Heavy Metal Blood Levels and Hearing Loss in Children of West Bengal, India

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Abstract

Introduction: Heavy metals are a major environmental threat in India and there are several health risks associated with it. The aim of this study was to investigate the relationship between the blood levels of lead, cadmium, arsenic, and mercury and a sensorineural hearing loss in children aged one to ten years.

Method: Heavy metal blood levels were determined using inductively coupled plasma mass spectrometry, with appropriate quality control.

Results: We found significantly higher blood lead concentration (mg/L; Mean ± SE) in children with a hearing loss (53.2 ± 4.4) compared to healthy controls (38.4 ± 4.7)/P = 0.03/.

Conclusion: Children’s blood lead levels ≥ 50 mg/L compared to the levels < 10 mg/L were associated with increased probability of hearing loss (OR, 48.8; 95% CI, 41.9–55.6). The differences in the blood levels of cadmium, arsenic, and mercury between the children with a hearing loss and controls were statistically insignificant (P > 0.05).

Keywords: Arsenic, blood, cadmium, children, hearing loss, lead, mercury

INTRODUCTION

Heavy metals are naturally occurring elements that have a high atomic weight and a density at least five times greater than that of water. Most commonly encountered heavy metals are lead, mercury, cadmium, and arsenic. For example, some heavy metal-specific gravities (g/cm³) are: arsenic, 5.7; cadmium, 8.7; iron, 7.9; lead, 11.3; and mercury, 13.5. As these heavy metals are not metabolized by the body, they accumulate in the soft tissues or in the bones causing toxic effects. Heavy metals may enter the human body through food, water, air, or absorption through the skin; when they come in contact with humans in residential and occupational settings, as well as from the general environment.[1] A growing body of evidence suggests that exposure to ototoxic environmental and industrial chemicals also may impact the auditory system and lead to hearing loss.[2]

Cadmium and lead are regarded as some of the main contributors to diseases associated with heavy metal poisoning.[3] Low levels of lead and cadmium exposure may contribute to hearing loss. Hearing loss was seen at metal concentrations common in the general population and below current workplace standards. Hearing ability has dropped about 14% to 19% in the USA.[4] Investigational studies suggest that lead exposure can cause degeneration in the inner ear receptor cells and affect latency in auditory nerve conduction velocity,[5-7] and that cadmium exposure causes apoptosis and alters the arrangement of inner ear receptor cells leading to an elevation in auditory thresholds.[8,9] However, few epidemiologic studies of the association between low-level lead exposure and hearing loss have been conducted in the general population.[10] It has also been suggested that lead exposure results in hearing loss, which in turn may be responsible for developmental learning disabilities.[4]

Research conducted among pregnant women showed that higher lead levels in women’s blood in the first and second...
trimester are main contributors to the risk of premature birth.\textsuperscript{[11]} No level of lead in children’s blood is currently thought to be safe, but the US Centers for Disease Control and Prevention (CDC) identified 10 µg/dL as a level of concern before 2012, and since 2012 it has been lowered to 5 µg/dL.\textsuperscript{[12]}

Osman et al.\textsuperscript{[13]} reported delayed wave I of the auditory brainstem response in children exposed to lead. There are substantial differences in the literature regarding the effects of lead on hearing assessed by pure tone audiometry. Mercury causes hearing loss and neuro-behavioral development disorder in children. Methyl mercury is considered a more toxic compound than mercuric chloride.\textsuperscript{[14]}

