

Blood Lead Levels in children 2 through 59 months old in Bhutan



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ABSTRACT

Introduction: Pediatric lead exposure has long-term health, public health, and economic consequences; however, it is an under-recognized problem in low and middle-income countries. Our objective was to determine the prevalence of elevated blood lead levels (EBLLs) ($\geq 5\mu\text{g/dL}$) and evaluate risk factors for EBLLs in infants and children in two cities in Bhutan. **Methods:** A cross-sectional study of children 2 through 59 months old was conducted in Thimphu and Phuentsholing, Bhutan during 2018. Blood was obtained by finger-stick and tested using a LeadCare II analyzer (Meridian Bioscience). Data were double entered in EpiData 3.1 and validated. Excel, Prism8, and STATA/IC 15.1 were used for analysis. **Results:** Among 531 participants, the prevalence of EBLLs was 43.88%. The prevalence of EBLLs in girls and boys was 37.40% ($n=262$) and 50.19% ($n=269$), respectively ($p=0.004$). The prevalence in Thimphu was 52.35% ($n=361$), compared to 25.88% ($n=170$) in Phuentsholing ($p<0.001$). In Thimphu, 70.47% ($n=149$) of 1 - 4 year old children tested in spring had EBLLs, compared to 51.45% ($n=138$) in autumn ($p=0.001$). Of the risk factors assessed, only regularly eating with fingers or hands was significantly associated with EBLLs ($p<0.001$). **Conclusion:** Nearly half of participants in two cities in Bhutan had elevated blood lead levels. Seasonal exposure to lead appears to be present. The high prevalence in this study is alarming. Further studies are urgently needed to both characterize the sources of lead and validate these findings on a larger scale.

Keywords: Bhutan; Blood lead level; Lead exposure; Lead poisoning; Prevalence.

INTRODUCTION

Lead is a naturally occurring, widely used metal that is detrimental to human health. Multiple medical, psychological, and economic problems are associated with lead exposure¹, and the damage is irreversible and long lasting². There is no level of lead in one's blood that is safe^{3,4}. Compelling proof exists that lead causes learning problems, neurological damage and behavioral changes in children even at blood lead levels (BLLs) below the current Centers for Disease Control and Prevention (CDC) cut-off of $5\mu\text{g/dL}$ ^{5,6}. A 2017 article reported that children with elevated blood lead levels had lower IQ scores and a lower socioeconomic status three decades later as adults⁶. Lead has been associated with immunological, renal, reproductive, endocrine and cardiovascular problems later in life^{3,7,8}. The economic toll on a society that has a high prevalence of elevated blood lead levels (EBLLs) is staggering, as it mentally stunts a society while adding many mental, physical, and social problems⁹. Exposure to lead is an under-recognized problem in Low and Middle Income Countries (LMICs), and the financial cost of lead exposure disproportionately affects LMICs⁹. In 2013, a study estimated that the yearly economic loss in LMICs from lead exposure was \$977 billion. In Asia, the yearly economic loss was

estimated at \$700 billion (1.88% of GDP)⁹. Based on Bhutan's GDP, the estimated economic loss related to lead exposure could be in the billions of Ngultrum per year¹⁰.

People are exposed to lead through its use in many common products, both past and present. Its use in paint, petrol and batteries is well-known, but it can be found in many other products and can contaminate food or spices. When products containing lead disintegrate, lead can contaminate surfaces inside and outside of homes where it can be accidentally ingested. Infants and toddlers are at the highest risk because of their hand-to-mouth behavior, and they also absorb a higher percentage of lead compared to adults. When iron and calcium are deficient in children's diets, lead is absorbed more readily³. Bhutan's National Nutrition Survey 2015 found 83.4% of children between 6 - 23 months did not receive iron-rich foods in their diets¹¹. These children have the potential to absorb lead more readily and suffer the detrimental consequences the rest of their lives.

WHO's 2010 publication, "Childhood Lead Poisoning" states it best, "When exposure to lead is widespread, low level toxicity can damage health, reduce intelligence, damage economies, and incapacitate the future leadership and security of entire countries"². However, the extent of exposure to lead is virtually unknown in Bhutan. The objective of the study was to determine the prevalence of EBLLs ($\geq 5\mu\text{g/dL}$) in infants and children in Thimphu and Phuentsholing and to determine risk factors.

