

## Original article

# Contributory influence of supplemental intake and environmental pollutants on baseline serum and human milk levels of lead, cadmium, chromium, and zinc in lactating women in Lagos, Nigeria

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## Abstract

**Background:** Routine heavy metal assessment is important in lactating women.

**Objectives:** This study aimed to investigate the concentration of lead, cadmium, and chromium and the protective role of zinc in the peripheral blood and breastmilk of lactating women within four weeks of delivery at the Lagos University Teaching Hospital (LUTH). It also sought to explore the link between heavy metals and supplemental intake, or environmental pollutants linked to heavy metals.

**Methods:** A total of 40 subjects were recruited for the study. The sociodemographic characteristics, supplemental intake use, and possible exposure to some sources of heavy metals were assessed using standard methods.

**Results:** The results revealed that 67.0% of the subjects had lived in Lagos for more than 120 months ( $\geq 120$ ). About 60% reported using eye kohl and some used hair dye. The serum level of lead was observed to be higher in those with a higher intake of Vitamins D and B. The serum level of cadmium was also observed to be lower in those who took calcium as a supplement. Zinc levels were lower in the participants who reported taking folic acid as a supplement.

**Conclusion:** Supplementations with vitamins, calcium and folic acid as well as exposure to hair dye, paints and radiation have the potential to alter the serum and breastmilk levels of important microelements like cadmium, lead and zinc in lactating women.

**Keywords:** Cadmium, lactating women, lead, supplements, zinc.

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Breastmilk is still the best-known natural nutrient-laden food source for infants. Breastmilk contains the right amounts of nutrients a newborn baby requires for growth and immunity.<sup>(1)</sup> In addition, breastmilk contains enzymes and antibodies that help in fighting infections in infants. Unfortunately, breastmilk can be contaminated by environmental pollutants that may be harmful to the infant. There is the possibility of a baby not being protected from toxic metals from its mother's blood and breast milk during pregnancy and lactation.<sup>(2)</sup>

The process of industrialization, urbanization and economic development continue to transform the world over, including cosmopolitan cities like Lagos State, Nigeria. These transformations come with environmental pollution and contamination most of which are man-made. These pollutants include pesticides, biphenyls (PCBs), hexachlorocyclohexane isomers (HCHs), and heavy metals. The longer an individual resides in a megacity with attendant exposure to pollution such as heavy metals, and industrialization the more these toxins stay in the body.<sup>(3)</sup> Heavy metal toxicities have been linked to dietary habits, industrial exposure, driving barefoot, contamination from wrongly coated food containers, some water sources, tooth filling and use of prosthetics dying of hair, ingestion of chips or flakes of lead-based paints, and so on.

The issue of heavy metal contaminants raises great worries for infants born particularly in highly polluted areas. Infants are most vulnerable to heavy metal poisoning compared to adults.<sup>(4)</sup> Metallic elements categorized by relatively high density and toxicity at low concentrations are referred to as heavy metals. These elements include lead, cadmium, and chromium.<sup>(5)</sup> Research reports emphasize the fact that heavy metals can be transferred from the mother's blood to a breastfed infant.<sup>(6,7)</sup>

Lead, cadmium, and chromium can thus cross the placenta and blood-brain barrier and can be found in maternal milk. This poses a potential risk for newborns.<sup>(8)</sup> The mother's milk is the sole source of food for the infant for the first six months, thus it is crucial to monitor the concentration of heavy metals in the maternal circulation and breastmilk as well as their determinants. Exposure to lead, cadmium, and chromium is most associated with toxicity and poisoning in humans.<sup>(9)</sup> Lead toxicity for instance has been associated with neurodevelopmental delay in children.<sup>(10)</sup> The heavy metal toxicants can stimulate

or suppress the immunomodulatory workings and can also influence indirectly the various organs and systems in the body.<sup>(11)</sup> Zinc, a micronutrient element has been reported to possess ameliorative properties against heavy metal toxicity.<sup>(12)</sup>

The reports from the International Centers for disease control reveal that breastmilk is not routinely assessed for environmental pollutants.<sup>(9)</sup> There is thus the need for regular monitoring of the mother's milk to evaluate the exposure of the mother and the baby also to heavy metal poisoning. Evaluation of heavy metals in breast milk is a non-invasive way to monitor the effects of environmental pollution on humans.<sup>(8, 13)</sup>

This study aimed to investigate the concentration of lead, cadmium, and chromium in the peripheral blood and breastmilk of lactating women within four weeks of delivery at the Lagos University Teaching Hospital, Lagos State, Nigeria. This study also explored the association between heavy metals, the role of zinc, and selected sources of exposure to environmental pollutants.

