

Measuring the Level of Heavy Metals in Hair Samples of Autistic Children in Baghdad

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Abstract

Autism is a severe developmental condition characterized by repetitive or stereotypic behavior, neurodevelopmental abnormalities, and communication issues. Genetic and environmental variables, as well as their interactions, have been recognized as contributing factors to autism, even though the pathophysiological reasons for the disorder are still largely unknown and under dispute. Although it is well known that a variety of variables can lead to autism, environmental factors have garnered a lot of attention lately. A significant portion of the global discourse has been on neurotoxins, including lead, mercury, cadmium, and iron. Some contend that these and other hazardous metals play a part in the onset of the illness. The research is being conducted in Baghdad, Iraq. In this study, 66 children with verified DSM-V diagnoses of autism and 39 children from volunteers acted as controls. To examine hair samples, X-ray fluorescence (XRF) is utilized. The findings indicate that lead, cadmium, mercury, and iron levels are not substantial. The mean values of lead $0.005 \pm 0.002 \mu\text{g/g}$ concentrations were in $0.0014 \pm 0.0007 \mu\text{g/g}$, mercury $0.00039 \pm 0.00012 \mu\text{g/g}$ and iron $0.194 \pm 0.063 \mu\text{g/g}$ respectively for children with autism, while for the control the mean values were $0.001 \pm 0.000 \mu\text{g/g}$, $0.0003 \pm 0.0001 \mu\text{g/g}$, $0.00031 \pm 0.00017 \mu\text{g/g}$, $0.130 \pm 0.077 \mu\text{g/g}$. The elevated amounts in autistic children as compared to the control group and the international normal value may be attributed to environmental pollution in Iraq.

Keywords: Autism, Hair, heavy metals, Iron, Lead, Cadmium, X-ray fluorescence

قياس مسنوى المعادن الثقيلة في عينات شعر الاطفال النوحديين في بغداد

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المستخلص

التوحد هو حالة تنموية حادة تتميز بسلوك غير نمطي، وتشوهات في النمو العصبي، ومشاكل في التواصل. تم التعرف على المتغيرات الجينية والبيئية، وكذلك تفاعلاتها، كعوامل مساهمة في مرض التوحد، على الرغم من أن الأسباب الفيزيولوجية المرضية لهذا الاضطراب لا تزال غير معروفة إلى حد كبير وموضع خلاف. على الرغم من أنه من المعروف أن مجموعة متنوعة من المتغيرات يمكن أن تؤدي إلى مرض التوحد، إلا أن العوامل البيئية حظيت بالكثير من الاهتمام في الآونة الأخيرة. وكان جزء كبير من الخطاب العالمي يدور حول السموم العصبية، بما في ذلك الرصاص، والزنبق، والكاديوم والحديد. ويرى البعض أن هذه المعادن الخطرة وغيرها تؤدي دوراً في بداية المرض. ويجري البحث في بغداد، العراق. في هذه الدراسة، كان ٦٦ طفلاً تم التحقق من تشخيصهم بالتوحد بواسطة الدليل التشخيصي والإحصائي للاضطرابات العقلية (DSM-V) و ٣٩ طفلاً من المتطوعين بمثابة عناصر تحكم. لفحص عينات الشعر، يتم استخدام الأشعة السينية (XRF) تشير النتائج إلى أن مستويات الرصاص والكاديوم والزنبق والحديد ليست كبيرة. كانت القيم المتوسطة لتركيزات الرصاص 0.002 ± 0.005 مايكروغرام / غرام هي 0.0014 ± 0.0007 مايكروغرام / غرام والزنبق 0.00012 ± 0.00039 مايكروغرام / غرام والحديد 0.00194 ± 0.0063 مايكروغرام / غرام على التوالي للأطفال المصابين بالتوحد، بينما كانت القيم المتوسطة للتحكم 0.0001 ± 0.0000 مايكروغرام / غرام 0.0003 ± 0.0001 مايكروغرام / غرام، 0.00031 ± 0.00017 مايكروغرام / غرام، 0.130 ± 0.077 مايكروغرام / غرام. إن ارتفاع الكميات لدى الأطفال التوحديين مقارنة بالمجموعة الضابطة والقيمة الطبيعية الدولية يمكن أن يعزى إلى التلوث البيئي في العراق

