BLOOD LEAD LEVELS AMONG INFANTS AND CHILDREN WITH RICKETS AT KENYATTA NATIONAL HOSPITAL

BY

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This research dissertation is submitted in partial fulfillment for the degree of Masters of Medicine (Pediatrics and Child Health), University of Nairobi
DECLARATION

I declare that this dissertation is my original work and has not been submitted for the award of a degree in any other university.

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DEDICATION

This book is dedicated to Jack Barasa and Tanya A. Barasa- the most wonderful family.
ACKNOWLEDGEMENTS

First and foremost, I would like to thank God Almighty for providing me with an opportunity to study and for daily grace to handle all that comes my way.

I would also like to extend my sincere appreciation to the following:

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- All the children and their mothers/caregivers who participated in this study.
- My family members and friends for praying for me, being considerate and patient with me during the entire period of my study.
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<table>
<thead>
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<th>Abbreviation</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>KNH</td>
<td>Kenyatta National Hospital</td>
</tr>
<tr>
<td>BLL</td>
<td>Blood Lead Levels</td>
</tr>
<tr>
<td>WHO</td>
<td>World Health Organization</td>
</tr>
<tr>
<td>CDC</td>
<td>Centre for Disease Control</td>
</tr>
<tr>
<td>ATDSR</td>
<td>Agency for Toxic substances &amp; Drugs Research</td>
</tr>
<tr>
<td>GIT</td>
<td>Gastrointestinal Tract</td>
</tr>
<tr>
<td>PPM</td>
<td>parts per million</td>
</tr>
<tr>
<td>HB</td>
<td>Haemoglobin</td>
</tr>
<tr>
<td>HCT</td>
<td>Haematocrit</td>
</tr>
<tr>
<td>1, 25 D3</td>
<td>1, 25 dihydrocholecalciferol</td>
</tr>
<tr>
<td>μg</td>
<td>micrograms</td>
</tr>
<tr>
<td>FEP</td>
<td>free erythrocyte protoporphyrin</td>
</tr>
<tr>
<td>ZPP</td>
<td>zinc protoporphyrin</td>
</tr>
</tbody>
</table>
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ABSTRACT

BACKGROUND & OBJECTIVES:

Modern environmental health hazards including lead are becoming a public health issue of increasing concern. Lead, a toxic heavy metal accounts for 0.6 % of the global burden of disease. Centre for Disease Control (CDC) defines elevated blood lead levels as those above 10μg/dl. This is a public health action level, not a toxicity threshold since there are no safe blood lead levels. In Kenya, elevated blood lead levels have been found in significant proportions in both children and adults. Lead is known to interact with the metabolism of calcium and vitamin D at various levels in the body and lead toxicity is associated with low levels of both calcium and Vitamin D, the main causes of rickets in our set up. This study primarily aimed to determine the blood lead levels in infants and children with rickets and compare these to the levels of an age matched comparative group without rickets.

METHODOLOGY:

This was a hospital based descriptive cross sectional study with a comparative arm carried out at the Kenyatta National Hospital over a period of three months. Infants and children aged 3 months to 3 years who fit the case definition of rickets, and an age matched comparative group without rickets were recruited. A questionnaire was administered and a venous blood sample drawn to determine blood lead levels.

RESULTS:

A total of 100 cases were identified, 50 with rickets and 50 without rickets. The mean age was 9.7 months with 78% being infants. A male preponderance (68%) was noted among the children with rickets. Two percent of the cases with rickets had elevated lead levels compared to 0% in the comparative arm. The mean lead level was 2.2ug/dl in the rickets group and 2.5μg/dl in the comparative group. Having a diagnosis of rickets was not associated with elevated lead levels (p=1.000). However, low level lead exposure (≥2μg/dl), was noted in 68% of those with rickets and in 82% of the comparative group.
None of the child or socio-demographic factors investigated was found to be associated with elevated lead levels ≥10μg/dl.

**CONCLUSION:**

The prevalence of elevated lead levels in children with rickets was 2% while that in the comparative group was 0%.

Elevated lead levels were not found to be associated with rickets in this study.

Low levels of blood lead below CDC/WHO defined public health action threshold were found with 71% of the infants and children sampled having levels above ≥2μg/dl.

**RECOMMENDATION:**

Due to the serious adverse effects associated with lead toxicity even at low levels, this study justifies reinforcement of preventive measures and public health education to mitigate the effects of lead toxicity.
1.0 BACKGROUND & LITERATURE REVIEW

Environmental health hazards are physical, chemical or biological factors external to person with potential to adversely affect health. According to the WHO, a third of Africa’s disease burden can be attributed to environmental health hazards (1). Traditional health hazards such as lack of sanitation, hygiene and access to safe water have been the major contributors but with rapid urbanization and development over the last four decades in the continent, modern health hazards are on the rise as key contributors and are expected to supersede traditional health hazards (2). Lead is a heavy metal with no known physiological function that enters the environment as a result of numerous human activities. (3, 4) Lead and other heavy metals are some of the modern environmental health hazards (MEHHs) which result from rapid development with inadequate safeguards to the environment and health. (2) Lead toxicity accounts for 0.6% of global burden of disease. (3) Children under the age of 14 years constitute 42.9% of the Kenyan population (5) and are a unique subpopulation in regards to vulnerability to environmental health hazards and more specifically lead.

1.1 EPIDEMIOLOGY

Occupational and accidental lead poisoning are exceptional and relevant to public health, but it is environmental lead poisoning that is most worrisome.

Other than occupationally exposed individuals, children and pregnant women constitute the group that is most vulnerable to the toxic effects of lead, even at low levels. Lead levels in blood are reported to peak at around two years of age (4)

Children are a unique subpopulation in regards to lead poisoning for various reasons. They consume more water & food and inhale more air volume per body weight. Children also tend to engage in age appropriate hand to mouth activity and spend more time in one environment. This implies that if their environment is contaminated with lead, they are likely to absorb more lead from it than adults. They also have a higher proportion of the total body lead burden in blood which is the surrogate marker for neurotoxicity. Exposure to lead can occur transplacentally and via breastmilk therefore infants born to mothers with elevated blood lead levels get exposed to lead much earlier in life. (6, 7)
Lead poisoning in children in their home environment was first reported in the 1890s in Australia. After similar reports were documented in the west, nationwide periodic surveys have been conducted in some countries eg USA to assess the magnitude of the problem. Reports in these countries indicate a decline in the blood lead levels of children over the years. In Africa isolated surveys have documented lead levels way above the recommended CDC public health action level.