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METHODS

Design

A cross-sectional study of infants/children 2 through 59 months old was conducted at the Community Health Department at Jigme Dorji Wangchuck National Referral Hospital (JDWRH) in Thimphu and at the Maternal Child Health (MCH) clinic at Phuentsholing General Hospital (PGH).

Study setting

Thimphu and Phuentsholing were selected as they are the two largest cities in Bhutan. Thimphu is the largest city and Phuentsholing is the industrial hub of the country on the border with India. These two locations see large numbers of children seeking preventive MCH services daily. Data collection was done in Thimphu from 30th April to 18th May 2018 (subsequently referred to as “spring”) and from 13th October to 2nd November. Data collection in Phuentsholing was done between 13th and 15th November 2018. Data collected in October/November is subsequently referred to as “autumn”. These dates reflect when the research team was available for data collection.

Study population

Due to the lack of prior local data, a broad range of ages from 2 through 59 months was included in the study. The target was to enroll 500 participants-70% from Thimphu and 30% from Phuentsholing, with equity between gender and age groups. Since infants less than six months of age come more often for immunizations and growth monitoring, the group less than one year old was subdivided into ≥ 2 to < 6 months and ≥ 6 to < 12 months in order to reduce overrepresentation of the youngest infants in the < 1 -year-old age group. Infants less than two months of age were excluded based on the assumption they had minimal exposure risk.

Inclusion/Exclusion

Any infant/child ≥ 2 and < 60 months old who came to the Community Health Department or MCH clinic was eligible. Participants were excluded if they were previously in the study, had received a blood transfusion in the prior 60 days, required hospitalization, or if parents did not provide informed consent. Participants were also excluded if they had lived outside of Bhutan for over two months in the prior year.

In Phuentsholing, due to a housing shortage, some Bhutanese live in Jaigaon, India, the city adjacent to Phuentsholing. Participants who had an MCH handbook and received MCH services at PGH but lived in Jaigaon were not excluded. Participants were enrolled as they presented at both locations until each age category was filled.

Data collection

After meeting inclusion and exclusion criteria and obtaining parental informed consent, a structured interview questionnaire

was administered by one of the investigators. The infant’s MCH handbook, when available, was used for birth and other information. In addition, demographic data and thirty questions related to lead exposure risks, such as the presence of peeling paint in the dwelling, proximity to construction or busy roads, and the use of traditional medicines were asked. These questions were developed based on other studies as well as local context¹²⁻¹⁴.

Using aseptic techniques, blood was collected by a finger stick, or heel stick in those < 1 year of age. Samples were immediately tested on a LeadCare II device (Meridian Bioscience, Cincinnati, OH). LeadCare II is a device that is FDA-approved and CLIA (Clinical Laboratory Improvement Amendments)-waived, with confirmed accuracy appropriate for lead screening¹⁵.

Data analysis

Data were double entered in EpiData 3.1 (Odense, Denmark) and validated. Excel and Prism8 (San Diego, CA) were used to calculate and compare prevalence rates and associations, using Fisher’s exact or Chi-squared tests. STATA/IC 15.1 (College Station, TX) was used to assess the association between the time of testing (spring and autumn) and EBLLs. Those demographic variables which were significant at $p < 0.20$ in a bivariate analysis were included in a multivariable logistic regression model to calculate adjusted odds ratios (OR) with 95% confidence intervals.

Ethical considerations

The study was approved by Bhutan’s Research Ethics Board of Health (REBH) (*Ref. No. REBH/Approval/2017/074, dated 13/11/2017*). The Essential Medicines and Technology Division (EMTD) issued a letter, dated 15th November 2017, approving the use of LeadCare II for research purposes in Bhutan. Clearance from each hospital administration was also received, along with informed consent from parents or guardians.

RESULTS

The 531 participants were divided evenly between females (49.34%) and males (50.66%), and a similar number of participants were studied in each of the five age ranges (Table 1). Two-thirds of the participants were from Thimphu, and 92% provided a household income range.

