Blood Levels of Heavy Metals among Mothers and their Autistic Children in South-South, Nigeria

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Abstract

Environmental pollution resulting from oil exploration is a significant public health concern, particularly for pregnant women and their offspring. This study investigated the levels of heavy metals (lead (Pb), mercury (Hg), and arsenic (As)) and their association with autism spectrum disorder (ASD) in South-South Nigeria, a region heavily impacted by oil exploration and petrochemical activities. Employing a descriptive correlational design, blood samples were collected from mothers and children with autism, as well as from healthy controls, across five government-recognized autism centres in Rivers and Bayelsa States. A total of 872 participants were included: 218 mothers of children with autism, 218 healthy mothers, 218 children with autism, and 218 healthy children from the same mothers. Heavy metal concentrations were analyzed using a Finnigan MAT95XP Mass Spectrometer, ensuring high precision in detecting heavy metal toxicants. Data were analyzed using descriptive statistics, chi-square tests, and t-tests at a significance level of 0.05. The results showed significantly higher concentrations of Pb and Hg in mothers and children with autism compared to controls ($p < 0.001$), while As levels were undetectable in all groups. These findings suggest a possible link between exposure to these heavy metals and autism, highlighting the urgent need for further research and targeted remediation efforts in high-exposure regions such as South-South Nigeria.

Keywords: Mother, Child, Autism, Environmental, Heavy metals

Introduction

Heavy metals such as lead (Pb), mercury (Hg), and arsenic (As) are naturally occurring environmental toxicants found in varying concentrations (Philip-Slaboh et al., 2023). However, anthropogenic activities such as mining, fossil fuel combustion, and industrial manufacturing have led to their widespread distribution, increasing environmental contamination (Skogheim et al., 2021). These heavy metals have been extensively studied

due to their neurotoxic effects, with growing evidence suggesting their role in neuronal mutations and neurodevelopmental disorders, including autism spectrum disorder (ASD) (Ijomone et al., 2020).

Human exposure to heavy metals occurs through multiple pathways, including contaminated drinking water, food sources, and air. Lead exposure is commonly linked to industrial waste polluting water sources, while mercury exposure primarily results from consuming contaminated seafood such as fish and shellfish (Papadopoulou et al., 2019). Given that riverside communities depend heavily on seafood and are susceptible to water pollution, residents in such areas may face a heightened risk of heavy metal exposure.

Pregnant women and infants are particularly vulnerable to these toxicants. During pregnancy, maternal exposure can lead to the accumulation of heavy metals in the bloodstream, allowing them to cross the placental barrier and accumulate in the amniotic fluid (Skogheim et al., 2021). Some heavy metals can penetrate the underdeveloped blood-brain barrier of the fetus, potentially leading to neurological impairments (Long et al., 2019). Furthermore, toxicants accumulated in maternal tissues can be transferred to infants through breastfeeding, increasing the risk of developmental disruptions (Mohamed et al., 2015).

Autism spectrum disorder (ASD) is a complex neurodevelopmental condition characterized by social communication deficits, cognitive delays, and repetitive behaviours (Liew et al., 2015). While ASD is diagnosed within the first three years of life and occurs across diverse populations, its global prevalence has risen dramatically from 0.6% in 2000 to approximately 4% in 2021 (Miani et al., 2021; Skogheim et al., 2021). Male children are disproportionately affected, with a male-to-female ratio of about 5:1 (Dickerson et al., 2017; Rynkiewicz et al., 2016).

The etiology of ASD remains complex, with both genetic and environmental factors contributing to its development. While genetic mutations account for approximately 37% of ASD cases, environmental factors, including exposure to heavy metals, have been implicated in nearly 55% (Long et al., 2019). Maternal exposure to toxic metals such as mercury, lead, and cadmium has been linked to adverse neurodevelopmental outcomes, interfering with normal brain development and potentially leading to ASD (Rahbar et al., 2021). Other risk factors include advanced maternal age (above 40 years), preterm birth, and maternal deficiencies in essential nutrients such as folic acid, vitamin D, and fatty acids (Lyall et al., 2013; Sullivan et al., 2014).

Despite global recognition of these risk factors, there is a need for localized research to understand the specific environmental exposures affecting different populations. Nurses and healthcare professionals play a crucial role in supporting children with ASD through behavioural interventions and communication therapies (Zwaigenbaum et al., 2015).

However, the rising prevalence of ASD underscores the need for preventative strategies targeting environmental risk factors.

