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calcium might be because calcium absorption became low when serum 25-(OH)(D) concentrations was below 20 nmol/ L. At these concentrations serum alkaline phosphatase excretion increased.⁽²¹⁾

Conclusions:

The current study suggests an association between 25- OH vitamin (D) concentration and ADHD in childhood. The low levels of serum vitamin (D) among the ADHD children suggest the need for regularly monitoring of serum vitamin (D) levels and treatment of patients with vitamin (D) deficiencies. In addition, lifestyle and diet should be modified and directed towards eliminating the nutritional deficiencies in the society.

Limitations of the study:

Although important findings presented in the current study, the sample size was not big enough to warrant possibility of generalization of findings.

Recommendations:

1. More studies should be done to include large samples of patients.
2. Relationship between serum vitamin (D) level and ADHD warrants further investigation to define the exact role of vitamin (D) in the pathogenesis of ADHD.
3. Further studies with wider scope on large number of patients entailing vitamin (D) administration as an adjuvant treatment are needed to clarify the therapeutic effects of vitamin (D) supplementation.

Conflict Of Interest:

There is no conflict of interest.

Acknowledgment:

I would also like to express my heartily appreciation and thankfulness to all my colleagues for their support and cooperation. I wish to express my deep thanks and gratitude to my supervisors (the co- authors) for their constructive criticism, scientific instructions, and discussion throughout this work. Finally, I owe a special dept of gratitude to my patients and their families for their help, and positive participation in our study.

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Table (3) Laboratory results of the two study groups

Laboratory	Groups		P- Value
	Patients (N= 30)	Control (N= 30)	
Vitamin D3	14.42± 9.39	26.58± 16.13	0.001**
Calcium	8.72± 0.52	9.46± 0.40	0.000**
Phosphorous	3.18± 0.80	3.62± 0.79	0.035*
Alkaline Phosphatase	166.50± 69.56	104.77± 77.35	0.002**

*P- value is significant at 0.05 level, **P- value is highly significant at 0.01 level.

Normal values: The normal level of 25- OH- vitamin (D): sufficient> 30 ng/ml, insufficient 21- 29 ng/ml, deficient< 20 ng/ml, intoxication> 150 ng/ ml. Serum calcium 8.5- 10.5. Serum phosphorous 3- 4.5 mg/dl. Serum alkaline phosphatase 44- 147 IU/l.

There was a highly significant difference between serum vitamin (D) level in cases and controls with P value 0.001 as vitamin (D) was deficient in cases (mean 14.415) and sufficient in controls (mean 26.5833). Although serum calcium level was within normal for both groups, significant difference was present as lower calcium level in cases (mean 8.720) than controls (mean 9.457) with a P value 0.000. Significant difference reported between serum Phosphorus level in cases and controls with a P value 0.035. Serum Alkaline phosphatase was significantly higher in cases (mean 166.50) than controls (mean 104.77) with a P value 0.002.

Discussion:

The role of environment and more specifically the role of nutrition in the prevention and treatment of the symptoms of the disease have been attracting the attention of Russell et.al.⁽²³⁾ Diet therapy is a simple and inexpensive method that can be readily accepted by the parents and adopted by the children. The role of micronutrients such as iron, zinc and omega-3 on the prevention and control of the symptoms have been extensively studied. However, the role of vitamin (D) has attracted less attention. This condition exists despite the fact that vitamin (D) deficiency is associated with psychiatric diseases such as autism, schizophrenia, and depression. The vitamin is not only involved in bone metabolism and serum calcium regulation but also has significant effect on many body organs.

In the early stages of life, vitamin (D) deficiency might harm neuronal development and function. Vitamin (D) regulates the synthesis of neurotrophic factors such as neurotrophin NT 3, NT 4, nerve growth factor and glial cell line- derived neurotrophic factor, which are important for cell differentiation and survival.⁽¹¹⁾

Although the vitamin (D) receptor and activating enzymes are prominent in the hypothalamus and substantia nigra, the precise function of vitamin (D) in the brain is not fully understood. There are associations between low serum vitamin (D) and depression, mood disorders, premenstrual mood changes, decreased cognitive function and autism spectrum disorder.⁽¹⁴⁾

The association between vitamin (D) deficiency and ADHD in young children has not reported enough in the literature. Therefore, the aim of this study is to investigate the level of 25OHD in ADHD.