The prevalence of elevated blood lead levels (EBLLs) was 43.88% among infants/children 2 – 59 months old. Another 36.53% had detectable blood lead levels between 3.3 $\mu\text{g/dL}$ (the lower limit detected by the LeadCare II device) and $< 5 \mu\text{g/dL}$. Half (50.19%) of males had EBLLs, compared to 37.40% of females ($p=0.004$). Fifty-two percent of Thimphu participants had EBLLs, compared to 25.88% in Phuentsholing ($p<0.001$). A significant difference in EBLLs was also present between income groups ($p=0.038$) (Table 1).

Table 1. Demographics and blood lead levels of infants and children (n=531) in Thimphu and Phuentsholing, Bhutan, 2018

Characteristic	Frequency of Participants n (%)				p-value [†]
	Total*	Non-reportable BLL <3.3 µg/dL	BLL 3.3 – 4.9 µg/dL	EBLL ≥5.0 µg/dL	
Total	531	104 (19.59)	194 (36.53)	233 (43.88)	
Sex					
Female	262 (49.34)	59 (22.52)	105 (40.08)	98 (37.40)	0.004
Male	269 (50.66)	45 (16.73)	89 (33.09)	135 (50.19)	
Age (months)					
2 – 11	105 (19.77)	51 (48.57)	37 (35.24)	17 (16.19)	
12 – 23	106 (19.96)	15 (14.15)	32 (30.19)	59 (55.66)	
24 – 35	105 (19.77)	9 (8.57)	36 (34.29)	60 (57.14)	
36 – 47	110 (20.72)	14 (12.73)	51 (46.36)	45 (40.91)	
48 – 59	105 (19.77)	15 (14.29)	38 (36.19)	52 (49.52)	
Location					
Thimphu	361 (67.98)	47 (13.02)	125 (34.63)	189 (52.35)	<0.001
Phuentsholing	170 (32.02)	57 (33.53)	69 (40.59)	44 (25.88)	
Monthly Household Income (Nu)					
<15,000 Nu	150 (28.25)	30 (20.00)	45 (30.00)	75 (50.00)	0.038 [‡]
15,000 – 24,999	144 (27.12)	25 (17.36)	52 (36.11)	67 (46.53)	
≥25,000	193 (36.35)	47 (24.35)	81 (41.97)	65 (33.68)	
Unknown/Did not answer	44 (8.29)	2 (4.55)	16 (36.36)	26 (59.09)	

Abbreviations: BLL: blood lead level; EBLL: elevated blood lead level

*Column percentage

[†]p values calculated using Fisher's exact test unless otherwise stated

[‡]Chi-squared test used

Among the 170 participants tested in Phuentsholing, 23 resided in India. Among those residing in India, 4 (17.39%) had EBLLs, ranging from 5.1 – 6.3 µg/dL. Among the 147 tested in Phuentsholing who lived in Bhutan, 40 (27.21%) had EBLLs.

Overall, BLLs ranged from non-recordable (<3.3 µg/dL) to 30.9 µg/dL. Among participants with EBLLs, the majority had BLLs between 5.0 – 9.9 µg/dL (Figure 1).

Figure 2 shows the prevalence of EBLLs by age groups. The under-12-months age group was sub-divided to better show the increasing prevalence in the first year of life. The 24 to 35 month old children had the highest prevalence (57.14%) followed closely by the 12 to 23 month old age group (Figure 2).

A statistical difference in the prevalence of EBLLs between seasons was identified in children 1 to 4 years of age tested in Thimphu. Also, a statistical difference in the prevalence of EBLLs in the autumn between Thimphu and Phuentsholing was identified (Table 2).