## **Materials and methods**

### ***Study area***

The observational study was carried out at the Postnatal wards and postnatal clinics of the Department of Obstetrics and Gynecology, Lagos University Teaching Hospital (LUTH).

### ***Study design***

The study subjects were eligible breastfeeding mothers with neonates (new-born less than 28 days old), who gave informed written consent to enroll in the study. Ethical approval was obtained from the Health research ethics committee of the College of Medicine, University of Lagos (CMUL/HREC/06/19/55).

### ***Exclusion criteria***

The exclusion criteria for the study were complaints of ill health of either the mother or the baby, hypertensive, diabetic, and asthmatic women, not being able to express milk, and smoking.

### ***Data collection***

The informed consent followed the international committee of medical journal editors (ICMJE) requirements. After obtaining informed consent, each

potential study subject was counseled on the study, and relevant information was collected using a structured questionnaire designed for the study. The questionnaire sought information on socio-demographic characteristics (such as marital status, level of education, occupation, lifestyle, and smoking); obstetric variables (such as mode of delivery, gestational age at delivery, previous pregnancies; the number of children, and infant feeding practices); nutritional/dietary history (such as consumption of nutritional supplements); and possible exposure to known sources of heavy metals (such as the use of cosmetics like eye kohl, lipsticks, and hair dyes, and exposure to chemicals like paints, pesticides, herbicides, and asbestos). Thereafter, biological (blood and breastmilk) specimens were obtained from each subject as per study protocol.

#### ***Determination of lead, cadmium, chromium, and zinc in maternal blood and breastmilk***

Five milliliters of venous blood were drawn from the antecubital vein and stored in plain vacutainer tubes, and 3 - 5 milliliters of expressed breastmilk were collected into a sterile sample bottle (after the subject cleaned her nipples and washed her hands properly). The blood sample was allowed to clot at room temperature for 20 minutes and centrifuged at 3,000 rpm for 15 minutes using an Eppendorf 5415C centrifuge (Eppendorf AG, Germany). The serum was collected in 2mL sterile cryovials. The biological samples (sera and breastmilk) were properly labeled and stored at - 20°C until analysis.

#### ***Laboratory methods and techniques***

The specimens were analyzed for selected heavy metals, (lead, cadmium, and chromium) as well as zinc at the Central Research Laboratory, University of Lagos using atomic absorption spectroscopy. The stored samples were allowed to thaw on the workbench and all assays were performed at room temperature as a batch.

#### ***Statistical analysis***

Data were analyzed using STATA software version 13 (College Station, TX: StataCorp LP). A test of normality was performed using the Shapiro-Wilk test. Qualitative data were presented as frequencies and percentages while quantitative data were represented as mean  $\pm$  standard deviation (SD). A  $P < 0.05$  was considered statistically significant.

#### **Results**

A total of 40 subjects were recruited for the study. Out of the 40 subjects, 30 (75.0%) had complete data and these were included in the final statistical analysis.

**Table 1** shows the baseline characteristics of the subjects. About two-thirds of the study subjects have resided in Lagos for more than 120 months. The study subjects were exposed to the use of eye kohl and lead cosmetics (60.0%). **Table 2** shows that blood lead level was significantly higher in the blood compared to human milk.

**Table 3** shows the use of supplements by the study subjects during pregnancy. Serum lead level was significantly higher ( $P < 0.05$ ) in subjects who took Vitamins D and B as supplements while serum cadmium level was significantly lower ( $P < 0.05$ ) in those who took calcium as a supplement. Serum zinc level was also significantly lower ( $P < 0.01$ ) in those who took Folic acid as a supplement.