In Africa, ASD prevalence is estimated at 1.5% (Zeidan et al., 2022), with reported rates in southern Nigeria ranging from 0.8% to 2.3%, exceeding the continental average (Ataben et al., 2024; Torty et al., 2024). The South-South region of Nigeria is a hub for oil exploration and petrochemical activities, raising concerns about heavy metal contamination and potential links to ASD. However, a search of major research databases revealed a paucity of studies examining this association in the region, highlighting a critical knowledge gap.

This study seeks to address this gap by investigating whether maternal exposure to heavy metals is associated with ASD in South-South Nigeria. The findings will provide valuable insights for public health policies, guide preventative interventions, and support the development of evidence-based nursing practices aimed at mitigating the burden of ASD in high-risk populations.

Methods

Ethical Clearance, Design and Area of the study

This study was approved by the University of Port Harcourt Institutional Review Board (IRB) and adhered to the Declaration of Helsinki guidelines (2013). A descriptive correlational design was adopted to examine the association between maternal heavy metal exposure and autism spectrum disorder (ASD) in children. The study was conducted across five government-recognized autism centers in Rivers and Bayelsa States, namely: University of Port Harcourt Teaching Hospital Autism Clinic, Alakahia, Port Harcourt; OLG Autism Centre in Woji, Port Harcourt; Choice Children's Hospital and Autism Centre in Abuloma Road, Port Harcourt; Blazing Heart Autism Centre in Mgbuoba, Port Harcourt; and Early Foundation Schools in Yenagoa.

Population, Sample Size Determination and Sampling

The study population consisted of 3,066 individuals, comprising 1,534 mothers, 766 autistic children, and 766 neurotypical children. A sample size of 872 (218 mothers of children with autism, 218 mothers of neurotypical children, 218 autistic children, and 218 neurotypical children) was determined using the sample size calculation formula for comparative studies as cited in Bolarinwa (2020): $n = [(Z_{\alpha/2} + Z_{\beta})^2 \times \sigma^2 (M_1 - M_2)^2] \div d^2$; where n = minimum sample size; $Z_{\alpha/2}$ = the critical value of the normal distribution at is 1.96 for a 95% confidence level; Z_{β} = the critical value of the normal distribution is 0.84 at a power of 80%; σ2 = the best guess pooled variance of the two samples is 1; M1 - M2 = the best guess mean difference between the two samples is 1; d = the Cohen's small effect size of interest is 0.2. A minimum sample size of 196 was computed. Then the minimum

sample size was increased by 10% using the non-response formula cited in Bolarinwa (2020) and mathmatically expressed as follows: nf = n ÷ (1-0.1); where nf = final sample size. The final sample size of 218 per group was computed.

Participants were selected using a simple random sampling technique. Inclusion criteria required that participants reside in Rivers or Bayelsa States and were not seriously ill, ensuring their ability to provide blood samples. Exclusion criteria included mothers who had received treatment for autism in the past and those who were currently pregnant.

Data Collection and Laboratory Analysis

Data collection involved the use of a semi-structured questionnaire and laboratory analysis of blood samples. The questionnaire comprised two sections: Section A contained structured (close-ended) questions on socio-demographic characteristics; Section B included open-ended questions assessing the concentration of heavy metals in blood samples.

Blood sample collection followed strict aseptic procedures. A trained phlebotomist, donning personal protective equipment (PPE) such as gloves, a laboratory coat, and face protection, collected 1 mL of venous blood from each participant using sterile 2 mL syringes fitted with 23G needles. The blood was stored in EDTA-coated specimen bottles to prevent coagulation. After collection, pressure was applied to prevent hemorrhage, and all used materials were disposed of in a biohazard waste container. Each blood specimen bottle was labeled with a unique identifier and placed in an insulated, temperature-controlled container (2–8°C) to preserve sample integrity during transport. Samples were delivered to the laboratory within 1.5 hours of collection.

In preparation for analysis, specimen tubes were washed with 5 mL of a 10% nitric acid solution and rinsed with 10 mL of distilled water. Each 1 mL blood sample was digested with 2.5 mL of 65% nitric acid using an ETHOS UP microwave digestion system (Milestone Inc., Shelton, USA) for 60 minutes at 90°C. The digest was then diluted to 10 mL with distilled water. Control samples were prepared using the same procedure with de-ionized water.

Heavy metal concentrations were measured using the Finnigan MAT MAT95XP Mass Spectrometer (Thermo Fisher Scientific, 1995). The spectrometer operates based on mass-to-charge ratio separation principles and has a high-performance resolution capable of detecting masses up to 9,000 Daltons. The device was calibrated using six reference points (0, 4, 6, 8, 15, and 25 ppm) on a linear scale. Each test batch was preceded by a blank analysis and method correction blank assessment. Every participant's sample was analyzed twice, and the mean value was recorded.