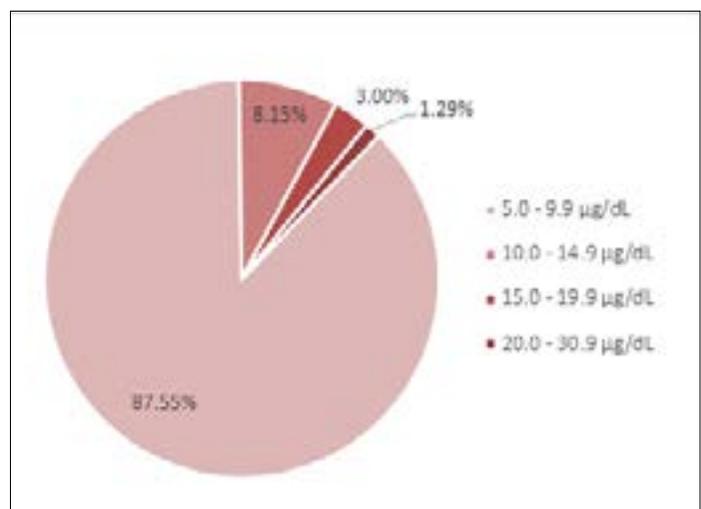


Figure 1. Percentage of participants with EBLLs (n=233) by blood lead level range in Thimphu and Phuentsholing, Bhutan, 2018

Among 1 to 4 year old children tested in Thimphu, those tested in the spring had 1.98 times (adjusted OR 1.98, 95% CI: 1.67 to 3.36) greater risk of EBLLs than those tested in the autumn after being adjusted for age, sex and household income. Increasing household income was also found to be significant and protective (Table 3).

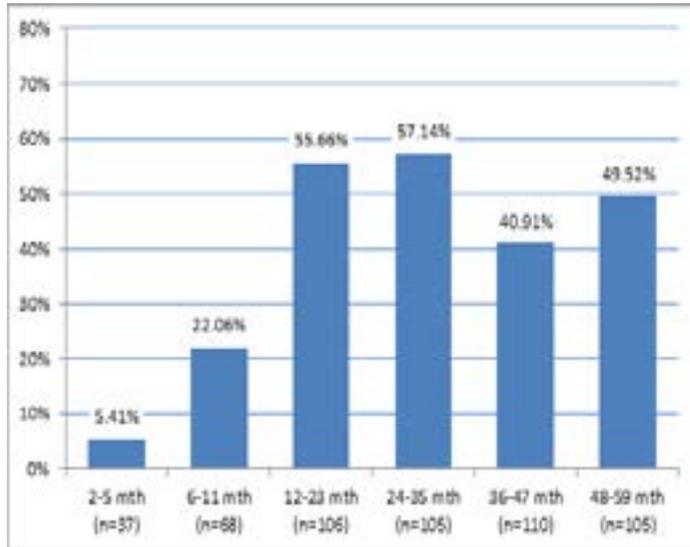


Figure 2. Prevalence of elevated blood lead levels by age (n=531) in Thimphu and Phuentsholing, Bhutan, 2018

Table 2. Comparison of the prevalence of elevated blood lead levels in 1-4 year olds tested between seasons in Thimphu or between Thimphu and Phuentsholing Bhutan, 2018

	Frequency of Participants n (%)			p-value*
	Total	BLL <5.0 µg/dL	EBLL ≥5.0 µg/dL	
Thimphu only:				
Total No. (%)	287	111 (38.68)	176(61.13)	
Season of testing†				
Spring	149	44 (29.53)	105(70.47)	0.001
Autumn	138	67 (48.55)	71 (51.45)	
Autumn only‡:				
Total No. (%)	277	166 (59.93)	111 (40.07)	
Location				
Thimphu	138	67 (48.55)	71 (51.45)	<0.001
Phuentsholing	139	99 (71.22)	40 (28.78)	

Abbreviations: EBLL: elevated blood lead levels

*p values calculated using Fisher's exact test

†Spring refers to April/May, Autumn refers to October/November

Table 3. Odds ratios for EBLLs in participants 1-4 years old (n=287) tested in Thimphu, Bhutan, 2018

