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Editorial

An overview of analytical considerations of Lead estimation in biological materials

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Introduction

Lead (Pb) is an environmental toxin which is cumulative and affects the neurological, hematological, gastrointestinal, cardiovascular, musculoskeletal and renal systems. Hypertension, ischemic heart disease, kidney disease and infertility are among the long-term complications. Pb has neurotoxic effects on children, and even modest amounts of exposure can impede cognitive development and produce behavioural problems. India, Indonesia, Philippines, Nigeria, China, Pakistan, Brazil, Mexico, Peru, France and United States of America are countries with a major prevalence of Pb toxicity. Major route of Pb entry into body is through oral cavity; around 5–15% of Pb is absorbed by gastrointestinal tract and the rest is excreted in the feces. Pb particles can also be inhaled; ciliary action of respiratory epithelial cells enables the movement of Pb particles down the laryngopharynx.[1]

Cooking utensils containing Pb and unusual sources like spices, cultural powders, traditional eye cosmetic to ward off the "evil eye" (kohl) are the notable sources of Pb exposure among children in India. The feasibility of traditional Indian medicine as a source of Pb exposure has been explored in pediatric Ayurveda treatment subjects in South India.[2]

There is a pressing need to develop plans for human biomonitoring studies, blood Pb prevalence screening, and other public health actions related to the assessment of human Pb exposure.

Common indications for estimation of lead

The common indications for estimation of Pb are constituent of active management of Pb exposure. By the time, signs and symptoms of Pb poisoning develop significant damage would have already occurred to the target organs which emphasizes the need to screen and identify Pb exposure.[3]

Thus, the vulnerable population with risk of lead exposure needs to be monitored. Lead is estimated in the assessment of risk in the highly exposed population. In comparison to adults, children are at an inordinate risk of developing lead toxicity due to the immature central nervous system and an incomplete blood brain barrier which is permissible to Pb.[2]

Primary prevention and secondary prevention strategies comprising of control of sources of exposure and case management in children can be planned in accordance with screening blood lead levels. Follow-up testing along with parental education, counseling and remediation of potential sources of lead in children's environment is critical.[3]

Prevalence screening is also done to know the ill effects expected in highly Pb exposed population viz police personnel, workers in industries related to paints and varnishes. Surveillance monitoring is done in population studies of at risk group.

Specimen for Lead estimation

Monitoring of Pb in biological materials is universally acknowledged to be dependent on the quality of sample collection and sample handling. The various specimens for Pb estimation include whole blood, plasma, urine, nail clippings and hair. Blood Pb estimation is required to ensure proper case management. Apart from blood sample, urine is also routinely collected.[5,6] However, Pb can be estimated in almost all biological materials.

Sample collection

Around 4 mL whole blood needs to be collected in the metal free Royal Blue Top (K2EDTA) tube for blood Pb levels estimation. If not venous blood then capillary blood can also be used in field testing for prevalence screening.[7]

Studies have revealed that either random or 24 hours urine sample may be collected in sterile acid washed urine container free from any metal and values are not altered. However, urine is not a reliable marker because it may be affected by alterations in kidney function, interindividual and circadian variation.[7,8] It is mandatory that the sample collection containers should be acid washed or certified lead free to be acceptable.[7]

Pb is ambivalent in nature. The pervasiveness of Pb means that the risk of sample contamination is high and contamination can occur at each stage: sample collection, storage, transport and manipulation. A clean location should, therefore, be identified for collection and storage of samples– this is particularly important if samples are being collected in the field.

Potential sources of contamination include:

- Environment in which blood sample collection is performed
- Contamination on the skin especially finger prick
- Collection supplies like tubes, needles, pipette tips, vials which may come in contact with the blood sample
- Laboratory environment where the assay shall be performed[6]

Analyzing venous blood for Pb is preferred for confirmation of exposure, diagnosis, decisions on the medical management of Pb poisoning and prognosis. Capillary blood sampling, involving a finger (or heel prick for babies), is relatively easy and quick and is less invasive but may be affected by contamination (giving false positives)[4]

Analytical methods used to measure lead

Blood Pb concentration measurement can be performed using laboratory methods and point-of-care or field-testing methods.