**Table 4** shows the use of supplements during pregnancy on breast milk elements. Cadmium levels were significantly lower ( $P < 0.05$ ) in subjects who took Folic acid as a supplement. Zinc levels were significantly lower ( $P < 0.001$ ) in those who took Folic acid as a supplement. **Table 5** shows a significant link between hair dye use and a significantly high serum cadmium level of the subjects who used it. Serum lead level was significantly lower ( $P < 0.05$ ) in subjects who took borehole water. Zinc levels were significantly higher ( $P < 0.01$ ) in subjects exposed to paints and radiation. **Table 6** shows that cadmium and zinc levels were significantly higher in subjects exposed to paints and radiation.

**Table 1.** General characteristics of the subjects.

| Characteristics                 |                      | Frequency (%)     |
|---------------------------------|----------------------|-------------------|
| Age (years)                     | 15 - 34              | 22 (73.0)         |
|                                 | 35 - 54              | 8 (27.0)          |
|                                 | <b>Total</b>         | <b>30 (100.0)</b> |
| Marital status                  | Single               | 2 (7.0)           |
|                                 | Married              | 28 (93.0)         |
|                                 | <b>Total</b>         | <b>30 (100.0)</b> |
| Number of full-term pregnancies | 1                    | 10 (34.0)         |
|                                 | 2                    | 7 (23.0)          |
|                                 | 3                    | 8 (27.0)          |
|                                 | 4                    | 4 (13.0)          |
|                                 | 5                    | 1 (3.0)           |
|                                 | <b>Total</b>         | <b>30 (100.0)</b> |
| Use of eye kohl and lead        | Yes                  | 18 (60.0)         |
|                                 | No                   | 12 (40.0)         |
|                                 | <b>Total</b>         | <b>30 (100.0)</b> |
| Region of origin                | Northern Nigeria     | 5 (17.0)          |
|                                 | Southeast Nigeria    | 10 (33.0)         |
|                                 | South-South Nigeria  | 3 (10.0)          |
|                                 | Southwest Nigeria    | 11 (37.0)         |
|                                 | Foreigner            | 1 (3.0)           |
|                                 | <b>Total</b>         | <b>30 (100.0)</b> |
| Level of education              | Secondary level      | 14 (47.0)         |
|                                 | Tertiary level       | 16 (53.0)         |
|                                 | <b>Total</b>         | <b>30 (100.0)</b> |
| Employment                      | Unemployed           | 3 (10.0)          |
|                                 | Employed             | 27 (90.0)         |
|                                 | <b>Total</b>         | <b>30 (100.0)</b> |
| Mode of delivery                | Vaginal delivery     | 17 (57.0)         |
|                                 | Cesarean delivery    | 13 (43.0)         |
|                                 | <b>Total</b>         | <b>30 (100.0)</b> |
| Gestational age delivery        | Preterm (< 37 weeks) | 4 (13.0)          |
|                                 | Term (≥ 37 weeks)    | 26 (87.0)         |
|                                 | <b>Total</b>         | <b>30 (100.0)</b> |
| Parity                          | 1                    | 10 (33.0)         |
|                                 | 2                    | 8 (27.0)          |
|                                 | 3                    | 7 (23.0)          |
|                                 | ≥ 4                  | 5 (17.0)          |
|                                 | <b>Total</b>         | <b>30 (100.0)</b> |
| Cigarette smoking               | Yes                  | 0 (0.0)           |
|                                 | No                   | 30 (100.0)        |
|                                 | <b>Total</b>         | <b>30 (100.0)</b> |
| Period of residence in Lagos    | < 120 months         | 10 (33.0)         |
|                                 | ≥ 120 months         | 20 (67.0)         |
|                                 | <b>Total</b>         | <b>30 (100.0)</b> |

**Table 2.** Baseline levels of lead, cadmium, chromium, and zinc in the blood and breastmilk.

|                   | Blood        | Breastmilk    | P - value |
|-------------------|--------------|---------------|-----------|
| Lead (µmol/L)     | 0.12 ± 0.09  | 0.05 ± 0.06   | 0.001     |
| Cadmium (µmol/L)  | 0.03 ± 0.04  | 0.03 ± 0.04   | 0.7249    |
| Chromium (µmol/L) | 0.07 ± 0.07  | 0.06 ± 0.06   | 0.3773    |
| Zinc (µmol/L)     | 22.42 ± 7.38 | 21.46 ± 13.89 | 0.7411    |

Values represent mean ± standard deviation.