الكلمات المفتاحية: التوحد، الشعر، المعادن الثقيلة، الحديد، الرصاص، الكاديوم، مضان الأشعة
السينية

Chapter one Introduction

Autism spectrum disorder (ASD) is a neurodevelopmental disorder that may be brought on by several environmental and genetic causes (Boris *et al.*,2004: Sadek *et al.*, 2017: Guo *et al.*,2012). Children with autism also face challenges in speaking, interacting with others, and engaging in limited, repetitive, and stereotypical activities and hobbies (Santos *et al.*,2010). Men's appearance is three to four times that of women's, and it happens within the first three years of life (Sener *et al.*,2014). The researchers linked ASD to the interaction of genetic and environmental factors, even though the exact etiology of the disorder is unknown. Heavy metals harm human health, and exposure to these metals has been increased by industrial and anthropogenic activities and modern industrialization. Contamination of water and air by toxic metals is an environmental concern, and hundreds of millions of people worldwide are affected. Food contamination with heavy metals is another concern for human health (AL-Amier, 2018). The concentration of heavy metals in water resources, air, and food is assessed in this regard heavy metals are released in the form of air particulates in the urban atmosphere as liquid or solid particles (Abdallah and Abdulhay, 2017). Due to their potential to induce birth defects, fetal developmental problems, neurological diseases, developmental delays, learning challenges, and behavioral abnormalities- all of which may lead to ASD- heavy metals are regarded as reproductive and developmental toxins (Blaurock-Buschand and RABAH,2011).

Heavy metal levels in hair may be measured concerning autistic disorders and can be used to diagnose autism (Al-Ayadhi ,2005). This procedure can reveal prolonged exposure to the metal. There is also proof that certain metallic elements are essential to maintaining human health. Numerous health conditions, both physical and mental, may be exacerbated by a trace element deficiency. Excessive amounts of hazardous metals or insufficient essential elements may cause an imbalance (Barlow *et al.*,1986). Many things, such as industrial paint, dust, fish, vegetables, construction materials, chemical goods, vehicle fumes, fertilizers, and the environment itself, can lead to the buildup of heavy metals (Eqani *et al.*,2020: Mohammadabadi *et al.*,2020: Liang *et al.*,2017). Children with autism may be genetically predisposed to brain damage linked to autistic behavioral traits due to a decreased ability to metabolize hazardous environmental substances (Geier *et al.*, 2009). The purpose of this study is to measure the concentration of some heavy metals, including lead, cadmium, mercury, and iron in hair samples from children with autism and compare the findings to those from matched, healthy, normal children. It can also be a way to keep an eye on the severity of ASD and a way to support kids who have the disorder in managing their condition.

Chapter two Material and methods

Patients

In this study autism syndrome diagnosis (ASD) groups were composed of patients who attended Medical City Hospital for Mental and Psychological Diseases in Baghdad, a specialized center for the treatment of autism, and a private clinic, in addition to patients who were reached through advertising by social media or from acquaintances and friends.

The study included 66 patients who live in Baghdad /Iraq, they were diagnosed by a psychiatric and neurological consultant. Their ages ranged between 3- 15 years and the duration of the disease was different. All patients underwent a cognitive assessment with psychological tests in the psychological counseling department of the hospital.

Psychologists use a variety of information sources to diagnose ASD, including patient interviews, behavioral observations, cognitive and language tests, medical exams to rule out other conditions, and interviews with parents or other adults who can provide insight into the patient's social, emotional, and behavioral development.

Controls

The control group consisted of 39 healthy participants; their ages ranged between 3-15 years. The selected children had no previous family history of ASD or any mental or neurological diseases.

1.Measurement of heavy metals in hair samples

The measurements were done at the University of Baghdad's German Laboratory, Department of Geology, College of Science. It was essential to use a mortar to turn the hair samples into powder before subjecting the specimen to X-rays. Following preparation, specimens and hair were put in a sample container and subjected to fluorescence spectroscopy. The detector sent its output pulses to a multi-channel analyzer that was attached to a computer so that they could be processed. Each analog pulse is converted by a multichannel analyzer into digital data for processing, after which it is stored in memory and the processed data is shown by counting against the channel number. In this work, the amount of these components in healthy and patient hair samples was used to assess the dependency of spectrum alterations. spectrum patterns from fluorescence spectroscopy were compared between healthy and patient hair samples. Based on the fluorescence X-ray signals that a material emits, X-ray fluorescence (XRF) is a non-destructive elemental analytical technique that may be used to identify the elements present in a material and calculate their percentages.