Characteristics	Participants n	Participants with EBLL n (%)	Crude OR	Adjusted OR
Season of testing*				
Autumn	138	71 (51.45)	Ref	Ref
Spring	149	105 (70.47)	2.25 (1.39 to 3.66)*	1.98 (1.67 to 3.36)*
Age (yrs.)				
1	70	45 (64.29)	Ref	Ref
2	72	48 (66.67)	1.11 (0.56 to 2.22)	1.11 (0.53 to 2.29)
3	75	39 (52.00)	0.60 (0.31 to 1.17)	0.70 (0.35 to 1.41)
4	70	44 (62.86)	0.94 (0.47 to 1.87)	1.21 (0.58 to 2.56)
Sex				
Female	134	75 (55.97)	Ref	Ref
Male	153	101 (66.01)	1.53 (0.95 to 2.46)	1.24 (0.74 to 2.06)
Household Income (Nu)				
≥25,000	103	48 (46.60)	Ref	Ref
15,000-24,999	82	55 (67.07)	2.33 (1.28 to 4.26)*	2.09 (1.13 to 3.89)*
<15,000	75	53 (70.67)	2.76 (1.47 to 5.18)*	2.45 (1.28 to 4.70)*
Not reported	27	20 (74.07)	3.27 (1.27 to 8.41)*	1.91 (1.11 to 7.62)*

*Significant at p<0.05

Table 4. Risk factors for elevated blood lead levels (n=531) in Thimphu and Phuentsholing, Bhutan, 2018

Tested risk	Total	BLL <5.0 µg/dL	EBLL ≥5.0 µg/dL	p-value*
Where participant lives[†]				
Thromde (developed area)	476	266	210	0.886
Outside thromde (less developed area)	54	31	23	
Age of home				
<10 years old	225	131	94	0.521
≥10 years old	261	144	117	
Do not know	45	23	22	
Type of home				
Traditional Bhutanese	61	25	36	0.294 [‡]
Modern (brick/cement)	423	248	175	
Wooden	30	14	16	
Other (temporary structure)	15	10	5	
Do not know	2	1	1	
Any peeling paint in home				
Yes	179	104	75	0.710
No	341	191	150	
Do not know	11	3	8	
Construction/renovation inside home				
Yes	62	38	24	0.416
No	467	259	208	
Do not know	2	1	1	
Construction/renovation close to home				
Yes	278	148	130	0.163
No	253	150	103	
Use of home-made metal cooking spoon				
Yes	463	255	208	0.292
No	67	42	25	
Do not know	1	1	0	
Regularly eats with fingers/hands[§]				
Yes	328	150	178	<0.001
No	154	102	52	

Abbreviations: BLL: blood lead level; EBLL: elevated blood lead level; Nu: Ngultrum

*p values calculated using Fisher's exact test, without including "do not know" category, unless otherwise stated

[†]n=530 (1 missing with a BLL <3.3)

[‡]Chi-squared test used, without including "do not know" category

[§]Only asked of those ≥6 months of age (n=494)

Table 4 shows a sampling of the questions asked from the questionnaire related to potential lead exposure risk factors. The only significant risk factor was regularly eating with fingers/hands (p<0.001).

DISCUSSION

Nearly 44% of infants/children in this study were found to have EBLLs. These findings are as alarming as prevalence levels of 48-66% found in studies in India, Nepal, and Haiti^{12,13,16}. By comparison, in developed countries where lead prevention programs have been active for decades, the prevalence of EBLLs

is as low as 1.3%¹⁴.

This study found significantly more boys had EBLLs compared to girls. This finding is seen in a study from Bangladesh as well as among refugee children coming to the US from many different countries^{17,18}. However, other studies do not find this sex difference.

Lead exposure is often connected to areas of mining of metal ore and smelting, industrial and recycling centers, and locations with poorer air quality. Although mining metal ore and smelting is not done in Bhutan, the other characteristics were present in Phuentsholing.

The lower prevalence of EBLLs (25.88%) in Phuentsholing, the industrial hub of Bhutan, compared to Thimphu (52.35%) was unexpected. Even after controlling for the season and age range among children 1 through 4 years old, there was still a significant difference in the prevalence between the cities (28.78% vs. 51.45%).

Although including children who live in Jaigaon, India was thought to be a risk for elevating the prevalence rate of EBLLs in Phuentsholing, the opposite was the case. The prevalence of EBLLs among the 23 participants who resided in India was 10% lower than those tested at PGH who lived in Bhutan, lowering the overall prevalence of EBLLs in Phuentsholing.

As healthcare is free in Bhutan, people may seek services from the facility of their choice. Most people would seek routine preventive services close to home, however since JDWRH is the national referral hospital, a few participants tested there may have been from outside of Thimphu, so the findings are not specific to a city, but to the participants availing services at each site.