In the present study, age of participating children ranged between (5-

10) years (60- 120 months) old with a mean age for cases 90.27± 19.424. Out of 30 cases 21 (70%) were males and 9 patients (30%) were females. As regard controls the mean age group was 89.1± 19.945. Out of 30 controls 19 (63.3%) were male and 11 controls (36.6%) were females. All cases and controls were within the normal IQ range. The two groups were matched as regard age, sex and IQ with no statistically significant difference. Beshri et.al.⁽⁶⁾ found that male children have 2.5 and 5.6 times more chance than female children to be diagnosed as ADHD.

The current study found a highly significant difference between serum vitamin (D) level in cases and controls. Vitamin (D) was insufficient in cases and sufficient in controls. In a study conducted in Turkey among 7 to 18 years old children where a significant difference (P<0.05) in mean serum vitamin (D) level between cases (20.9± 19.4 ng/ml) and control groups (34.9± 15.4 ng/ml) was demonstrated.⁽¹⁵⁾

Another study on 1331 cases of ADHD and the same number of control groups healthy individuals under the age of 18 found out that the mean range serum vitamin (D) level of ADHD children (16.6± 7.8 ng/ml) was lower than in the control group (23.5± 9.9 ng/ml). In addition, 8.15% of the ADHDs had normal vitamin levels in their serum.⁽¹⁷⁾

Morales et.al.⁽²⁰⁾ examined whether maternal vitamin (D) status in pregnancy is associated with risk of ADHD- like symptoms in offspring. They concluded that higher maternal circulating levels of 25-(OH)D3 in pregnancy are associated with lower risk of developing ADHD- like symptoms in childhood.

Shang- Guan and Zhao⁽²⁴⁾ examined serum 25 hydroxyvitamin (D) levels in children with attention deficit hyperactivity disorder (ADHD) and to explore the relationship between vitamin (D) level and ADHD and concluded that serum levels of 25 hydroxyvitamin (D) in children with ADHD are lower than in healthy children, suggesting vitamin (D) level might be related to ADHD.

Sharif et.al.⁽²⁵⁾ stated that there was a statistically significant difference in serum vitamin (D) levels between the cases and the control groups. The serum vitamin (D) level of 21.6% children in the cases (ADHD) group was normal. The remaining 78.4% children had serum vitamin (D) level below normal. In the control group, 48.6% of the children had normal serum vitamin (D) level. None of the children in both groups showed toxic level of vitamin (D). Analysis has shown a statistically significant difference (P= 0.04) in serum vitamin (D) levels between the ADHD and the control groups.

The incidence of ADHD is much lower in areas with sunny weather and sunlight can have a protective effect against the disease. Phototherapy and sunray have been used as a treatment. A hypothesis in this regard is that sunrays increase the level of vitamin (D) level.⁽⁴⁾

Although serum calcium and phosphorous level were within normal for both groups in our study, highly significant difference was present as lower calcium level in cases than controls, significant difference as lower phosphorous in cases than controls. Serum Alkaline phosphatase was highly significant higher in cases than controls. The lower level of serum

the last 6 months before the study.

- b. Children with congenital syndromes, neurological or metabolic disorders.
- c. Subjects excluded due to the either refusal of the mother or difficulty to obtain venous blood from very uncooperative subjects.