2.Ethical Clearance

All participating children's parents or legal guardians gave their consent. Before any data was collected, the parents or guardians were given a thorough description of the study, its goals, and its advantages, and their agreement was acquired. Furthermore, the youngsters gave their full consent to participate in the study and were not exposed to any form of force or improper influence. The ability to remove their children from their studies at any time without facing any repercussions was granted to parents or guardians.

Chapter three Results and discussion

1. The concentration of some heavy metal in the hair

Long-term exposure to metals can be identified and utilized as a diagnostic test for autism by analyzing levels of heavy metals in the hair of autistic individuals. Furthermore, it has been demonstrated that several metallic elements are essential to maintaining human health. A shortage of trace elements can be a factor in many different medical and psychiatric conditions. A harmful metal excess or a key element shortage might be the cause of the imbalance (Aljumaili *et al.*, 2023). The concentration of heavy metals in the hair of individuals with autism spectrum disorders and control groups is displayed in Table (1).

Table (1): Concentration of heavy metals in the hair of autism spectrum disorders patients and control group

Parameters ($\mu\text{g/g}$)	Control Mean \pm SE	Patients Mean \pm SE	P- Value	Significant P \leq 0.05
Lead	0.001 \pm 0.000	0.005 \pm 0.002	0.117	NS
Cadmium	0.0003 \pm 0.0001	0.0014 \pm 0.0007	0.2109	NS
Mercury	0.00031 \pm 0.00017	0.00039 \pm 0.00012	0.69771	NS
Iron	0.130 \pm 0.077	0.194 \pm 0.063	0.543	NS
NS \rightarrow	Non-Significant Difference at p \leq 0.05			

Measurement of the lead concentration in hair specimens for patients

The level of lead in the hair of children can vary depending on several factors, including their environment, diet, and lifestyle. The Centers for Disease Control and Prevention (2022), considers any hair lead level above 40 $\mu\text{g/g}$ to be high in children. Figure (1) shows a difference in the concentration of lead between autistic and control children. The mean value for the level of lead concentrations in ADS patients was $0.005 \pm 0.002 \mu\text{g/g}$, while it records a concentration of $0.001 \pm 0.000 \mu\text{g/g}$ in healthy control which the result was not significant ($P \leq 0.05$) table (1).

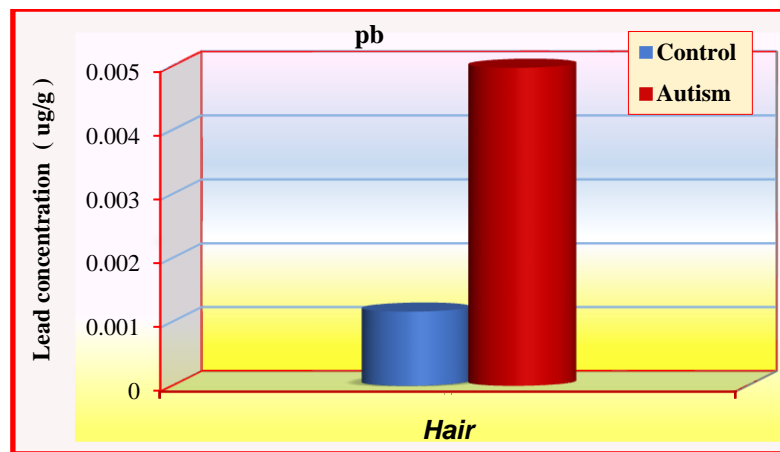


Figure (1): Comparison between the concentration of lead in the hair of autism spectrum disorders patients and control groups

Since children are more prone to put items in their mouths and so absorb more lead, they are particularly vulnerable to lead poisoning. Conversely, lead buildup in children is influenced by an underdeveloped immune system (Naranjo *et al.*, 2020). Given that children's brains and neurological systems may absorb four to five times more lead than those of adults, there is a strong correlation between excessive acute lead exposure and the likelihood of disease incidence (WHO, 2022). Because children's neural systems are more sensitive than adults, lead can have more severe effects on them than on adults. Many neurological system diseases, including headaches, nausea, tremors, and numbness, can be brought on by exposure to lead (Mason *et al.*, 2014). Mohamed *et al.* (2015) investigated the lead, mercury, and aluminum levels in Egyptian children diagnosed with autism. Compared to controls, the mean concentrations of these heavy metals in the hair of autistic individuals were much greater. Living close to petrol stations and maternal fish eating were favorably linked with their levels.