In India, around 63 million people suffer from significant hearing loss.\textsuperscript{[15]} The National Sample Survey 58th round (2002) surveyed disability in Indian households and found that hearing disability was second most common cause of disability and top most cause of sensory defect. In urban areas, hearing loss was 9% of all disability and in rural areas, it was 10%.\textsuperscript{[16]} A person who is not able to hear as well as someone with normal hearing—hearing thresholds of 25 dB or better in both ears—is said to have hearing loss. It is diagnosed when hearing testing finds that a person is unable to hear 25 decibels in at least one ear.\textsuperscript{[17]} Hearing screening is recommended for all newborns. Approximately 50% of all cases of congenital hearing loss are attributable to environmental factors. The other 50% of cases are thought to be inherited, i.e. of genetic causes. Of these hereditary cases, approximately 30% are classified as syndromic. The other 70% of hereditary cases are classified as non-syndromic.\textsuperscript{[18]} Syndromic hearing loss occurs with signs and symptoms affecting other parts of the body. In contrast, non-syndromic hearing loss is a partial or total loss of hearing that is not associated with other signs and symptoms. This is the otherwise perfectly normal child with the exception of hearing loss.\textsuperscript{[15]}

The aim of this study was to investigate the relationship between heavy metal exposure and hearing loss in children diagnosed with non-syndromic hearing loss in the eastern region of India.

MATERIAL AND METHODS

The study protocol was reviewed and approved by the Institutional Ethics Committee. An informed consent was obtained from the parent/guardian of each child. The study was conducted from May 2015 to August 2018 on 1213 children who visited the ENT Department of Ramakrishna Mission Seva Pratishtan due to their hearing problem. Out of 1213 examined children 70 cases were diagnosed as non-syndromic hearing loss (NSHL) patients. These cases and 30 normal hearing healthy controls were screened by otoacoustic emissions (OAE) and brainstem evoked response audiometry (BERA).

The primary purpose of distortion product otoacoustic emission (DPOAE) tests is to determine cochlear status, specifically hair cell function. This information can be used to (1) screen hearing status (particularly in neonates, infants or individuals with developmental disabilities), (2) partially estimate hearing sensitivity within a limited range, (3) differentiate between the sensorineural and conductive hearing loss, and (4) test for functional (feigned) hearing loss. The information can be obtained from patients who are sleeping or even comatose because no behavioral response is required.\textsuperscript{[19]}

Both the controls and patients aged between one and ten years were screened using OAE as the first level of screening. This was done in the Department of Otorhinolaryngology at Ramakrishna Mission Seva Pratishthan hospital using a GSI Audio Screener, which is a completely automated analysis system that gives a “PASS” or “REFER” result. Absence of emissions for 2 out of the 4 frequencies tested (2 kHz, 3 kHz, 4 kHz, and 5 kHz) was given a “REFER” result. Patients who failed the initial screening were subjected to repeat testing with DPOAE and if they failed again after 6 months refer to BERA test.

BERA/Auditory Brainstem Response (ABR) testing was performed using clicks presented through Oticon BC vibrator. The stimulus either in the form of click or tone pip is transmitted to the ear via a transducer placed in the insert earphone or headphone. The wave forms of impulses generated at the level of brain stem are recorded by the placement of electrodes over the scalp.\textsuperscript{[20]} BERA was used to confirm the hearing loss of the patients less than 5 years of age who failed the OAE screening.

A detailed medical history and pedigree structure were obtained through personal interviews with the selected individual families. The medical history included obstetric and perinatal data (whether the mother experienced history of infection, diabetes mellitus or other infections, or was exposed to medication, drugs or any vaccinations during pregnancy) and information on area of origin, consanguinity, onset, course, and duration of hearing loss, symmetry of the hearing impairment, history of chronic diseases such as middle ear infections, medical treatment, noise damage, trauma, cardiovascular diseases, jaundice in early age, ototoxic agents, head trauma, fever, and ear operations. Two milliliters of venous blood was obtained from the patients and controls in Ethylenediaminetetraacetic acid (EDTA) vials.

A simple and high-throughput method was used for the determination of arsenic (As), cadmium (Cd), lead (Pb), and mercury (Hg) in blood samples. All measurements were conducted using an Inductively Coupled Plasma Mass Spectrometer (ICP-MS) (Elan DRC II PerkinElmer, United States). A clean laboratory and laminar-flow hood capable of producing class 100 were used for preparing solutions. High purity deionized water (resistivity 18.2 MΩ-cm) obtained using a Milli-Q water purification system was used throughout. All solutions were stored in high-density polyethylene bottles. Plastic bottles and glassware materials
were cleaned by soaking in 10% v/v HNO₃ for 24 h, rinsing five times with Milli-Q water, and dried in a class 100 laminar flow hood before use.