Methods:

The children who fulfilled the criteria mentioned above will be subjected to the following:

1. A semi- structured interview to their parents or caregivers empathizing the demographic data as age, sex, residency, type of school, school grades, the history of the illness, family history including consanguinity between parents, presence of similar condition in family and other psychiatric disorders in the family. The condition of pregnancy, labor, type of labor, feeding, vaccination, milestones, scholastic achievement, relations, detentions, social and communication skills. Past history of medical or surgical condition or trauma, neuropsychiatric symptoms and signs noted by the caregiver.
2. Full Detailed Dietetic History.
3. Full detailed developmental history especially teething and motor development.
4. Full examination specially to exclude any chronic illness or vitamin (D) deficiency manifestation such as:
 - a. Bone deformity: genu valgum, genu varum, frontal posing.
 - b. Growth retardation and developmental delay.
 - c. Muscle Weakness.
5. Full psychiatric clinical assessment to identify psychiatric symptoms/ disorders as aggression, conduct disorder, anxiety, mood symptoms and disorders, phobia, delayed speech and tics.
6. ADHD diagnosis was based on using Diagnostic and Statistical Manual of Mental Disorders, Fifth Edition (DSM5) (American Psychiatric Association, 2013).⁽³⁾
7. Arabic versions of the Conner's rating scales revised, long versions, parent form.⁽¹⁰⁾
8. Conner's Parent Rating Scale- revised (CPRS- L); Long version (CPRS- R- L)⁽⁷⁾

Aim:

It is applied to detect subtypes and severity of ADHD, and assess scores in co- morbid cases of ADHD and bipolar disorders to compare to cases with ADHD only. Can be used between (3- 17) years.

Items are scored on 14 subscales of symptoms namely: oppositional, cognitive problems/ inattention, hyperactivity, anxious shy, perfectionism, social problems, psychosomatic, ADHD index (screening test for inattention), Conners' global index (CGI): restless- impulsive, CGI emotional liability, CGI total index: (screening test for hyperactivity), DSM- IV: inattentive, DSM- IV hyperactive- impulsive and DSM- IV total.

CPRS is an 80item questionnaire with an average administration time of 25- 30 minutes. It scores the parents' report of their child's behavior

during the past month on a 4- point- response scoring, namely (0: Not true or seldom true, 1: Just a little true, occasionally, 2: Often true, quite a bit, and 3: Very often true, very much true).

Assessment of intelligence quotient (IQ) using the Arabic version of the Stanford- Binet Intelligence Scale, 5th Edition (SB5).⁽¹²⁾

Assessment of intelligence quotient (IQ) using the Arabic version of the Stanford- Binet Intelligence Scale, 5th Edition (SB5).⁽²²⁾

The Full- Scale IQ (FSIQ) is derived from the sum of all the tasks in the SB5. It covers both the Verbal and Nonverbal domains of cognitive ability in a balanced design and taps the five underlying factor index scales of the SB5. The FSIQ provides a global summary of the examinee's current general level of intellectual functioning as measured by the SB5. The FSIQ is considered a reliable measure of g, or the general ability to reason, solve problems, and adapt to the cognitive demands of the environment. The test includes Fluid Reasoning and knowledge and Quantitative Reasoning and Visual Spatial Reasoning.

Measurement of 25 hydroxyvitamin (D) using commercial ELISA kits:

1. Venous blood samples (5 ml) were collected under sterile conditions from each patient and control subjects to measure serum 25-hydroxyl vitamin (D) level by enzyme linked immunosorbent assay (ELISA) technique. The 25 OH vitamin (D) ELA kit is used for determination of 25 OH vit (D) in human serum and plasma. It was manufactured at immune diagnostic company, Australian patent, kits' number.⁽²⁷⁾
2. Estimation of serum calcium, phosphorous and alkaline phosphatase using commercial kits.

Results:

Table (1) Demographic data distribution of the two study groups

	Groups				P- Value	
	Patients		Control			
	N (30)	%	N (30)	%		
Age Months Mean± S.D.	90.27± 19.424		89.1± 19.945		0.842	
Sex	Male (Total 40)	21	70%	19	63.3%	0.584
	Female (Total 20)	9	30%	11	36.7%	
IQ	94.13± 9.73		95.62± 9.81		0.671	

Age of participating children ranged between (5- 10) years (60- 120 months) old with a mean age for cases 90.27± 19.424 months, 21 patients out of 30 (70%) were males and 9 patients 30% were females. As regard controls the mean age group was 89.1± 19.945 months, 19 controls out of 30 (63.3%) were male and 11 controls 36.6% were females. All cases and controls were within the normal IQ range. The two groups were matched as regard age, sex and IQ with no statistically significant difference.