In Kenya, a study done in 2005 to evaluate occupational and environmental lead exposure in both children and adults found an overall prevalence of 15% (8). Seventy percent of pregnant women sampled in 1998 in 2 antenatal clinics within Nairobi had levels above the WHO cut off of 10μg/dl. (9) In Kibera, Nairobi, 7% of children between the age of 6 months to 59 months evaluated had elevated blood lead levels in 2007 (10). Around the Dandora dumpsite the levels were elevated in 50% of children between the ages of 4 to 18 years. (11)

Lin Lan et al evaluated 155 children aged of 6 months-59 months in China with a diagnosis of rickets for blood lead levels between 1999-2002. Eighty four percent of the children evaluated had blood lead levels above 10μg/dl-the CDC/WHO public action threshold (12). The average blood levels in children aged 14 years and below in China between 1994-2004 was 33.8%. (13)
Table 1: prevalence of elevated blood lead levels

<table>
<thead>
<tr>
<th>investigator</th>
<th>year</th>
<th>setting</th>
<th>sample size</th>
<th>% elevated</th>
</tr>
</thead>
<tbody>
<tr>
<td>Kimani et al(8)</td>
<td>2005</td>
<td>Nairobi/Olkalau</td>
<td>308-adults &amp; children</td>
<td>25%</td>
</tr>
<tr>
<td>Kimani et al(11)</td>
<td>2005</td>
<td>Dandora-Nairobi</td>
<td>328(4-18yrs)</td>
<td>50%</td>
</tr>
<tr>
<td>Olewe et al(10)</td>
<td>2007</td>
<td>Kibera-Nairobi</td>
<td>387(6-59 months)</td>
<td>7%</td>
</tr>
<tr>
<td>Tenge(14)</td>
<td>1996</td>
<td>Nairobi</td>
<td>475(6-17yrs)</td>
<td>0%</td>
</tr>
<tr>
<td>Owago et al(9)</td>
<td>1998</td>
<td>Nairobi</td>
<td>223(pregnant women)</td>
<td>70.4%</td>
</tr>
<tr>
<td>Boseela et al(15)</td>
<td>2002</td>
<td>Egypt</td>
<td>164(9-60 months)</td>
<td>26.2%</td>
</tr>
<tr>
<td>Lin lan et al(12)</td>
<td>1999-2002</td>
<td>China</td>
<td>155(6-59months)</td>
<td>84.6%</td>
</tr>
</tbody>
</table>

Despite the phasing out of leaded fuels, lead from other sources continues to be an important contributor to environmental pollution. This is shown by various studies done in different countries to evaluate blood lead levels after effective ban of leaded fuels as illustrated in table 2.

Table 2: prevalence of elevated blood lead levels after the phasing out of lead in fuels

<table>
<thead>
<tr>
<th>investigator</th>
<th>study year</th>
<th>phase-out year</th>
<th>location</th>
<th>sample size</th>
<th>% elevated</th>
</tr>
</thead>
<tbody>
<tr>
<td>Graber et al(16)</td>
<td>2009</td>
<td>2005</td>
<td>Kampala</td>
<td>163</td>
<td>20.5%</td>
</tr>
<tr>
<td>Riddel et al(17)</td>
<td>2003</td>
<td>2000</td>
<td>Phillipines(rural)</td>
<td>2261</td>
<td>21%</td>
</tr>
<tr>
<td>Nichani et al(18)</td>
<td>2002</td>
<td>2000</td>
<td>India</td>
<td>754</td>
<td>33.2%</td>
</tr>
</tbody>
</table>
Evaluation of lead levels in soil, water, milk, vegetables and dust in different areas in Kenya show dangerously high levels of lead as shown in table 3.

Table 3: concentration of lead in the environment in Kenya

<table>
<thead>
<tr>
<th>site</th>
<th>soil μg/dl</th>
<th>milk μg/dl</th>
<th>vegetables μg/dl</th>
<th>air μg/dl</th>
<th>water μg/l</th>
</tr>
</thead>
<tbody>
<tr>
<td>Kibera(10)</td>
<td>3,000-90,000</td>
<td>-</td>
<td>0</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Thika (19)</td>
<td>133,790</td>
<td>44</td>
<td>2243</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Nairobi(19)</td>
<td>265,918</td>
<td>46</td>
<td>5053</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Nairobi-Dandora(11)</td>
<td>6.4-1350</td>
<td>-</td>
<td>-</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Metrological Dept-Nairobi(20)</td>
<td></td>
<td></td>
<td>2.9μg/m3</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Kisumu(21)</td>
<td>-</td>
<td></td>
<td></td>
<td></td>
<td>140-650</td>
</tr>
<tr>
<td>WHO upper limit</td>
<td>110μg/dl</td>
<td>20μg/dl</td>
<td>300μg/dl</td>
<td>1.5μg/m3</td>
<td>10μg/l</td>
</tr>
</tbody>
</table>

Lead contamination of household consumer products and house dust has been found in South Africa & Nigeria (2) as shown in table 4.

Table 4: selected levels of lead in consumable products in Africa.

<table>
<thead>
<tr>
<th>product</th>
<th>Levels ppm</th>
<th>country</th>
<th>Recommended (ppm)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Residential paint</td>
<td>290,000</td>
<td>South Africa(2005)</td>
<td>600</td>
</tr>
<tr>
<td>Paint on toys</td>
<td>145,000</td>
<td>South Africa(2007)</td>
<td>600</td>
</tr>
<tr>
<td>Household paint</td>
<td>189,000</td>
<td>Nigeria(2007)</td>
<td>600</td>
</tr>
<tr>
<td>crayons</td>
<td>8200-10650</td>
<td>South Africa(2004)</td>
<td>0.1</td>
</tr>
<tr>
<td>pencils</td>
<td>79-1160</td>
<td>South Africa(2004)</td>
<td>0.1</td>
</tr>
</tbody>
</table>
1.2 EXPOSURE

Lead is absorbed into the human body through the gastrointestinal system, the respiratory system and very rarely through the skin. (3, 4, 22) The placenta and breast milk can also serve as routes of exposure to the foetus and the breast fed child. (23, 24, 25)

Children may be exposed to lead through inhalation of lead particles suspended in the air. This lead could be from fumes of burning car batteries, industrial and automotive exhausts or open burning of industrial and domestic waste containing plastics and other contaminated material. Prior to the phasing out of lead from fuels, this was the main route of exposure. Absorption of lead through the respiratory tract depends on the particle size. Ninety percent of lead that is deposited in the lower airways is absorbed.

Ingestion of lead is currently the main route of exposure for children and infants. This may occur through ingestion of flaky paint, contaminated soil and dust, herbal drugs containing lead and lead compounds, consumption of food grown in contaminated soil, cooked or stored in lead utensils and use of water contaminated with lead. (2) Absorption of lead through the gastrointestinal system varies with age. Children absorb 50% of ingested lead as compared to 10% in adults. (2, 3, 4, 26) Absorption also varies with diet and micronutrient deficiencies. More lead will be absorbed in the fasted state and zinc, calcium and iron deficiencies increase lead absorption. (22, 27, 28) Experimental studies show a four to eight fold increase in absorption of lead when calcium deficient diets are ingested. (28) Calcium supplementation has been suggested and investigated as one of the ways of reducing lead absorption from the GIT tract. (29, 30) Consumption of calcium deficient diets has been described as an important contributor to development of rickets.