1.2. Measurement of the cadmium concentration in hair specimens for patients and control groups

Toxic metal cadmium can lead to major health issues, particularly in young children. Exposure to even low concentrations of cadmium can harm a child's developing brain and neurological system, resulting in behavioral issues, learning impairments,

and other health issues (NIEHS,2023). Figure (2) shows a difference in concentration of cadmium between autistic and control children, the mean value for the level of cadmium concentration in ADS patients was $0.0014 \pm 0.0007 \mu\text{g/g}$, while it records a concentration of $0.0003 \pm 0.0001 \mu\text{g/g}$ in healthy control which was also not significant ($P \leq 0.05$). The Agency for Toxic Substances and Disease Registry considers any hair cadmium level above $100 \mu\text{g/g}$ to be high in children (ATSDR,2022).

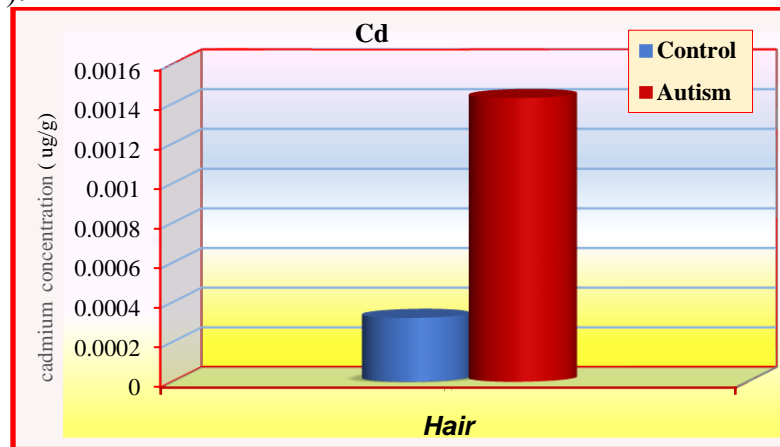


Figure (2): Comparison between the concentration of cadmium in the hair of autism spectrum disorders patients and control groups

The current results are consistent with the study of Aljumaili *et al.* (2023) investigated the levels of lead, iron, and cadmium in autistic children's hair in Al-Anbar province, Iraq. The study examined hair as a long-term exposure indicator. The study revealed significantly higher levels of both lead and iron in autistic children compared to controls and international reference values. Meanwhile, cadmium level was slightly higher in children with autism than in controls and within the international normal values. These findings suggest that lead and iron exposure may be environmental risk factors for ASD in this province. The results indicated the possibility that environmental pollution in Al-Anbar province may contribute to the high levels of lead and iron in children with autism.

1.3. Measurement of the mercury concentration in hair specimens for patients

Toxic metal mercury can lead to major health issues, particularly in young children. Any hair mercury level in children over $1.1 \mu\text{g/g}$ is deemed excessive by the Centers for Disease Control and Prevention (CDC). The levels of mercury in hair samples from children with autism and the control groups are displayed in Figure (٣). Compared to healthy controls, the mean mercury levels in ADS patients were $0.0014 \pm 0.0007 \mu\text{g/g}$, whereas it was $0.00031 \pm 0.00017 \mu\text{g/g}$. Table 1 shows that the results were not substantially different ($P < 0.05$).

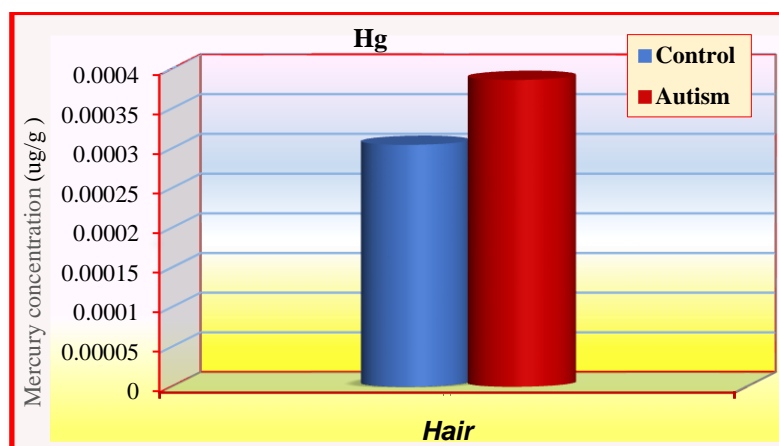


Figure (3): Comparison between the concentration of mercury in the hair of autism spectrum disorders patients and control groups