\* $P < 0.05$ , significant value.

**Table 3.** Effect of supplemental intake of vitamins during pregnancy on baseline levels of lead, cadmium, chromium, and zinc in the blood.

| Supplement taken | Elements    |             |             |              |
|------------------|-------------|-------------|-------------|--------------|
|                  | Lead        | Cadmium     | Chromium    | Zinc         |
| Vitamin C        | 0.08 ± 0.06 | 0.05 ± 0.05 | 0.07 ± 0.05 | 21.28 ± 7.66 |
|                  | 0.14 ± 0.09 | 0.03 ± 0.03 | 0.07 ± 0.08 | 22.99 ± 7.38 |
| P - value        | 0.0803      | 0.2023      | 0.9544      | 0.5603       |
| Vitamin D        | 0.08 ± 0.08 | 0.04 ± 0.04 | 0.08 ± 0.08 | 22.17 ± 7.58 |
|                  | 0.17 ± 0.06 | 0.03 ± 0.03 | 0.06 ± 0.07 | 22.85 ± 7.37 |
| P - value        | 0.0105*     | 0.5098      | 0.5542      | 0.8136       |
| Vitamin B        | 0.09 ± 0.09 | 0.05 ± 0.04 | 0.07 ± 0.07 | 24.05 ± 7.59 |
|                  | 0.15 ± 0.08 | 0.02 ± 0.03 | 0.07 ± 0.08 | 20.99 ± 7.13 |
| P - value        | 0.0455*     | 0.0842      | 0.8483      | 0.2643       |
| Calcium          | 0.09 ± 0.09 | 0.05 ± 0.05 | 0.07 ± 0.05 | 24.89 ± 7.42 |
|                  | 0.14 ± 0.08 | 0.02 ± 0.03 | 0.07 ± 0.08 | 20.77 ± 7.09 |
| P - value        | 0.1564      | 0.0391*     | 0.8981      | 0.1364       |
| Folic acid       | 0.05 ± 0.07 | 0.06 ± 0.01 | 0.06 ± 0.01 | 35.10 ± 0.42 |
|                  | 0.13 ± 0.08 | 0.03 ± 0.04 | 0.07 ± 0.08 | 21.51 ± 6.77 |
| P - value        | 0.2548      | 0.4359      | 0.7622      | 0.0093**     |
| Iron             | 0.10 ± 0.10 | 0.03 ± 0.02 | 0.04 ± 0.05 | 25.61 ± 7.89 |
|                  | 0.13 ± 0.09 | 0.04 ± 0.04 | 0.08 ± 0.08 | 21.44 ± 7.12 |
| P - value        | 0.5051      | 0.6551      | 0.1592      | 0.1957       |

Values represent mean ± standard deviation.

Significant value = (\* $P < 0.05$ , \*\* $P < 0.01$ ).

Values at the top represent without supplementation, while values below represent with supplementation.

**Table 4.** Effect of supplemental intake of vitamins during pregnancy on baseline levels of lead, cadmium, chromium, and zinc in the breastmilk.