Dermal absorption of lead is not a significant mode of lead uptake unless the lead is present in its more lipid soluble organic form. (4)
1.3 DISTRIBUTION & EXCRETION.

Figure 1: distribution & excretion of lead in the body

**LEAD IN BLOOD**

- found bound to red blood cells.
- Half life is one to two months.
- redistributed to other tissues.
- the surrogate marker for toxicity

**Lead in Bone**

- Contains the bulk of the body lead, (74% in children).
- Half life is 30 years.
- Can be mobilized during pregnancy & lactation thereby increasing the blood lead levels.

**Lead in soft tissue**

Has a predilection for nervous tissue and the proximal tubules of the kidney.

**Excretion**

- Excreted in urine
- Significant amounts found in breast milk.
- Can also be excreted in sweat.
- Transplacental transfer occurs.
1.4 HEALTH EFFECTS OF LEAD

1.41 MECHANISM OF TOXICITY

The toxic properties of lead can be attributed to two main mechanisms.

- Lead’s ability to distort structural proteins and enzymes. Lead is a divalent cation that binds strongly to sulfhydryl groups on proteins leading to alteration of the function of the protein. This is the mechanism by which it causes haemolytic anaemia by inhibiting the function of D-ALA, an important enzyme in heme synthesis.

- Lead has the ability to mimic or inhibit calcium activity. It inhibits the entry of calcium into cells even when present in low concentrations. Many calcium-binding proteins have a higher affinity for lead than for calcium. Lead bound to these proteins may alter function, resulting in abnormal intra- and intercellular signaling. Neurotransmitter release is, in part, a calcium-dependent process that is adversely affected by lead.

- A third mechanism prevents the development of the normal tertiary brain structure. In immature mammals the normal neuronal pruning process that results in elimination of multiple intercellular brain connections is inhibited by lead. Failure to construct the appropriate tertiary brain structure during the first few years of life may result in a permanent abnormality.

1.42 CLINICAL PRESENTATION

Lead poisoning can be acute or chronic. Acute poisoning is rare & fatal.

**GI symptoms** include anorexia, abdominal pain, vomiting, and constipation, often occurring and recurring over a period of weeks. Children with BLLs >20μg/dL are twice as likely to have GI complaints as those with lower BLLs.

**CNS symptoms** are related to increasing cerebral edema and increased intracranial pressure. Headaches, change in mentation, lethargy, papilloedema, seizures, and coma
leading to death are rarely seen at levels <100μg/dL but have been reported in children with a BLL as low as 70μg/dL.

Behaviour disturbances e.g autism and intellectual impairment are noted even at very low blood lead levels. There is no clear cutoff blood lead level for the appearance of hyperactivity, but it is more likely to be observed in children who have levels >20μg/dL.

Renal tubular dysfunction and a reversible Fanconi syndrome may be induced by lead toxicity.

Lead is known to interfere with enzyme activity in the human body. Inhibition of D-ALA, an important enzyme in heme synthesis leads to shortened red cell survival and may contribute to a hemolytic anemia, though most cases of anemia in lead-poisoned children are due to other factors such as iron deficiency and hemoglobinopathies.

### 1.43 LEAD & RICKETS

Rickets is a disease of growing bone characterized by inadequate mineralization due either isolated or combined deficiencies of calcium, phosphates and vitamin D. There is reported resurgence of vitamin D deficiency and rickets worldwide (31). In Kenya, there is anecdotal evidence of increasing prevalence of rickets.

**LEAD and VITAMIN D**

Lead toxicity is associated with a decrease in circulating forms and function of 1, 25 dihydrocholecalciferol, the active form of vitamin D.

Lead interferes with hydroxylation of 25 hydrocholecalciferol by the 1α hydroxylase in the proximal tubules of the kidney. It is also postulated that lead increases the breakdown of 1,25 D3 thereby leading to reduced circulatory levels of the active D3. This is mainly seen in children with high lead levels above 60ug/dl, but depressed levels of vitamin D are seen even at lower levels. Levels of vitamin D comparable to those seen with children diagnosed with renal failure have been observed in children with lead toxicity. (32)
At the receptor level, vitamin D usually stimulates synthesis of osteocalcin, a protein constituent of bone that’s important for mineralization, this effect is inhibited by lead. (33) Lead competitively inhibits the vitamin D stimulated absorption of calcium in the intestines. The effects of lead on vitamin D are more apparent in children with chronic nutritional deficiency of calcium, zinc or Iron.

**LEAD & CALCIUM**

Lead and calcium are both absorbed at the small gut. Calcium competitively inhibits lead absorption at this site. Low calcium levels in the diet, the reported main cause of rickets in the developing world (36), are noted to increase the absorption of lead up to 4-8 fold. (34)

Calcium deficiency in experimental studies also increases the susceptibility of lead toxicity. It has also been noted that in calcium deficiency there is an increase in the bone lead content which can then act as an endogenous source of blood lead levels in times of physiological stress. Lead toxicity on the other hand causes an increase in calcium loss from the proximal convoluted tubules of the kidney and may further exacerbate the hypocalcaemia. (33)

**1.5 EVALUATION OF LEAD TOXICITY**

A large proportion of lead in the body is contained in the bone- approximately 74% in children. Therefore the most accurate assessment of the total body lead burden is the bone lead levels. (4) However, blood lead levels have been accepted as the best surrogate of lead toxicity as they represent the lead available to cause neurotoxicity. It is important to note that there are no safe blood lead levels. In 1991 US CDC established 10μg/dl as the lowest level of concern in children. This is an action threshold used as a screening tool and is not a toxicity threshold. This advisory level has been continually reduced from 60μg/dl (1960-1970) to 30μg/dl (1971-1985) to 25μg/dl (1985-1991). There is ongoing deliberations within the Advisory Committee on Childhood Lead Poisoning &Prevention (ACCLPP) on the possibility of further reducing the threshold after adverse effects and even increased mortality in adults were clearly demonstrated in those with levels above 2μg/dl. (37, 38, 39, 40)
STUDY JUSTIFICATION.

Lead is now known to be toxic at levels below the CDC/WHO action of 10μg/dl. Community prevalence studies in our set up continue to show that lead levels in blood are still significantly high even after the elimination of lead in fuel. In Kenya, there is anecdotal evidence of increasing cases of rickets in children. Lead toxicity alters the metabolism of Vitamin D, reducing its levels and may be a cause of rickets. It has also been known to affect the response to treatment of Vitamin D deficient rickets. Children who are fed low calcium diets and those who have hypocalcaemia have higher body lead burdens and also have the potential for more toxicity. Lead toxicity may have an impact on the clinical presentation of rickets.