Data Analysis

Categorical data from Section A of the questionnaire were analyzed using descriptive statistics such as frequencies and percentages. Continuous data from Section B were summarized using measures of central tendency (mean) and dispersion (standard deviation). Hypothesis testing was conducted using Chi-square and t-tests, with statistical significance set at a 5% level.

Results

Table 1: Demographic characteristics of the mothers

Category	**Mothers of children with Autism (n = 218)**	**Mothers of healthy children (n = 218)**	**p value**
	f (%)	f (%)	
Age			<0.001
20-29	6 (2.8)	71 (32.6)	
30-39	98 (44.9)	79 (36.2)	
40-49	114 (52.3)	68 (31.2)	
Parity			0.846
1-3	89 (40.8)	91 (41.7)	
4-6	129 (59.2)	127 (58.3)	
Educational level			0.220
Primary	70 (32.1)	81 (37.2)	
Secondary	66 (30.3)	72 (33.0)	
Tertiary	82 (37.6)	65 (29.8)	
Occupation			0.521
Farmer	53 (24.3)	60 (27.5)	
Trader	45 (20.6)	53 (24.3)	
Office worker	65 (29.9)	54 (24.8)	
Unemployed	55 (25.2)	51 (23.4)	

Chi squared was used for analysis, p value < 0.05 is significant; f = frequency, % = percent

Table 1 showed that mothers of children with autism were significantly older than mothers of healthy children ($p < 0.001$), with a higher proportion aged 40-49 years. Parity, education, and occupation showed no significant differences between groups ($p > 0.05$). Most mothers had 4-6 children, and occupations were similarly distributed across both groups.

Table 2: Demographic characteristics of the children

Category	**Child with Autism (n = 218)**	**Healthy child (n = 218)**	**p value**
	f (%)	f (%)	
Age of child			0.328
12-23 months	87 (39.9)	72 (33.0)	
24-35 months	63 (28.9)	70 (32.1)	
36-47 months	68 (31.2)	76 (34.9)	
Gender			0.502
Male	109 (50.0)	102 (46.8)	
Female	109 (50.0)	116 (53.2)	
Maternal occupation			
Farmer	59 (27.0)	66 (30.3)	0.197
Trader	54 (24.8)	57 (26.2)	
Office worker	41 (18.8)	45 (20.6)	
Unemployed	64 (29.4)	50 (22.9)	

Chi squared was used for analysis, p value < 0.05 is significant; f = frequency, % = percent

Table 2 revealed that there were no significant differences in age, gender, or maternal occupation between children with autism and healthy children (p > 0.05). Age distribution was similar across both groups, with most children aged 12-23 months. Gender was evenly split in autistic children, while slightly more healthy children were female.

Table 3: Heavy metals (Pb, Hg, and As) concentration in the blood of mothers

Heavy metal concentration in blood	**Mothers of children with Autism (n = 218)**	**Mothers of healthy children (n = 218)**	**t-test**	**df**	**p value**
Lead (Pb)			33.10	434	<0.001
Mean (µg/L)	4.12	2.82			
SD	0.40	0.41			
Mercury (Hg)					
Mean (µg/L)	2.20	1.64	16.61	434	<0.001
SD	0.39	0.32			
Arsenic (As)					

Mean (µg/L)	<LOD	<LOD	0.00	0.00	1.00
SD	0.00	0.00			

<LOD = below the limit of detection, df = degree of freedom, $p < 0.05$ is significant, n = sample size, µg/L = microgram per litre

Table 3 demonstrated that Mothers of children with autism had significantly higher blood concentrations of lead (4.12 µg/L vs. 2.82 µg/L, $p < 0.001$) and mercury (2.20 µg/L vs. 1.64 µg/L, $p < 0.001$) than mothers of healthy children. Arsenic levels were below the limit of detection in both groups ($p = 1.00$).

Table 4: Heavy metals (Pb, Hg, and As) concentration in the blood of children from the same mothers

Heavy metal concentration in blood	**Children with Autism (n = 218)**	**Healthy children (n = 218)**	**t-test**	**df**	**p value**
Lead (Pb)			32.38	434	<0.001
Mean (µg/L)	2.91	2.02			
SD	0.27	0.30			
Mercury (Hg)					
Mean (µg/L)	1.53	1.18	13.78	434	<0.001
SD	0.30	0.22			
Arsenic (As)					
Mean (µg/L)	<LOD	<LOD	0.00	0.00	1.00
SD	0.00	0.00			

df = degree of freedom, $p < 0.05$ is significant, n = sample size, µg/L = microgram per litre

Table 4 unveiled that Children with autism had significantly higher blood concentrations of lead (2.91 µg/L vs. 2.02 µg/L, $p < 0.001$) and mercury (1.53 µg/L vs. 1.18 µg/L, $p < 0.001$) than healthy children. Arsenic levels were below the limit of detection in both groups ($p = 1.00$).