| Supplement taken | Element     |             |             |               |
|------------------|-------------|-------------|-------------|---------------|
|                  | Lead        | Cadmium     | Chromium    | Zinc          |
| Vitamin C        | 0.04 ± 0.05 | 0.04 ± 0.06 | 0.08 ± 0.05 | 22.88 ± 21.27 |
|                  | 0.06 ± 0.06 | 0.02 ± 0.03 | 0.05 ± 0.06 | 20.76 ± 8.85  |
| <i>P</i> - value | 0.3753      | 0.2269      | 0.1745      | 0.7000        |
| Vitamin D        | 0.05 ± 0.06 | 0.04 ± 0.05 | 0.06 ± 0.06 | 22.62 ± 15.96 |
|                  | 0.05 ± 0.05 | 0.02 ± 0.03 | 0.04 ± 0.06 | 19.46 ± 9.68  |
| <i>P</i> - value | 0.9314      | 0.3         | 0.4401      | 0.5577        |
| Vitamin B        | 0.06 ± 0.06 | 0.04 ± 0.05 | 0.06 ± 0.06 | 24.26 ± 17.93 |
|                  | 0.05 ± 0.05 | 0.02 ± 0.03 | 0.05 ± 0.05 | 19.01 ± 8.97  |
| <i>P</i> - value | 0.7391      | 0.1186      | 0.5658      | 0.3097        |
| Calcium          | 0.06 ± 0.07 | 0.04 ± 0.06 | 0.05 ± 0.06 | 23.73 ± 19.93 |
|                  | 0.05 ± 0.05 | 0.02 ± 0.03 | 0.06 ± 0.06 | 19.96 ± 8.13  |
| <i>P</i> - value | 0.7027      | 0.1838      | 0.4846      | 0.4761        |
| Folic acid       | 0.05 ± 0.07 | 0.09 ± 0.13 | 0.09 ± 0.13 | 58.70 ± 27.44 |
|                  | 0.05 ± 0.06 | 0.03 ± 0.03 | 0.05 ± 0.05 | 18.80 ± 8.32  |
| <i>P</i> - value | 0.9337      | 0.037*      | 0.3925      | 0.0001***     |
| Iron             | 0.09 ± 0.07 | 0.05 ± 0.07 | 0.05 ± 0.07 | 29.06 ± 24.51 |
|                  | 0.04 ± 0.05 | 0.03 ± 0.03 | 0.06 ± 0.06 | 19.15 ± 8.15  |
| <i>P</i> - value | 0.0868      | 0.2445      | 0.6181      | 0.0991        |

Values represent mean ± standard deviation.

Significant value = (\* $P < 0.05$ , \*\*\* $P < 0.001$ ).

Values at the top represent without supplementation, while values below represent with supplementation.

**Table 5.** Effect of environmental pollutants during pregnancy on baseline levels of lead, cadmium, chromium, and zinc in the blood.

| Pollutants       | Element     |             |             |               |
|------------------|-------------|-------------|-------------|---------------|
|                  | Lead        | Cadmium     | Chromium    | Zinc          |
| Dye of hair      | 0.12 ± 0.08 | 0.03 ± 0.03 | 0.07 ± 0.08 | 22.14 ± 7.74  |
|                  | 0.13 ± 0.15 | 0.08 ± 0.06 | 0.05 ± 0.08 | 24.87 ± 1.60  |
| <i>P</i> - value | 0.789       | 0.0256*     | 0.564       | 0.554         |
| Pesticides       | 0.11 ± 0.08 | 0.03 ± 0.04 | 0.08 ± 0.08 | 22.31 ± 7.13  |
|                  | 0.15 ± 0.07 | 0.01 ± 0.00 | 0.01 ± 0.01 | 22.55 ± 15.91 |
| <i>P</i> - value | 0.5341      | 0.3837      | 0.2216      | 0.9663        |
| Smoke            | 0.12 ± 0.08 | 0.04 ± 0.04 | 0.07 ± 0.07 | 22.11 ± 7.04  |
|                  | 0.12 ± 0.09 | 0.02 ± 0.03 | 0.08 ± 0.08 | 23.63 ± 9.27  |
| <i>P</i> - value | 0.9201      | 0.4379      | 0.7899      | 0.6598        |
| Paints           | 0.13 ± 0.08 | 0.03 ± 0.04 | 0.07 ± 0.08 | 21.18 ± 6.66  |
|                  | 0.03 ± 0.06 | 0.06 ± 0.01 | 0.04 ± 0.03 | 33.63 ± 2.73  |
| <i>P</i> - value | 0.0736      | 0.2915      | 0.4321      | 0.004**       |

**Table 5.** (Cont.) Effect of environmental pollutants during pregnancy on baseline levels of lead, cadmium, chromium, and zinc in the blood.