The concentration of mercury may be close between patients and controls. This may be due to constant exposure to environmental pollution due to burning waste, exhaust smoke, and the friction of car tires. The three DSM-IV autism diagnostic criteria—difficulties communicating, repetitive and stereotyped patterns of behavior—can all be brought on by mercury exposure. Mercury can also cause symptoms that are common in ASD, including movement disorders, emotional/psychological changes, anomalies in senses, difficulties with abstract or sophisticated thought, significant sleep disruptions, and self-harming behavior (CDC,2023: WHO,2023). In the Mackenzie Valley of Canada, Packull-McCormick *et al.* (2022) evaluated the mercury levels in the First Nations settlements. The purpose of the study was to determine if hair mercury concentrations fluctuate year-round and if seasonal variations in fish diet are associated with hair mercury concentrations. Although preliminary findings indicated a potential connection between total fish intake and hair mercury levels, more thorough examination using statistical models.

1.4. Measurement of the iron concentration in hair specimens for patients and control groups

Iron levels in hair are not considered to be a reliable indicator of iron status in children. This is because iron levels in hair can be affected by many factors, including diet, environment, and health conditions.

Some studies have shown that iron levels in hair may be correlated with iron levels in the blood. However, other studies have shown no correlation between iron levels in hair and iron levels in the blood (Wozniak *et al.*, 2019). Figure (3) shows a difference in percentage between autistic and control children the mean value for the level of iron constrictions in ADS patients was $0.194 \pm 0.063 \mu\text{g/g}$ while it records a concentration of iron $0.130 \pm 0.077 \mu\text{g/g}$ in healthy control which the result not significant $P \leq 0.05$.

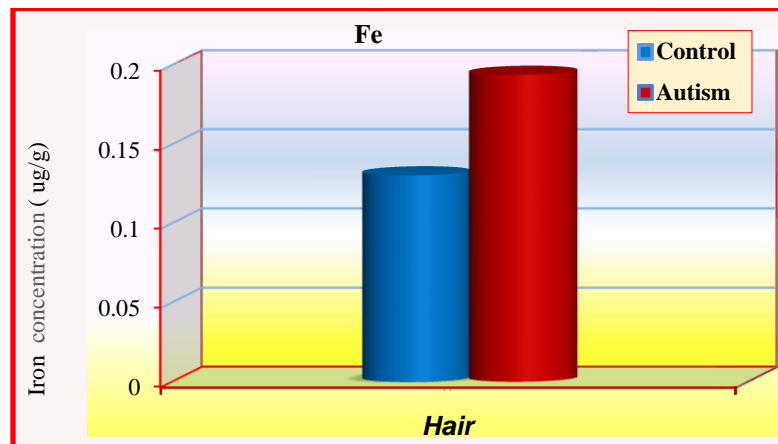


Figure (3): Comparison between the concentration of iron in the hair of autism spectrum disorders patients and control groups

Sahin *et al.* (2015) study examined the possible relationship between blood parameters and hair iron levels in individuals with different iron reserves. Those with iron deficiency anemia, transfusion-related anemia with varying iron contents, and a healthy control group were among the participants. Measurements were made of serum iron, hair iron content, and total iron-binding capacity (TIBC). The most important discovery was that patient groups with different body iron contents had considerably different hair iron levels. Furthermore, a correlation was seen between the measured blood indicators of iron status and these hair iron levels. This implies that the amount of iron in hair may serve as a helpful proxy for the body's total iron level.

Chapter four

Conclusion

The results show a correlation between Lead, cadmium mercury and Iron concentrations, and ASD in the hair of autistic Iraqi children, it was higher than normal rates due to continuous exposure to these substances, exposures during critical developmental periods may exacerbate symptoms. Heavy metals is risk factor in Iraq environmental.

Chapter five

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