For the proposed method with ICP-MS detection,²¹,²² multi-element stock solutions containing 1 g/L of each element were obtained from Perkin-Elmer (PerkinElmer, USA). Analytical calibration standards were prepared daily over the range of 0–50 μg/L for all elements by suitable serial dilutions. Patient and control blood samples (200 μl), were pipetted into (15 ml) conical tubes. Then, 500 μl of 10% v/v Tetramethylammonium hydroxide solution was added to the samples, incubated at room temperature for 10 min, and then the volume was made up to 10 ml with a solution containing 0.05% w/v EDTA + 0.005% v/v triton X-100. Rhodium was added as internal standard to obtain final concentration 10 μg/L. After that, samples were directly analyzed by ICP-MS.²³

STATISTICS

Statistical analyses were carried out using standard descriptive methods, Pearson Chi-Square test and Levene’s F test. Model assumptions were checked by means of residual analyses. Statistical significance was based on the level of two-tailed P-value of 0.05.

RESULTS

A total of 100 examinees (70 cases non-syndromic hearing loss and 30 controls) were investigated. Among the deaf patients, 42 (60%) were males and 28 (40%) females and among control cases, 19 (63%) were males and 11 (37%) females. Table 1 shows the general characteristics of the study participants. The majority of children were in the age group of two to less than four years, the majority of mothers of children with a hearing loss had secondary education while the majority of mothers of controls had a higher secondary education. The concentrations of lead in blood samples of patients with a hearing loss were significantly higher than in controls. A blood lead level greater than or equal to 50 μg/L compared to less than 10 μg/L was associated with increased odds of high-frequency hearing loss (OR, 48.79; 95% CI, 41.95-55.63).

On the opposite, the differences in blood levels of cadmium and mercury and arsenic between the children with a hearing loss and the controls were statistically insignificant [Table 2].

DISCUSSION

Our results clearly indicate that a hearing loss in children is positively related to blood lead concentrations. On the opposite, children’s blood levels of cadmium, mercury, and arsenic are not significantly related to hearing loss.

A number of previous studies have reported associations between blood lead levels and hearing impairment. Some studies have shown that occupational exposure to lead may induce higher hearing thresholds in an adult population.²⁴-²⁶

In a study on the general US population, Choi et al.⁴ found that the hearing threshold of the highest quintile groups, based on blood lead levels, was higher compared to the lowest quintile group. Also, it has been shown that a high prenatal blood lead level may adversely affect the development of the auditory system.²⁷,²⁸ Shargorodsky et al.²⁹ reported in a US adolescents study, that a high-frequency hearing