| Pollutants       | Element     |             |             |              |
|------------------|-------------|-------------|-------------|--------------|
|                  | Lead        | Cadmium     | Chromium    | Zinc         |
| Radiation        | 0.13 ± 0.09 | 0.03 ± 0.04 | 0.07 ± 0.08 | 21.51 ± 6.77 |
|                  | 0.05 ± 0.07 | 0.06 ± 0.01 | 0.06 ± 0.01 | 35.10 ± 0.42 |
| <i>P</i> - value | 0.2548      | 0.4359      | 0.7622      | 0.0093**     |
| Asbestos         | 0.12 ± 0.08 | 0.04 ± 0.04 | 0.07 ± 0.07 | 23.13 ± 7.10 |
|                  | 0.12 ± 0.09 | 0.04 ± 0.03 | 0.08 ± 0.09 | 22.20 ± 7.60 |
| <i>P</i> - value | 0.9524      | 0.9972      | 0.6329      | 0.7532       |
| Borehole         | 0.14 ± 0.09 | 0.03 ± 0.04 | 0.08 ± 0.08 | 21.33 ± 7.77 |
|                  | 0.04 ± 0.05 | 0.03 ± 0.02 | 0.07 ± 0.09 | 26.38 ± 2.99 |
| <i>P</i> - value | 0.0191*     | 0.9781      | 0.8744      | 0.1708       |

Values represent mean ± standard deviation.

Significant value = (\**P* < 0.05, \*\**P* < 0.01).

Values at the top represent without supplementation, while values below represent with supplementation.

**Table 6.** Effect of environmental pollutants during pregnancy on baseline levels of lead, cadmium, chromium, and zinc in the breastmilk.

| Pollutants       | Element     |             |             |               |
|------------------|-------------|-------------|-------------|---------------|
|                  | Lead        | Cadmium     | Chromium    | Zinc          |
| Dye of hair      | 0.05 ± 0.06 | 0.03 ± 0.04 | 0.06 ± 0.06 | 21.35 ± 14.55 |
|                  | 0.10 ± 0.00 | 0.07 ± 0.04 | 0.06 ± 0.05 | 22.50 ± 6.52  |
| <i>P</i> - value | 0.1385      | 0.0887      | 0.8901      | 0.8944        |
| Pesticides       | 0.05 ± 0.06 | 0.03 ± 0.04 | 0.05 ± 0.06 | 21.47 ± 14.42 |
|                  | 0.05 ± 0.07 | 0.01 ± 0.01 | 0.10 ± 0.04 | 17.50 ± 9.76  |
| <i>P</i> - value | 0.9659      | 0.4127      | 0.3511      | 0.7075        |
| Smoke            | 0.05 ± 0.05 | 0.03 ± 0.03 | 0.05 ± 0.05 | 19.51 ± 8.43  |
|                  | 0.07 ± 0.08 | 0.03 ± 0.07 | 0.07 ± 0.08 | 29.28 ± 26.46 |
| <i>P</i> - value | 0.5322      | 0.9496      | 0.4085      | 0.125         |
| Paints           | 0.06 ± 0.06 | 0.03 ± 0.03 | 0.05 ± 0.05 | 18.89 ± 8.47  |
|                  | 0.03 ± 0.06 | 0.08 ± 0.09 | 0.06 ± 0.10 | 44.67 ± 31.09 |
| <i>P</i> - value | 0.5322      | 0.03*       | 0.8498      | 0.0011**      |
| Radiation        | 0.05 ± 0.06 | 0.03 ± 0.03 | 0.05 ± 0.05 | 18.80 ± 8.32  |
|                  | 0.05 ± 0.07 | 0.09 ± 0.12 | 0.09 ± 0.13 | 58.70 ± 27.44 |
| <i>P</i> - value | 0.9337      | 0.037*      | 0.3925      | 0.0001***     |
| Asbestos         | 0.05 ± 0.05 | 0.04 ± 0.05 | 0.05 ± 0.06 | 21.86 ± 15.49 |
|                  | 0.08 ± 0.07 | 0.01 ± 0.02 | 0.06 ± 0.05 | 20.49 ± 11.32 |
| <i>P</i> - value | 0.1572      | 0.1661      | 0.8718      | 0.8146        |
| Borehole         | 0.06 ± 0.06 | 0.03 ± 0.05 | 0.06 ± 0.06 | 21.73 ± 15.91 |
|                  | 0.04 ± 0.05 | 0.03 ± 0.02 | 0.06 ± 0.07 | 21.30 ± 3.84  |
| <i>P</i> - value | 0.5152      | 0.8757      | 0.8484      | 0.9531        |

Values represent mean ± standard deviation.