UTILITY

This study will raise awareness on the possibility of lead toxicity in children diagnosed with rickets. The results may justify screening of blood lead levels in children with rickets. It will also give information that could justify further studies to evaluate what effects lead toxicity may have on the response to treatment of rickets in our set up.
STUDY OBJECTIVES

Primary

1. To determine the proportion of children diagnosed with rickets in KNH who have elevated blood lead levels and compare the proportion to that of an age matched group without rickets

Secondary

1. To identify the child and socio-demographic factors associated with elevated blood lead levels e.g. age, sex, residence.
2.0 METHODOLOGY

2.1 Study Design

This was a hospital based descriptive cross sectional study with a comparative arm

2.2 Study Location

Patients were consecutively recruited from the Kenyatta National Hospital pediatric wards, Pediatric Outpatient clinic and the Pediatric Emergency Unit. Kenyatta National Hospital is the main national referral teaching and research hospital in Kenya, located in the capital city, Nairobi. It also serves the population of Nairobi and its environs as a Provincial Hospital with a catchment population of two million people.

2.3 Study Period

The study was carried out over a period of three months.

2.4 Study Population

The study population included:

- Children and infants 3 months to 3 years diagnosed with rickets.
- Age matched comparative group from the pediatric eye and/or surgical clinic.

2.5 Selection and Enrollment

2.51 Inclusion Criteria

1. Children and infants aged 3 months to 3 years, recruited at diagnosis or within 1 month of diagnosis of rickets.

2. Children and infants of parents who gave informed consent.

2.52 Exclusion Criteria

1. Children and infants with a previous diagnosis of lead toxicity.
2. Children and infants of parents or caregivers who declined to give informed consent.
2.53 Case Definition Of Rickets

Rickets was defined by presence of clinical signs and confirmatory radiological findings. Clinical Rickets was diagnosed if two or more of the following were present; craniotabes, frontal bossing, bilateral widening of the wrists, rachitic rosary, Harrison’s sulcus and pigeon’s chest. (41)

Radiologic evidence of rickets was said to be present if any of the following features were confirmed by a radiologist: generalized osteopenia, fraying and cupping of distal ends of radius & ulna and widening of the costochondral junctions. (42)

2.54 Comparative Group

Comparative age matched group was recruited from the Paediatric Eye Clinic and Ear Nose & Throat (ENT) clinic and was examined for clinical signs of rickets prior to enrollment.

2.6 Sample Size

The sample size was determined by using the Two Sample Comparison of Proportions formula shown below.

FORMULA

\[ N = \frac{1}{P_1 \cdot P_2} \left[ P_1 (1-P_1) + P_2 (1-P_2) \right] \]

- \( P_1 = 7\% \) based on the Kibera study (10)
- \( P_2 = \) estimated to be 35\% based on the reported 4-8 fold increase in lead absorption when calcium deficient diets are administered. (28)
- Power \( (1-\beta) = 0.9 \)
- \( \alpha = 0.05 \)
- Ratio of sample sizes = 1:1

\[ N = 100 \]
2.7 DATA COLLECTION, MANAGEMENT & ANALYSIS

2.71 Patient Recruitment/ Questionnaire administration

Potential Study Participants were identified by screening patient files and/or admission records. The principal Investigator identified those patients with rickets who meet the eligibility criteria. The Investigator/ assistants then approached the patients/ caregivers and explained the purpose and methods of the study allowing the parent/ caregiver to provide voluntary and informed consent.

Consent was given in written form, on a pre-designed consent forms which were availed to the caregiver. The consent form provided described the purpose of the study, the study procedure to be followed as well as the potential benefits and risks of participating in the study. The investigator then conducted the consent discussion and ensured the parent/guardian understood the information provided on the consent form. Questions regarding the study from the parent/guardian were answered prior to signing the consent form. Consent obtained was voluntary and free from coercion.

Parents/ guardians accepting to take part in the study signed the consent form which was countersigned by the investigator. Records were kept regarding reasons for non-participation of eligible participants.

A pretested questionnaire was administered to the consenting parents or caregivers. Subjects were recruited via consecutive sampling till the desired sample size was attained.

2.72 Blood sample acquisition

The venepuncture site was swabbed with alcohol and left to air dry. 3-5mls of venous blood were collected directly into lead free (EDTA–containing) vacutainer tubes in a sterile manner by a qualified phlebotomist. Each tube was inverted several times to mix the EDTA and the blood so as to prevent coagulation. Samples were stored at about 4 degrees Celsius and sent for laboratory analysis within 12 hours. Samples were prepared for analysis using standard methods (vanLoon, 1985; NIOSH 1977)
Blood lead measurements on digested sample solutions were performed using atomic absorption spectrometer (AAS-Spectr-AA-10, BARIAN). Water used was distilled-deionised. All the glassware used in this study was decontaminated by soaking overnight in 5% HNO3 and rinsed thoroughly in deionised water. The lead standard solutions were freshly prepared daily, and checked for constancy of absorption before taking the readings. Standard and blank samples were analysed for every 15 samples analysed to ensure adequate quality control.

2.73 Data Analysis & Management

The data was collected using a structured questionnaire and results of blood lead levels. A database was designed in MS Access. On completion of the data entry exercise, the data was exported in a SPSS Version 17.0 for analysis.

Data was summarised using means and medians for continuous variables such as age and lead levels. Categorical variables such as sex, age group, injuries and related factors were summarised by calculating the proportions. The proportions were calculated within 95% confidence intervals and means, with standard deviations or medians with inter-quartile ranges, derived as appropriate to provide descriptive summaries of the data.

Results were presented in descriptive form using frequency tables, pie charts, graphs and cross tabulation.

The association between various socio-demographic variables and elevated lead levels were tested independently using chi-square and fisher’s exact test whenever a category had less than five subjects. Odds ratios and chi-square test for linear trend were used to determine significance.

The main study outcome was calculated as the percentage of children with elevated lead levels in the two study groups (children with and without rickets). A statistical test for comparison of two proportions was conducted to determine if the proportions of children with main outcome in the study groups were statistically different. Within each group the chi square test was used to identify patient& demographic factors showing
significant association with elevated lead levels. In case of small value a Fisher’s exact test was conducted.
2.9 ETHICAL CONSIDERATIONS

1. The study was undertaken after approval by the Department of Paediatrics, University of Nairobi and the Ethical review committee, Kenyatta National Hospital.

2. The purpose of the study was carefully explained to the Children’s parents or caregivers and a written consent obtained prior to enrolling any child in the study.

3. Strict Confidentiality was observed throughout the entire study period. The study participants were given study identification numbers which were used as the linking identifier for clinical & laboratory databases. Access to study data for reasons other than those specified in this application will not be permitted without further application to the KNH Scientific and Ethics committee.

4. Clinically important findings and laboratory results were availed to the teams managing the children.

5. The benefits for the patients who participated in the study was that they had a thorough assessment (history taking and physical examination) as well as a blood test to determine the lead levels and appropriate referral according to the findings.
3.0 RESULTS

Figure 2: flow chart on screening and recruitment of study patients

56 subjects with clinical signs of rickets & confirmatory radiological features identified

informed consent from parents/caregivers sought

5 declined

Questionnaires administered to 50 parents/caregivers

venous blood samples drawn from 50 infants and children

58 age- matched comparative subjects identified.