| Characteristic                      | Children with a hearing loss (n = 70) | Healthy controls (n = 30) | Pearson Chi-Square P-value |
|-------------------------------------|--------------------------------------|---------------------------|---------------------------|
| Sex                                 |                                      |                           |                           |
| Male                                | 42                                   | 19                        | 0.75                      |
| Female                              | 28                                   | 11                        |                           |
| Age                                 |                                       |                           |                           |
| <2years                             | 4                                    | 0                         | 0.002                     |
| 2–<4 years                          | 29                                   | 6                         |                           |
| 4–<6 years                          | 7                                    | 4                         |                           |
| 6–<8 years                          | 15                                   | 2                         |                           |
| 8–<10 years                         | 15                                   | 18                        |                           |
| Speech level                         |                                       |                           |                           |
| No speech                            | 33                                   | 0                         | 0.001                     |
| Few word                            | 29                                   | 3                         |                           |
| Normal                              | 8                                    | 27                        |                           |
| Literacy of the mother              |                                       |                           |                           |
| Illiterate                          | 11                                   | 0                         | 0.009                     |
| Primary⁴                            | 2                                    | 1                         |                           |
| Secondary⁵                          | 34                                   | 10                        |                           |
| Higher secondary⁶                   | 10                                   | 13                        |                           |
| Graduation or above                 | 13                                   | 6                         |                           |
| Congenital anomaly                  |                                       |                           |                           |
| Yes                                 | 20                                   | 1                         | 0.005                     |
| No                                  | 50                                   | 29                        |                           |
| Cardiovascular disorder             |                                       |                           |                           |
| Yes                                 | 8                                    | 0                         | 0.054                     |
| No                                  | 62                                   | 30                        |                           |
| Jaundice                            |                                       |                           |                           |
| Yes                                 | 13                                   | 1                         | 0.44                      |
| No                                  | 57                                   | 29                        |                           |
| Developmental milestones            |                                       |                           |                           |
| Delayed                             | 32                                   | 0                         | 0.001                     |
| Normal                              | 38                                   | 30                        |                           |
| Psychometric analysis               |                                       |                           |                           |
| Down Syndrome                       | 1                                    | 0                         | 0.51                      |
| Normal                              | 69                                   | 30                        |                           |
| Special investigation               |                                       |                           |                           |
| OAE/ BERA                           |                                       |                           |                           |
| Pass                                | 0                                    | 30                        | 0.001                     |
| Fail                                | 70                                   | 0                         |                           |
| Total                               | 70                                   | 30                        |                           |

⁴Primary—Upto Class IV; ⁵Secondary—Class V to Class X; ⁶Higher secondary—Class XI to Class XII.
impairment was more frequent among those with a blood lead level over 2 μg/dl compared to a reference group.

Park et al.\textsuperscript{[10]} have concluded that the potential of lead to affect hearing in the general population is accumulating steadily. To our knowledge, this is the first Indian epidemiologic study that associates children’s high blood lead level and hearing impairment.

Using an auditory brainstem response test we show that high blood lead levels are associated with the conductivity along the auditory pathways. Likewise, Jones et al.\textsuperscript{[30]} reported that lead changed the axonal structure and function of the brainstem auditory nuclei. An alternative hypothesis is that toxic metals affect intracellular calcium homeostasis\textsuperscript{[31]} and accordingly, chronic lead exposure induces auditory hair cell death.\textsuperscript{[4]}

We found no evidences about cadmium, mercury, and arsenic adverse effect on children’s hearing. Mercury affects hearing, with central conduction time delay (ABR I-V, III-V), but cochlear function may be unaffected. McBride and Williams\textsuperscript{[32]} reported that cadmium causes dose-dependent loss of hearing in rats, and a delay in only Wave I of the auditory brainstem response implies cochlear dysfunction. A zinc-enriched diet reduces the ototoxic effect of cadmium. DeAbreu and Suzuki\textsuperscript{[33]} examined the effect of cadmium fumes and noise exposure and showed that hearing loss at 4 kHz and 6 kHz was more severely affected with combined exposure. An advantage of the present study is that it was conducted on a representative sample of the eastern part of Indian population, and the findings can be generalized to the entire Indian population. Although evidence for the link between hearing impairment and heavy metals such as lead is accumulating, most studies were based on animal experiments.\textsuperscript{[34-36]} Epidemiologic studies targeting the general population, such as this study, are rare.

The strengths and limitations of this study should be considered. A case-control method of this study is helpful in causality investigations. Data from our study are comprehensive and nationally representative sample of participants. The audiometric assessment of hearing loss is the criterion standard objective measure and has been shown to be reliable in numerous studies. Although blood lead levels have a relatively short half-life of approximately 30 days, they are the accepted measure of assessing current lead exposure.\textsuperscript{[37]} We included the known possible risk factors for hearing loss in the analyses, the data on risk factors for children hearing loss are currently limited, and other confounders may exist that were not included in our study.

**CONCLUSIONS**

In conclusion, we show that a high blood lead level in children is associated with hearing loss. Therefore, lead exposure should be lowered to protect children’s hearing.

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**Conflicts of interest**

The authors declare that they have no competing interests.

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