Significant differences exist between these methods in context to their analytical measurements (limits of detection, accuracy and precision), costs (purchase and maintenance costs, laboratory infrastructure requirements, reagents and supplies) and technical requirements (sample preparation, calibration and skilled personnel). In addition to the setting and resources of the laboratory these aspects will impact the decision about the choice of method.

Laboratory methods

Flame atomic absorption spectrometry (AAS), graphite furnace atomic absorption spectrometry (GFAAS), anode stripping voltammetry (ASV), inductively coupled plasma-atomic emission spectroscopy (ICP/AES) & inductively coupled plasma mass spectrometry (ICP/MS) are the most commonly employed laboratory methods for Pb estimation.[9]

Laboratory methods based on Atomic Absorption Spectrometry, Inductively coupled plasma mass spectrometry (ICP/MS) and Anodic Stripping Voltammetry require trained staff, though the level of training and skill required varies with the instrumentation.[4]

Laboratory methods represent confirmatory evidence of lead exposure. For laboratories intending to analyse multiple trace elements in a given sample ICP/MS is recommended. Added advantage with ICP/ MS is the isotope ratio of the lead in the sample can be determined which makes it feasible to identify the particular source of lead exposure in environment.[4]

Point of Care testing (POCT)

Devices based on portable ASV are very simple to use and do not require trained laboratory personnel. It is imperative to consider and mitigate environmental factors that may interfere with their reliability when using these devices in the field.

The environmental factors include contamination risks, a clean space for sampling and testing, for adequately cleansing the skin of test subjects the availability of water and soap is a must, apposite temperature conditions for the optimal functioning of the analyzers and uninterrupted electricity supply.[4]

POCT devices are mainly utilized for screening purpose. However, these can be used for active management of lead poisoning in resource poor setting when there is no access for sample transport to referral laboratory.

Apart from direct estimation of lead in the biological materials there are biomarkers of lead exposure. Coproporphyrin, δ-aminolevulinic acid (ALA), and erythrocyte protoporphyrin (EP) concentrations and ALA dehydratase (ALAD) activity are some of the biomarker assays for monitoring lead exposure and toxicity. These are performed by means of standard clinical laboratory techniques.[10]

Unit of Measurement & Conversion Factor

Micrograms of Pb per decilitre of blood (µg/ dL), micrograms per litre (µg/L) and micromoles per litre (µmol/L) are the universally used units for reporting blood Pb concentration. The atomic mass of Pb (207.19) is used as conversion factor between mass and molar units.[4]

It is tacit to be careful when making the conversion as the denominator can be decilitres or litres.

- For conversion from mass to molar units the value should be divided by the atomic mass.
- For conversion from molar to mass units the value should be multiplied by the atomic mass.

Laboratory mandates

Referral laboratory should be accredited by a national accreditation body. In India, the referral laboratory should be National Accreditation Board for Testing and Calibration Laboratories (NABL) accredited. The referral laboratory should be having internal quality control in place and participating in proficiency testing programme.

Conclusion

Lead exposure is common in India probably due to lack of stringent curtailing sources of lead. Human biomonitoring protocols should be strengthened. Prevalence screening may be carried out using portable POCT devices based on anodic stripping voltammetry (ASV). For active case management laboratory methods based on Flame atomic absorption spectrometry (AAS), graphite furnace atomic absorption spectrometry (GFAAS), anode stripping voltammetry (ASV), inductively coupled plasma-atomic emission spectroscopy (ICP/AES), and inductively coupled plasma mass spectrometry (ICP/MS) remain confirmatory and help in identifying the source, clinical decision making and prognosis.

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