5 excluded based on presence of signs of rickets

rickets ruled out on basis of negative clinical signs

Informed consent sought

3 declined

Questionnaires administered to 50 parents/caregivers

Venous blood samples to determine lead levels drawn from the subjects
3.1 SOCIO DEMOGRAPHIC CHARACTERISTICS OF STUDY POPULATION

The median age of the children and infants recruited was 9.1 months, (IQR 5-11 months). 78% of the study population was below one year of age. There were 68% males in the group with rickets as compared to 44% in comparative group (p value 0.016).

FIGURE 3: age distribution of the study population

There were significant differences noted in maternal age, maternal parity and the birth intervals between the two groups. Mothers of children with rickets were older, with more children but longer birth intervals than their counterparts from the comparative arm. There were no differences in maternal level of education as shown in table 5.
Table 5: maternal characteristics of the study population

<table>
<thead>
<tr>
<th></th>
<th>rickets(SD)</th>
<th>comparative(SD)</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mean maternal age(years)</td>
<td>27.1(5.7)</td>
<td>25.8(4.6)</td>
<td>0.007</td>
</tr>
<tr>
<td>Maternal parity</td>
<td>2.1(1.1)</td>
<td>1.7(0.8)</td>
<td>0.019</td>
</tr>
<tr>
<td>Birth interval(years)</td>
<td>4.9(2.9)</td>
<td>3.2(1.5)</td>
<td>0.019</td>
</tr>
<tr>
<td>Maternal level of education</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>none</td>
<td>2(4%)</td>
<td>1(2%)</td>
<td>1.000</td>
</tr>
<tr>
<td>primary</td>
<td>14(28%)</td>
<td>19(38%)</td>
<td>0.388</td>
</tr>
<tr>
<td>secondary</td>
<td>28(56%)</td>
<td>25(50%)</td>
<td>0.644</td>
</tr>
<tr>
<td>Post secondary</td>
<td>6(12%)</td>
<td>3(6%)</td>
<td>1.000</td>
</tr>
</tbody>
</table>

There were no differences noted in the two groups in terms of gestation at birth, but there was a trend towards significance in birth weight as illustrated below.

Table 6: child characteristics of study population

<table>
<thead>
<tr>
<th></th>
<th>rickets</th>
<th>comparative</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mean birth weight(kg)</td>
<td>3.0</td>
<td>3.1</td>
<td>0.065</td>
</tr>
<tr>
<td>Gestation at birth (months)</td>
<td>8.9</td>
<td>8.9</td>
<td>0.73</td>
</tr>
</tbody>
</table>

Thirty seven (74%) of the subjects with rickets lived in urban areas as compared to 43 (86%) of the comparative group [OR (95%CI), 1.7(0.3-11] p=0.554. However, the group
with rickets was more likely to live within a km of a major highway than their counterparts without rickets [OR (95% CI), 4.5(1.9-10.8) p<0.0001.

In terms of housing, 44 (88%) of the subjects with rickets lived in stone walled houses in comparison to 46 (92%) of the comparative group [OR 1.05 p=0.379]. The walls of the houses the majority lived in were painted with 70% in the rickets group as compared to 72% of the comparative group with no differences between the two groups.

The subjects with rickets were 3 times more likely to have the paint on the walls chipping [OR (95% CI), 3(1.1-9.0) p<0.016] than the counterparts without rickets.

The summary of the other socio-demographic characteristics is as shown in table 7

**Table 7: socio-demographic characteristics of study population**

<table>
<thead>
<tr>
<th>variable</th>
<th>rickets N=50 (%)</th>
<th>comparative group N=50(%)</th>
<th>OR</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Source of water</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Municipal</td>
<td>42(84%)</td>
<td>38(76)</td>
<td>1.10</td>
<td>0.354</td>
</tr>
<tr>
<td>Borehole</td>
<td>6(12)</td>
<td>11(22)</td>
<td>0.50</td>
<td>0.244</td>
</tr>
<tr>
<td>others</td>
<td>2(4)</td>
<td>1(2)</td>
<td>0.45</td>
<td>0.344</td>
</tr>
<tr>
<td>Water storage containers</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>plastic</td>
<td>47(94)</td>
<td>47(94)</td>
<td>1.1</td>
<td>0.367</td>
</tr>
<tr>
<td>others</td>
<td>3(6)</td>
<td>3(6)</td>
<td>1.0</td>
<td>0.323</td>
</tr>
</tbody>
</table>

Various factors that affect lead exposure and or lead levels eg excessive pica activity, proximity to a dumpsite etc were compared between the two groups. The results are shown in figure 4.
Figure 4: comparison of factors affecting lead levels.

**Chart Title**

<table>
<thead>
<tr>
<th>Factor</th>
<th>Number with Factor</th>
<th>p-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Breasfeeding</td>
<td>40</td>
<td>0.006</td>
</tr>
<tr>
<td>Vitamin/mineral supplement</td>
<td>22</td>
<td>0.002</td>
</tr>
<tr>
<td>Sunlight exposure</td>
<td>36</td>
<td>0.001</td>
</tr>
<tr>
<td>Pica activity</td>
<td>4</td>
<td>0.006</td>
</tr>
<tr>
<td>Dumpsite proximity</td>
<td>14</td>
<td>0.822</td>
</tr>
</tbody>
</table>

ASSESSMENT FOR RICKETS

Patients met the eligibility criteria for rickets if they had 2 more of the predetermined clinical signs and confirmatory findings on a wrist radiograph as reported by a qualified radiologist.

Widening of the wrists was the commonest clinical sign identified in 88% (n=44) of the subjects with rickets, followed by rachitic rosary in 70% (n=35). Craniotabes, frontal bossing, and harrisons sulcus were identified in 28%, 46% and 14% respectively of the subjects sampled. [Figure 5]
Radiological findings were as summarized below.

**Table 8: Radiological findings of the subjects with rickets**

<table>
<thead>
<tr>
<th>characteristic</th>
<th>N (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Osteopenia</td>
<td>30 (60%)</td>
</tr>
<tr>
<td>Fraying &amp; cupping</td>
<td>47 (94%)</td>
</tr>
<tr>
<td>fractures</td>
<td>1 (2%)</td>
</tr>
</tbody>
</table>

The mean serum calcium was 2.0mmol (SD 1.5-2.2), while the mean serum phosphates was 1.6mmol (SD 1.3-1.8). Both the mean serum calcium and phosphates were within the normal ranges. 54% of the subjects with rickets had a mean calcium level below 2.2 mmol. The mean alkaline phosphatase level however, was above the normal range at 492IU (IQR 221-655).
3.2 LEAD LEVELS

3.21 Lead levels using the CDC/WHO defined 10μg/dl

The prevalence of elevated lead levels was 2% in the group with rickets compared to 0% in the comparative group (p value 0.479). The mean lead levels for the entire group was 2.2μg/dl (SD 2.28). The mean lead levels among the children with rickets were 2.2μg/dl (SD 2.4) and 2.46μg/dl (SD 1.09) in the comparative group with no differences between the two groups (p value 0.516).