Significant value = (\**P* < 0.05, \*\**P* < 0.01, \*\*\**P* < 0.001).

Values at the top represent without supplementation, while values below represent with supplementation.

## Discussion

The milk of the human breast is still a burden as studies are indicating its possibility of being contaminated by heavy metals such as lead (Pb), cadmium (Cd), and chromium (Cr). Breastmilk has been described as a course of transmission of cadmium, chromium, and lead from the mother's blood to an exclusively breastfed newborn.<sup>(14)</sup> Heavy metals thus appear to reach higher concentrations in the blood than in the breastmilk as observed in this study. The blood lead level was significantly higher in serum compared to breast milk. Lead levels are amassed in the bones of mothers over a long period and during pregnancy. It is released and increases from around 24 weeks of pregnancy until delivery because of an increase in bone turnover during this physiological period<sup>(15,16)</sup> into the blood and breast milk along with calcium. The mode of infant delivery could be a route of heavy metal exposure. The baseline milk lead level in this study is about sevenfold lower than a previous report in Northern Nigeria<sup>(17)</sup> and twofold lower than the study from Southwest Nigeria.<sup>(18)</sup> There were 57.0% of the subjects who delivered via the vagina in the study. A study from Iran reported that women who had vaginal delivery had more cadmium contamination than women who delivered through cesarean section.<sup>(19)</sup>

The use of supplements during pregnancy is recommended to women because they are important in the development of the unborn baby. There are varying reports concerning the use of supplements and their correlation with heavy metal levels. Gundacker C, *et al.*<sup>(20)</sup> reported a positive correlation between vitamin supplementation and milk metal levels. A study by Leotsinidis M, *et al.*<sup>(21)</sup> however found no effect of supplemental intake of trace elements and vitamins in women from Greece. Calcium supplemental intake during pregnancy might be the reason for the low serum cadmium levels as reported by other authors.<sup>(22)</sup> The intake of the iron supplement has been reported to be a significant negative predictor of chromium levels in the mother's blood<sup>(23)</sup> as observed in the study. Some studies have corroborated the theory that the absorption of iron from supplements may be inhibited by metals like chromium because they share and compete for the same protein binding sites and may be inversely correlated.<sup>(24,25)</sup> There are perhaps a few studies that have investigated the association between chromium exposure and pregnancy or lactation. A study reported

an association between lower excretion of chromium in maternal urine with higher consumption of multivitamins.<sup>(23)</sup>

Some physiological conditions like pregnancy and lactation raise chromium losses which would lead to chromium shortage particularly if chromium intakes are already low.<sup>(26)</sup> Maternal anemia has been linked with high breast milk lead levels.<sup>(2)</sup> Studies have also reported an association between iron deficiency and high lead levels in human beings.<sup>(27,28)</sup> This is however not the case in this study.

During reproductive age, women tend to suffer from vitamin D deficiency or insufficiency<sup>(29)</sup> even though the sun is the major source of vitamin D required by the body. Diets also provide small amounts of vitamin D<sub>3</sub> and Vitamin D<sub>2</sub> thus supplementation might be necessary. Vitamin D supplementation might also be essential during pregnancy first because maternal vitamin D insufficiency during this period is a common issue and a significant public health problem globally<sup>(30)</sup> and second because the fetus depends on the mother for vitamin D, calcium, and phosphorous which would be transmitted across the placenta. In this study, 36.7% of the subjects reported the use of vitamin D. There is a need for assessment of vitamin D supplementation and its use during pregnancy. There was a significant link between serum lead levels in the subjects who took vitamin D and in subjects who took vitamin B (53.3%) supplements. By implication, there is a need for continuous assessment of supplements and possible environmental pollutants that pregnant women could be exposed to. A synergistic link has been found between folic acid supplementation and serum zinc levels as reported in this study. Folic acid supplementation guards against a decline in serum zinc levels that could result from iron supplemental intake.<sup>(31)</sup> In this study, however, zinc levels were lower in subjects who took folic acid. The milk cadmium and zinc levels were lower in subjects who took the folic acid supplement. This may be because cadmium and zinc have the same oxidative states therefore cadmium can replace zinc present in metallothionein, thus preventing it from behaving as a free radical scavenger.<sup>(3)</sup> It is also worrisome that zinc levels were lower in breast milk because zinc plays a fundamental role in growth and development, and it is very essential for cellular metabolic processes.