Table (2) Anthropometric measures of the two study groups

	Groups		P- Value
	Patients N (30)	Control N (30)	
Weight (Kg) (Mean± S.D.)	24.87± 5.53	24.52± 4.73	0.793
Height (Cm) (Mean± S.D.)	123.53± 10.65	123.68± 9.69	0.955
BMI (Mean± S.D.)	16.07± 1.23	15.83± 0.77	0.370

Both cases and controls were within normal as regard anthropometric measures and matched with no statistically significant difference.

Introduction:

Attention deficit/ hyperactivity disorder (ADHD) is a childhood-onset neurodevelopmental condition characterized by inattention, hyperactivity, and/ or impulsivity that can persist into adulthood with deleterious effects on social, academic, and behavioral outcomes.⁽¹⁹⁾ ADHD has prevalence of (3- 7)% among scholar population and it is associated with learning disabilities and executive dysfunctions.⁽¹⁾

Both genetic and neuroimaging studies in the past (20- 30) years have supported on early speculation of the dopamine activity in the presentation of ADHD. Genetic factors associated with dopamine receptor regulation are consistently associated with ADHD and neuroimaging studies have shown that brain structure and function in dopamine- relevant regions are altered in patients with ADHD.⁽¹⁸⁾

Although medications for ADHD have a large effect size and psychosocial interventions can augment treatment success, more than 30% of children are still symptomatic despite combined treatment. This support the need to use complementary medicine and nutritional supplementation. There is no conclusive data to support nutrient deficiencies as a cause of ADHD. However, patients with ADHD have reduced levels of vitamin (D), zinc, ferritin, and magnesium.⁽²⁶⁾

Vitamin (D) is a fat- soluble steroid, either synthesized endogenously or taken from diet, converted in the liver to 25 hydroxyvitamin (D) (25 OHD), and this metabolite is exposed to a second hydroxylation in the kidneys to produce the active form (1,25 OHD). Although 1,25 dihydroxyvitamin (D) is the active form of vitamin (D), serum levels are not considered a useful measure of vitamin (D) status in the body because its half- life is short and it may remain normal even in deficiency secondary to up- regulation of the 1 α -OHase enzyme. 25 hydroxyvitamin (D) (25-OH) is agreed to be the best measure of Vitamin (D) and reflects both cutaneous synthesis and intake from food and supplements. The Recommended Dietary Allowance for individuals ages (1- 70) is 600 IU/ day.⁽¹⁶⁾

In addition to its regulation of calcium and phosphorous in the intestine and stimulation of bone cell mineralization, vitamin (D) is a neuroactive steroid that has been shown in both animal and human studies to be crucial for normal brain development. Vitamin (D) receptors and enzyme es are located in neuronal cells of the substantia nigra, hippocampus, hypothalamus, prefrontal cortex, and cingulate gyrus; many of these regions have also been shown to have abnormalities in ADHD.⁽⁹⁾

Vitamin (D) deficiency during development has deleterious effects on the dopamine system. In animal models, vitamin (D) has been shown to be associated with the production of tyrosine hydroxylase, the rate-limiting enzyme for dopamine synthesis. Experimentally low doses of 1 alpha, 1- 25 dihydroxyvitamin D3 (1,25-(OH) 2D3) (the hormonally active form of vitamin (D)) are able to protect the mesencephalic dopaminergic neurons against toxins that cause a decrease in glutathione content.⁽¹¹⁾

The United States food and drug administration (FDA) defines a biomarker as an objective measure of normal processes, pathological

processes or pharmacological response. For psychiatry, biomarkers could be used to screen, diagnose, or predict the development of psychiatric disorders. Because no single biomarker candidate may be sufficient for accurate and reliable diagnosis, the current trend in psychiatry has shifted towards identifying sets of biomarkers.⁽¹³⁾