3.22 Lead levels using ≥2μg/dl cut off

When the elevated lead level threshold was lowered to 2μg/dl, the lowest level currently known to be associated with adverse effects (37, 38, 39, 40), 71% of all sampled children had a level ≥2μg/dl. The comparative group had 82% of the subjects with levels ≥2μg/dl as compared to 68% in the rickets group with a trend towards a significant difference (p value 0.061).[Table 9]

Table 9: Results of blood lead levels

<table>
<thead>
<tr>
<th>Variable</th>
<th>Rickets</th>
<th>Comparative group</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mean lead levels (μg/dl)</td>
<td>2.2</td>
<td>2.5</td>
<td>0.516</td>
</tr>
<tr>
<td>&gt;10μg/dl</td>
<td>1(2%)</td>
<td>0(0%)</td>
<td>0.479</td>
</tr>
<tr>
<td>≥2μg/dl</td>
<td>30(60%)</td>
<td>41(82%)</td>
<td>0.061</td>
</tr>
</tbody>
</table>

3.3 Characteristics of those with elevated lead levels

There was one 8 month old male subject with elevated lead level at a level of 17μg/dl. He was born at 9 months gestation, weighing 2.8kgs and lived in the rural area, more
than 1km from a major highway and had no proximity to a dumpsite. The floor of the house he lived in was earthen and the walls were wooden and not painted. The water used in the household was sourced from a borehole and stored in plastic containers.

His mother was 30 years old with 3 children and had attained secondary school education. Both his parents were farmers.

This subject had rickets, had never received a mineral/vitamin supplement but was exposed to the sun daily for 1-2 hours. At the time of the study, he was breastfeeding and did not exhibit excessive pica activity.

3.4 Child and socio-demographic co-relates of elevated blood lead levels

We sought to determine child, maternal and socio demographic characteristics associated with elevated lead levels, ≥10μg/dl, which were found in only 1 subject. None of the child, maternal or socio-demographic characteristics assessed were associated with elevated blood lead levels, ≥10μg/dl. The characteristics assessed included age, gestation at birth, vitamin/mineral supplementation, residence, excessive pica activity, diagnosis of rickets etc.

3.5 Child & socio demographic correlates of levels≥ 2μg/dl

We sought to determine if any differences in child, maternal and socio-demographic characteristic existed if the postulated new definition of elevated blood lead level of ≥2μg/dl was used (37). Subjects who were breastfeeding at the time of the study and those who had rickets were less likely to have levels ≥2μg/dl. Excessive pica activity was associated with levels ≥2μg/dl. There was a trend towards significance for sunlight exposure being associated with levels above 2μg/dl. [Table 10]
### Table 10: Characteristics associated with Lead levels ≥2μg/dl

<table>
<thead>
<tr>
<th>Characteristic</th>
<th>Lead ≥2ug/dl (N=71, %)</th>
<th>Lead &lt;2ug/dl (N=29, %)</th>
<th>OR((95%CI))</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td>breastfeeding</td>
<td>42 (59.2)</td>
<td>25 (86.2)</td>
<td>0.2 (0.1-0.7)</td>
<td>0.007</td>
</tr>
<tr>
<td>Vitamin/mineral supplement</td>
<td>20 (28.2)</td>
<td>10 (34.5)</td>
<td>0.7 (0.3-1.9)</td>
<td>0.269</td>
</tr>
<tr>
<td>Sunlight exposure</td>
<td>60 (84.5)</td>
<td>18 (62.1)</td>
<td>1.9 (0.5-7.4)</td>
<td>0.095</td>
</tr>
<tr>
<td>Excessive pica activity</td>
<td>17 (24.3)</td>
<td>2 (6.9)</td>
<td>4.3 (0.9-20.1)</td>
<td>0.053</td>
</tr>
<tr>
<td>Dumpsite proximity</td>
<td>22 (31.1)</td>
<td>5 (17.2)</td>
<td>2.2 (0.7-6.4)</td>
<td>0.122</td>
</tr>
<tr>
<td>Painted walls</td>
<td>48 (67.6)</td>
<td>23 (79.3)</td>
<td>0.5 (0.2-1.5)</td>
<td>0.177</td>
</tr>
<tr>
<td>Urban residence</td>
<td>56 (78.9)</td>
<td>24 (82)</td>
<td>0.6 (0.1-2.2)</td>
<td>0.342</td>
</tr>
<tr>
<td>rickets</td>
<td>30 (42.2)</td>
<td>20 (68.9)</td>
<td>0.3 (0.1-0.8)</td>
<td>0.008</td>
</tr>
</tbody>
</table>

Results of multivariate regression analysis showed that sunlight exposure was the most significant factor associated with levels ≥ 2μg/dl. There was a trend towards significance for the protective effect of breastfeeding against levels ≥2μg/dl. [Table 11]
Table 11: Multivariate Regression analysis of characteristics associated with levels ≥2μg/dl

<table>
<thead>
<tr>
<th>Variable</th>
<th>OR(95% CI)</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Currently breastfeeding</td>
<td>0.3 (0.1-1.0)</td>
<td>0.057</td>
</tr>
<tr>
<td>Male gender</td>
<td>1.1 (0.40-3.20)</td>
<td>0.845</td>
</tr>
<tr>
<td>Within 1km of major highway</td>
<td>2.02(0.63-6.50)</td>
<td>0.236</td>
</tr>
<tr>
<td>Painted house</td>
<td>0.5(0.2-1.6)</td>
<td>0.223</td>
</tr>
<tr>
<td>Sun exposure</td>
<td>4.5(1.02-20.35)</td>
<td>0.045</td>
</tr>
<tr>
<td>Vitamin/mineral supplementation</td>
<td>1.1 (0.4-3.2)</td>
<td>0.924</td>
</tr>
<tr>
<td>Rickets</td>
<td>0.38 (0.1-1.2)</td>
<td>0.104</td>
</tr>
<tr>
<td>Excessive pica activity</td>
<td>2.5 (0.5-13.7)</td>
<td>0.283</td>
</tr>
</tbody>
</table>
DISCUSSION

In this study we found a low prevalence (1%) of elevated lead levels using the WHO/CDC definition of ≥10μg/dl for the entire study population. Two percent of the subjects with rickets had elevated lead levels compared with 0% in the comparative group. The subjects with rickets did not have a higher prevalence of elevated lead levels (p value=0.479).

Majority of the recruited subjects were infants (78%) and also resided in urban areas (70%) with no significant differences between the group with rickets and the one without.