Many hypotheses have been raised for the higher prevalence of EBLLs in Thimphu, including differences in the amount of demolition and construction, weather differences, and possible water contamination. Those will need to be investigated in future studies.

Increasing household income has been associated with a lower prevalence of EBLLs¹⁹. Although this study found similar findings, the prevalence of EBLLs in infants/children in the highest income households is still very high at 33.68%. Although income is slightly protective, still 1 in 3 children in the wealthiest households had EBLLs.

This study was not designed to study the difference in prevalence of EBLLs during different times of the year. However, when the data were analyzed, an unexpected 19% difference was noticed between the two testing periods among 1-4 year-old children in Thimphu. This comparison could only be done in Thimphu as all the data was collected in Phuentsholing in November. Even after controlling for age and gender, through multivariable logistic regression, the difference in EBLLs between the April/May testing and the October/November testing in Thimphu remained significant.

Although seasonal differences of 2.5 - 4% have been documented in US studies, the highest rates have been found in

the warmest months^{17,20}. As this findings are 5 to 8 times higher than those studies, and the higher prevalence in Thimphu was in the cooler season, many questions regarding the cause remain.

One possible cause of the lower prevalence of EBLLs in autumn compared to spring in Thimphu and the lower prevalence of EBLLs in Phuentsholing over Thimphu may be the moisture and weather conditions. Decreased soil moisture and dry windier condition in areas with lead-contaminated soil have been shown to increase BLLs²¹.

Many studies have identified paint, batteries in homes, or battery recycling as risk factors for lead exposure¹²⁻¹⁴. Of the 30 questions on risk factors tested, the only one significantly associated with EBLLs was regularly eating with fingers or hands. The combination of the high overall prevalence, lack of significant decrease in prevalence with increasing age, and the lack of additional statistically significant risk factors leads us to believe that lead is pervasive in the environment.

As India, Bhutan's largest trading partner, only implemented a 90ppm lead level limit in paint in November 2017, paint is a very likely source²². This, combined with a high number of buildings being demolished and replaced with larger structures in Thimphu, might be a source of wide-spread environmental lead contamination. From other studies and recalls of toys and consumer goods made in Asian countries, there are likely multiple other sources of lead in Bhutan as well.

From current knowledge and the findings from this study, we can encourage parents to regularly wash their infants/children's hands, especially before eating, to reduce lead exposure. Assuming that lead is present in dust, wiping dusty surfaces and floors in the home with damp cloths and minimizing infant's and children's exposure to peeling paint will likely reduce exposure to lead. Additionally, ensuring good nutrition, especially good intake of iron and calcium will help to prevent excess absorption of lead.

LIMITATIONS

As testing of venous blood lead levels was not available in Bhutan, a finger-stick testing method was utilized for this study. This approach has the potential of showing increased lead levels due to lead contamination, however CDC's guidelines were used to develop the best protocol for minimizing contamination²³. Also, although the study size was adequate to determine a high prevalence of EBLLs, the findings cannot be generalized to all of Bhutan, due to testing in only two cities.

CONCLUSION

Nearly half of infants/children studied in two cities in Bhutan had elevated blood lead levels. Seasonal exposure differences appear to be present. This study highlights an alarming situation and the urgent need for further studies to both characterize the sources of lead and validate these finding on a larger scale. A subsequent

long-term comprehensive public health intervention will also be needed once more information is available.

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AUTHORS CONTRIBUTION

Following authors have made substantial contributions to the manuscript as under:

PE: Concept, design, data collection and analysis, manuscript writing and review.

DP: Concept, Design, data collection and analysis, manuscript writing and review

PT: Design, data collection, manuscript writing and review

SP: Design, data collection, manuscript writing and review

MSG: Design, data collection and analysis, manuscript writing and review

SU: Design, manuscript writing and review

KE: Concept, Design, data collection and analysis, manuscript writing and review

Author agree to be accountable for all respects of the work in ensuring that questions related to the accuracy and integrity of any part of the work are appropriately investigated and resolved.

CONFLICT OF INTEREST

None

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