Most likely, no single ADHD biomarker can be identified. However, the use of a combination of markers may help to reduce heterogeneity and to identify homogeneous subtypes of ADHD. Those biomarkers may include Dopaminergic Biomarkers (e.g. Dopamine D4 Receptor (DRD4)), Noradrenergic Biomarkers (e.g. Neuropeptide Y (NPY)), Adrenergic Biomarkers for ADHD (e.g. Alpha- 2A- adrenergic receptor (ADRA2A)), Metabolism Enzymes as Biomarkers (e.g. Dopamine Beta Hydroxylase (DBH)), CNS Development Biomarkers (e.g. Brain Derived Neurotrophic Factor (BDNF)), Potential Biomarkers for environmental Risk factors (e.g. Iron, Zinc, Oxidative Stress), endophenotypes biomarkers (e.g. working memory, selective attention vigilance/ sustained attention, Theta/beta ratio and Reaction Time Variability).⁽¹³⁾

Some researches work to find the relation between ADHD and vitamin (D) and their results found that there is significance deficiency in serum vitamin (D) among ADHD children.⁽⁵⁾ Abdelsattar et.al.⁽²⁾ evaluated vitamin (D) in children with ADHD and they found that vitamin (D) is deficient among Egyptian children with ADHD.

As there is data suggests that vitamin (D) deficiency during development has deleterious effects on the dopamine system⁽¹¹⁾ and brain dopamine relative regions affected in ADHD.⁽¹⁸⁾ Vitamin (D) is important in cerebral function and it might have a role in the etiopathogenesis of ADHD.⁽⁸⁾ Our study aimed to investigate the level of 25OHD in children with ADHD aged 5- 10 years old.

Material and Methods

Subjects:

Subjects were recruited from the Psychiatric Clinic in the Collage of Postgraduate Childhood Studies. The study included 30 children diagnosed as ADHD according to the diagnostic and statistical manual of psychiatric disease (DSM)- 5 criteria.⁽³⁾ Another 30 subjects with matched age, sex- and social background and no chronic illness were taken as controls.

All parents agreed to undergo the assessment and had informal consent. The study was approved by ethical committee of faculty of postgraduate childhood studies (FPGCS), Ain Shams University.

1. Inclusion criteria:

- Age ranges between 5 and 10 years.
- Children fulfilled DSM 5 criteria for ADHD.
- Both Sexes Were Included.
- Not Taking Vitamin Supplementation.
- Written informed legal consent was signed by parents or caregivers of the children enrolling in the study.

2. Exclusion criteria:

- Patients with calcium supplements or vitamin (D) intake during

Study of Serum Vitamin D Level in a Sample of Egyptian Children with ADHD

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Summary

Background: Attention deficit/ hyperactivity disorder (ADHD) is a childhood- onset neurodevelopmental condition characterized by inattention, hyperactivity, and/ or impulsivity that can persist into adulthood with deleterious effects on social, academic, and behavioral outcomes. Vitamin (D) is a fat- soluble steroid, either synthesized endogenously or taken from diet, converted in the liver to 25 hydroxyvitamin (D) (25 OHD), and then second hydroxylation occur in the kidneys to produce the active form (1, 25 OHD). Vitamin (D) receptors and enzymes are located in neuronal cells of many brain regions which shown to have abnormalities in ADHD.

Materials: This study was done at the Psychiatric Clinic in the Faculty of Postgraduate Childhood Studies, Ain Shams University from April 2018 to August 2019. 123 children assessed to reach the thirty children diagnosed with ADHD who were conducted in this study. Another 30 age, sex- and social background- matched normal children with no chronic illness were taken as controls. Their ages ranged from (5- 10) years. ADHD diagnosis was based on using Diagnostic and Statistical Manual of Mental Disorders, Fifth Edition (DSM5). Conner's Parent Rating Scale Revised was applied. The serum level of human 25 hydroxyvitamin (D) was estimated by ELISA technique. Also, serum calcium, phosphorous and alkaline phosphatase were measured.