Discussion

This study found significantly higher blood lead (Pb) concentrations in mothers of children with autism compared to mothers of healthy children, suggesting a potential association between lead exposure and autism. Lead, a known neurotoxin, interferes with neurodevelopment, particularly during pregnancy, potentially leading to structural and functional impairments in the foetal brain (Capitão et al., 2022). The findings support previous research linking lead exposure to cognitive deficits and neurodevelopmental disorders, including autism (Skogheim et al., 2021). Although this study used a descriptive

correlational design, unlike Skogheim et al.'s (2021) prospective cohort study, both align in highlighting lead's neurotoxic potential. Lead exposure from industrial emissions, contaminated water, and lead-based paints underscores the need for stronger environmental regulations.

Similarly, mothers of children with autism exhibited significantly higher mercury (Hg) levels than those of healthy children, reinforcing mercury's neurotoxic role in autism risk. Mercury disrupts neurodevelopment through oxidative stress, cellular signaling interference, and neurotransmitter disruption (Elsagh et al., 2021). This contrasts with Golding et al. (2018), who found no significant mercury-autism association in the UK, possibly due to stricter environmental policies. However, studies by Hessabi et al. (2019) and Qin et al. (2018) support mercury's role in neurodevelopmental impairments. Mercury exposure sources, including industrial emissions and contaminated fish, necessitate improved public health interventions to mitigate exposure during pregnancy.

Conversely, arsenic (As) was undetectable in all maternal blood samples, suggesting it is not a contributing factor to autism in this study population. This aligns with Hessabi et al. (2019), who reported similar findings. While arsenic contamination poses risks in certain regions (Filon et al., 2020), its absence here indicates minimal exposure. However, long-term biomonitoring using alternative matrices, such as hair or nails, could provide further insights.

The study found significantly higher lead (Pb) concentrations in children with autism compared to healthy children, suggesting a potential link between lead exposure and autism. This aligns with previous research indicating that lead disrupts brain development by interfering with synapse formation, neurotransmitter release, and mitochondrial function (Mousavi et al., 2022). Moreover, lead-induced oxidative stress and inflammation have been implicated in autism pathology (Collin et al., 2022). However, these findings contrast with Rahbar et al. (2021), who reported no significant difference in blood lead levels between children with autism and healthy peers. The discrepancy may stem from differences in sample age groups, as older children may have had more time to excrete lead. Notably, this study controlled for maternal factors, enhancing the reliability of its conclusions.

Additionally, children with autism exhibited significantly higher mercury levels than their healthy siblings, reinforcing the hypothesis that environmental toxins contribute to autism risk. Mercury is a known neurotoxin that disrupts synaptic function and neurotransmitter systems (Li et al., 2018). This study's results align with Hessabi et al. (2019) and Qin et al. (2018), who also found elevated mercury levels in children with autism. In contrast, arsenic was below detectable limits, suggesting it may not play a significant role in autism within this population.

Limitations

This study has several limitations. First, while it identified elevated lead and mercury levels in children with autism, it cannot establish causation due to its observational design. Second, environmental exposure sources were not directly measured, limiting conclusions about specific pathways of heavy metal exposure. Third, the study focused on a specific geographic region, reducing generalizability to other populations with different environmental conditions. Additionally, while sibling comparisons controlled for genetic and maternal factors, other confounding variables, such as diet and medical history, could not be fully accounted for. Lastly, arsenic levels may have been undetectable due to methodological detection limits.

Conclusion

This study provides evidence of elevated lead and mercury levels in children with autism compared to their healthy siblings, suggesting a possible role of environmental toxicants in autism spectrum disorder (ASD). While causality cannot be established, these findings highlight the need for further investigation into the mechanisms linking heavy metal exposure to neurodevelopmental outcomes. The absence of detectable arsenic suggests that it may not be a significant factor in this population. Strict environmental regulations should be enforced to minimize exposure to neurotoxic metals.

Declarations

Ethics approval: This study was approved by the University of Port Harcourt Nigeria IRB.
Consent for publication: All contributors gave consent to publish this study.
Competing interests: The contributors declare no competing interests
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Authors' contributions: SB-D conceptualized the study. SB-D, AE, AF-B contributed in the design process and data acquisition. SB-D and CE contributed in the analysis, interpretation of results, drafting and revising the manuscript. SB-D, AE, AF-B, and CE approved the final version.

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