There are several potential sources of environmental pollutants during pregnancy that could

predispose a mother to lead, chromium, or cadmium exposure. The use of eye kohl and lead could be a predisposing factor to lead exposure.<sup>(32)</sup> In this study, 60.0% of the subjects reported the use of eye kohl and lead cosmetics. None of the subjects reported cigarette use and one of the most well-identified routes of exposure to lead and cadmium is through cigarette smoking<sup>(33)</sup>, this did not apply to this study. The blood cadmium level was significantly correlated with hair coloration (hair dye) in the women that had their hair dyed for cosmetic purposes. Cadmium is one of the heavy metals present in hair dyes<sup>(34)</sup> and cadmium is absorbed and taken in by the body and can survive in all adult tissues. Serum zinc levels in subjects exposed to paints and radiation were significantly correlated. Zinc plays a significant role in ameliorating side effects caused by environmental pollutants.<sup>(35)</sup> Also, the fact that cadmium and zinc have similar oxidative states<sup>(3)</sup> were also exhibited in the milk of subjects exposed to paints and radiation. The significant relationship between milk zinc level and serum lead level represents an interactive association that can cause serum lead metal to increase when milk zinc level increases or vice versa.

The results of this study indicate that the use of eye kohl and hair dyes should be discouraged during pregnancy by health caregivers and further research into the use of supplements and the association with heavy metals is important. It is hoped that lactating women are encouraged to donate both blood and breastmilk to assist in routine studies such as this. The present recommendation for termination of breastfeeding is when blood lead levels of lactating mothers are at  $> 1.932 \mu\text{mol/L}$ . This is greater than the serum lead concentration of  $0.12 \pm 0.02 \mu\text{mol/L}$  observed in this study. The significant relationship between milk zinc level and serum lead level depicts a synergistic interaction that can cause serum lead to increase when milk zinc level increases or vice versa. The study however was able to recruit only 40 subjects. This is because several of the women who were approached concerning the study did not want to give both blood and breast milk for the study. Out of the 40 subjects, only 30 (75.0%) had complete data and these were included in the final statistical analysis. This is probably because the study needed to obtain both blood and milk from the mothers. This was also observed in a study carried out in Ibadan, Nigeria.<sup>(18)</sup> The study recruited 92 lactating women, 15 study subjects declined blood sample collection after

expressing breast milk, 15 subjects declined breast milk expression after blood sample collection, and 11 subjects declined both breastmilk expression and blood sample collection. Based on these findings, it can be concluded that in this part of the world, people's cultural beliefs, and environmental factors still affect their approach to research.

### Conclusion

The period of pregnancy and even lactation interact with the environment. The period of pregnancy is also monitored through antenatal and post-natal routines. The breastmilk of the mother is a suitable medium in human biomonitoring because it helps to expose any toxic metal exposure levels in the mother which may affect the baby. This study reveals that the use of supplements and cosmetics monitoring should be included in the antenatal and post-natal routines. It is also important that the blood and milk of the mother should be monitored regularly to reduce environmental exposure in the infant.

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### Conflicts of interest statement

All authors have completed and submitted the International Committee of Medical Journal Editors Uniform Disclosure Form for Potential Conflicts of Interest. None of the authors disclose any conflict of interest.

### Data sharing statement

The present review is based on the references cited. All data generated or analyzed during the present study are included in this published article and the citations herein. Further details, opinions, and interpretation are available from the corresponding author on reasonable request.

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