Results: There was a highly significant difference between serum vitamin (D) level. It was deficient in cases (14.42 ± 9.39) and sufficient in controls (26.58 ± 16.13). Serum calcium and phosphorous level were within normal for both groups with highly significantly lower calcium level (8.72 ± 0.52) and significantly lower phosphorous level (3.18 ± 0.80) in cases compared to controls (9.46 ± 0.40) and (3.62 ± 0.79) respectively. Serum Alkaline phosphatase was highly significantly higher in cases (166.50 ± 69.56) than controls (104.77 ± 77.35).

Conclusion: 25 OHD is deficient in children with ADHD.

Keywords: Attention deficit hyperactivity disorder, vitamin (D), Calcium, Phosphorous, Alkaline Phosphatase.

دراسة فيتامين (د) بمصل الدم في اضطراب فرط الحركة ونقص الانتباه في عينة من الأطفال المصريين

المقدمة: اضطراب فرط الحركة ونقص الانتباه هو حالة تخص التطور العصبي وتبدأ في مرحلة الطفولة وتتسم بعدم التركيز، وفرط النشاط، و/أو الاندفاع التي يمكن أن تستمر في مرحلة البلوغ مع آثار ضارة على المخرج الأكاديمي والاجتماعي والسلوكي. فيتامين (د) هو ستيرويد يذوب في الدهون، وسواء أكان مصنعا داخل الجسم أو مأخوذا من الطعام، فإنه يتحول داخل الكبد إلى 25-هيدروكسي فيتامين (د)، ثم يتعرض هذا المستقلب لعملية هيدروكسيلية أخرى في الكلى ليتحول إلى الصورة النشطة 1-25 ثنائي هيدروكسي فيتامين (د). إن مستقبلات فيتامين (د) وإيزيماتته توجد في الخلايا العصبية في عدة مناطق في المخ؛ العديد من هذه المناطق تعاني من اختلالات في اضطراب فرط الحركة وتشتت الانتباه.

طريقة البحث: تمت هذه الدراسة في عيادة الطب النفسي بكلية الدراسات العليا للطفولة بجامعة عين شمس في الفترة من أبريل 2018 إلى أغسطس 2019. تم فحص 123 طفل للوصول إلى عدد 30 طفل مصابين باضطراب فرط الحركة ونقص الانتباه والذين تم ادراجهم في الدراسة. بالإضافة إلى عدد 30 طفل متماثلين مع المجموعة الأولى من حيث العمر والجنس والبيئة الاجتماعية ولا يعانون من أمراض مزمنة كمجموعة ضابطة. أعمار الأطفال تتراوح من خمس إلى عشر سنوات وتم تشخيص اضطراب فرط الحركة وتشتت الانتباه بواسطة DSM-5 (تم تطبيق اختبار كونر المعدل للوالدين تم قياس 25 هيدروكسي فيتامين (د) في مصل الدم بواسطة الاليزا كما تم قياس نسبة الكالسيوم والفوسفور والفوسفاتيز القلوية في مصل الدم).

النتائج: هناك فروق إحصائية ذات دلالة عالية فيما يتعلق بمستوى فيتامين (د) في المصل حيث أنه ناقص في مجموعة الحالات (14.42 ± 9.39) وكافي في المجموعة الضابطة (26.58 ± 16.13). مستوى الكالسيوم والفوسفور في حدود الطبيعي في المجموعتين مع وجود فرق إحصائي ذا دلالة عالية وانخفاض معدل الكالسيوم (8.72 ± 0.52) في الحالات وفرق إحصائي ذا دلالة وانخفاض معدل الفوسفور (3.18 ± 0.80) حيث أن كليهما أقل في مجموعة الحالات من المجموعة الضابطة (9.46 ± 0.40) و (3.62 ± 0.79) على التوالي. فيما يخص الفوسفاتيز القلوية فهناك فرق إحصائي ذو دلالة عالية حيث وجد أن معدله اعلى في مجموعة الحالات (166.50 ± 69.56) من المجموعة الضابطة (104.77 ± 77.35).

الاستنتاج: فيتامين (د) ناقص في الأطفال المصابين باضطراب فرط الحركة ونقص الانتباه.

الكلمات الدالة: اضطراب فرط الحركة ونقص الانتباه، فيتامين (د)، كالسيوم، فوسفور، فوسفاتيز قلوية.