Some significant differences were noted between the subjects with rickets and the comparative group with regards to maternal age, maternal parity and birth intervals. The mothers’ of children with rickets were older and had more children than their counterparts in the comparative arm. This is probably because depletion of maternal calcium and Vitamin D - possible causes of rickets, that are found more commonly in older multiparous women with short birth intervals. (43). Another possible explanation for the differences noted in two of the maternal characteristics (higher parity and shorter birth intervals) would be that the subjects with rickets were all hospitalised, probably reflecting the severest forms of rickets or rickets with comorbidities compared to the comparative group that consisted of outpatients.

The prevalence of elevated lead levels in this study was 1% and the mean blood lead level was 2.2μg/dl. These values are much lower than what has been previously defined in Kenya but comparable to that of the US population ten years ago demonstrated to be at 1.6% for levels ≥10μg/dl, with mean blood levels at 2.1μg/dl during the National Survey of children aged 1-5 years in 1999-2000(44). A prevalence of 25% was defined in 2005 among children and adults by Njoroge et al (8) and in 2007, 7% of children aged 6-59 months had levels ≥10μg/dl in a study conducted by Olewe et al in Nairobi (10).These figures demonstrate an obvious downward trend probably reflective of reduced environmental lead contamination since the phasing out of leaded fuels in Kenya in 2005. Some significant differences in terms of age of the subjects described in
these two studies may also account for the low prevalence found in our study. The study in 2005 constituted of predominantly adults with children below 14 years accounting for 12%, 14% of the subjects in the study by Olewe et al were below one year while in our study, majority of the subjects (78%) were infants. Few studies have been done to examine lead level patterns in children with levels below 10μg/dl, but longitudinal studies in those with higher levels show peaks at 18-36 months of age (45).

Similar trends in reduction of blood lead levels after phasing out of lead in fuels, however, have been noted in India (46), South Africa (47) and USA (48).

One subject (1% of the sampled population) had lead levels above 10μg/dl, the CDC/WHO defined elevated blood lead level. This was an 8 month old infant with rickets who resided in the rural area, had excessive pica activity and levels of 17ug/dl. None of the subjects in the comparative group were found to have levels ≥10μg/dl.

The prevalence amongst subjects with rickets was much lower (2%) than that found in Guangzhou, China by Lin Lan et al between 1999-2002 at 84.6% among children with rickets aged 6 months to 59 months (12). The study in China recruited 155 outpatients, 22.6% of the subjects had subclinical rickets and the age distribution was not availed to determine how many were infants. The study was carried out in an inner city that was highly industrialized and part of the study period included time before the phasing out of lead in fuels in China which was completed in 2000. In our study, 70% of the population resided in urban areas, none of the children had subclinical rickets and it was carried out 7 years after the phasing out of lead in fuels in Kenya which was completed in 2005.

None of the child, maternal and socio- demographic characteristics investigated was found to be significantly associated with elevated blood lead levels as defined by CDC/WHO. This could have been due to the fact that only one subject was found to have levels ≥10μg/dl making it difficult to make statistical inferences.

An interesting observation was that some significant differences in child and socio-demographic characteristics were realized when the threshold of high lead levels was lowered to ≥2μg/dl, the lowest level currently known to be associated with adverse effects.(37, 38, 39)
The comparative group had more children with levels ≥2μg/dl (82%) than their counterparts with rickets (68%) with a trend towards significance (p=0.061). However, this is likely to be due to the confounding effects of excessive pica activity, breastfeeding and sunlight exposure.

Excessive pica activity, a known risk factor for lead accumulation was found to be more common among the children in the comparative group (15%) than the group with rickets (4%) [p=0.006]. There was a trend towards significance for association of excessive pica activity with levels ≥2μg/dl [OR 4.3 (0.9-20.1) p=0.053]. The comparative group was probably reflective of active well children with appropriate attainment of milestones who were more likely to engage in hand to mouth activities than their counterparts who had rickets therefore being exposed to more lead through the GIT system.

Breastfeeding was found to be protective against levels ≥ 2μg/dl [OR 0.2 (0.1-0.7) p=0.007]. The group with rickets was more likely to be breastfeeding (80%) than the comparative arm (54%) [p=0.006]. Considering that the majority of the subjects were infants, those who are predominantly breastfed may tend to consume less volumes of other foods eg cow’s milk and vegetables which have been demonstrated to have significant amounts of lead in some studies (19).

Exposure to sunlight which was found in a higher proportion of the comparative arm (96%) than the rickets arm (72%) [p value=0.001] and was trending towards significance for association with levels ≥2μg/dl [p value=0.095]. After the multivariate regression analysis, sunlight exposure was significantly associated with levels ≥2μg/dl (p value 0.045). Subjects who are sunlight exposed probably play outside or spend a significant amount of time outdoors and may therefore represent exposure to a contaminated environment (air and soil) hence the higher lead levels.

There was a trend towards significance for the protective effect of breastfeeding, with those breastfeeding being associated with levels <2μg/dl (p value 0.057) after the multivariate regression.
The main strength of this study was that it analysed biological samples in a highly specialized laboratory with excellent quality control making the results reliable. The recruitment of a comparative arm allowed for comparison of blood lead levels measured between the two groups. This is the first study conducted in children after the complete phase out of leaded fuels in Kenya and is probably reflective of the effects of reducing environmental contamination with lead from fuels. It is also the study on lead levels with the highest proportion of infants in Kenya, probably demonstrating the levels of lead exposure in this young and vulnerable age group that were previously undefined. The recruitment of the comparative group from the outpatient clinics was the major limitation of the study and may have been a significant contributor to the baseline differences observed between the subjects with rickets and those without rickets. The low prevalence of lead levels in the population (1 subject), also made it difficult to make statistical inferences on characteristics associated with elevated blood lead levels.

**CONCLUSION**

The prevalence of elevated blood lead levels was low in the entire study population at 2% in the group with rickets and 0% in the comparative group with no significant differences between the two groups.

None of the child or socio-demographic factors assessed was associated with levels ≥10μg/dl

In this study, rickets was not associated with elevated blood lead levels.

**RECOMMENDATION**

Since lead exposure even at low levels has serious adverse outcomes, reinforcement of preventive measures and public health education to reduce or eliminate exposure is paramount.

A larger study is justified to be able to account for some of the confounders of lower lead levels in the group with rickets.
References


33. Horkwitz M. Metabolism of Vitamin D in Lead Poisoning. *Nutrition reviews* a. 198; 39: 372-374


45. National Health and Examination Survey (NHANES 2000)
   www.cdc.gov/nchs/nhanes.htm


   www.cfpub.epa.gov
APPENDIX 1: QUESTIONNAIRE

QUESTIONNAIRE-BLOOD LEAD LEVELS AMONG CHILDREN WITH RICKETS & CONTROLS AT KNH

STUDY SERIAL NO……….. CONTACT OF PRIMARY CAREGIVER……………..
AGE (MONTHS)……….. SEX………………..
BIRTH WEIGHT……….. GESTATION AT BIRTH…………………..

LEAD LEVELS………..

1. Maternal age (years)………..
2. Maternal parity………..
3. Birth interval (last)………..
4. Maternal level of education
   None Primary secondary post secondary
5. Occupation of primary care giver……………..
6. Child currently breastfeeding
   1. Yes 2. No Total duration of breastfeeding (months) …………..
7. Use of vitamin/mineral supplement other than Vitamin A
   1. No 2. Yes (i) Duration of use…..
   (ii) Specify type………..
8. Sunlight exposure
   1. No 2. Yes (i) Frequency (times/week)
   (ii) With clothes [yes] [no]
   (iii) Duration (hours)………..
9. Residence
   • Urban
   • Peri-urban
10. How long has the child lived in the current residence?
   1. <6 months  2. > 6 months

11. Distance from a major highway
   1. < 1km  2. > 1km

12. What type of floor is in the house the child lives?
   - Earthen
   - Wooden
   - Cement
   - Tiled
   - Other

13. What type of walls are in the house the child lives?
   - Stone walls
   - Wooden
   - Earthen

14. Are the walls painted?
   1. No  2. Yes. If yes go to b

   (b) Is the paint chipping?
   1. Yes  2. No

15. Sources of water
   - Municipal water
   - Borehole
   - Rainwater
   - River
   - Other
16. What type of container is used to store the water?
   - Metal
   - Plastic
   - Earthenware
   - Other. Specify……………

17. Does the child exhibit excessive pica activity?
   1. Yes  2. No

18. Do you live near a dumpsite? (within 3kms)
   1. Yes  2. No

**Part b: TO BE FILLED FOR THOSE WITH A DIAGNOSIS OF RICKETS**

1. Date of diagnosis……..

2. Clinical findings
   - Craniotabes [Yes] [No]
   - Frontal bossing [Yes] [No]
   - Widening of wrists [Yes] [No]
   - Rachitic rosary [Yes] [No]
   - Harrison’s sulcus [Yes] [No]
   - Pigeon’s chest [Yes] [No]

1 Radiological Findings
   - Generalized osteopenia [yes] [no]
   - Flaying & cupping of distal ends [yes] [no]
   - Fractures present [yes] [no]

2 Biochemistry
   Serum calcium (mmol)………..
   Serum phosphates (mmol)……..
3 Treatment prescribed:
   (i) Vitamin D a. none b. injectable form c. oral drops
   (ii) Calcium supplements (specify)……….
APPENDIX 2......CONSENT FORM

BLOOD LEAD LEVELS AMONG INFANTS & CHILDREN WITH RICKETS CONSENT FORM

PATIENTS STUDY IDENTIFICATION NUMBER:……………………

DATE:…………………………

STUDY TITLE: BLOOD LEAD LEVELS AMONG CHILDREN DIAGNOSED WITH RICKETS AT KNH.

INVESTIGATOR: DR IMMACULATE WAMBUI KARIUKI

RESIDENT- DEPT OF PAEDIATRICS & CHILD HEALTH

UNIVERSITY OF NAIROBI

TEL-0721877053

SUPERVISORS

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University of Nairobi
INVESTIGATOR’S STATEMENT

We are requesting you and your child to kindly participate in this research study. The purpose of this consent form is to provide you with the information you will need to help you decide whether to participate in the study. This process is called ‘Informed Consent’. Please read this consent information carefully and ask any questions or seek clarification on any matter concerning the study with which you are uncertain.

INTRODUCTION

Rickets is a disease characterized by insufficient mineralization of the growing ends of bones. Rickets is caused by deficiencies of Vitamin D, calcium or phosphorus which are important for mineralization of bone. Lead is a toxic heavy metal that has been found in significant amounts in the environment. Lead causes a reduction in the levels of calcium, Vitamin D & phosphorus through various mechanisms. Children who are calcium deficient as maybe the case in rickets also tend to accumulate more lead in their body.

The blood lead levels in children with rickets in KNH have not been described. This study seeks to determine the blood lead levels in the children diagnosed with rickets and compare that to the lead levels of children without rickets.

BENEFITS

The results of this study will be made available to you and the clinician taking care of your child. You will also receive education on how to prevent lead toxicity in your home environment. The results of this research will be used by clinicians in this clinic and other clinics so as to improve care of children diagnosed with rickets.

RISKS

The risks anticipated in this study are related to the venepuncture your child will get to draw a sample of blood for determining the blood lead levels. This may include bleeding
at the venepuncture site, pain etc. Refusal to participate in this study will not in any way change the care given to your child.

**CONFIDENTIALITY**

The information obtained about you, your child and your family will be kept in strict confidence. No specific information regarding you, your child or your family will be released to any person without your consent. We will however, discuss overall findings regarding all children assessed but nothing specific will be discussed regarding your child. We will also not reveal the identity of you or your child in these discussions.

**VOLUNTARINESS**

The study will be fully voluntary. There will be no financial rewards to you for participating in the study. One is free to participate or withdraw from the study at any point. Refusal to participate will not compromise your child’s care in any way.

**STUDY PROCEDURE**

This study will be carried out in two main ways . The first will involve answering of a series of questions regarding your child and his/her home environment. The second will involve a sterile procedure carried out by a qualified phlebotomist where 2-3 mls of blood will be drawn from your child’s vein with care to avoid risk of infection at the venepuncture site. The sample will be taken to the laboratory for processing to determine the lead levels. No other procedures or tests will be carried out on the blood sample and the sample will be disposed off after processing in an appropriate manner. The results of the test will be availed to you at a later date.

**PROBLEMS OR QUESTIONS:**

If you ever have any questions about the study or about the use of the results you can contact the principal investigator, **Dr Immaculate Kariuki** by calling **0721877053**.

If you have any questions on your rights as a research participant you can contact the **Kenyatta National Hospital Ethics and Research Committee (KNH-ESRC)** by calling **2726300 Ext. 44355**.
Consent Form: Participant’s Statement:

I ________________________________ having received adequate information regarding the study research, risks, benefits hereby AGREE DISAGREE (Cross out as appropriate) to participate in the study with my child. I understand that our participation is fully voluntary and that I am free to withdraw at any time. I have been given adequate opportunity to ask questions and seek clarification on the study and these have been addressed satisfactorily.

Parent’s/Care giver’s Signature: __________________________ Date ______

______________________________ declare that I have adequately explained to the above participant, the study procedure, risks, benefits and given him/her time to ask questions and seek clarification regarding the study. I have answered all the questions raised to the best of my ability.

Interviewers Signature _________________ Date _______________
APPENDIX 3......Budget

The following are estimates of the costs to be incurred by the researcher while conducting this study. The researcher will fund the study from her own resources.

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<th>Unit Cost (KShs)</th>
<th>Total (KShs)</th>
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