# Original article

# Blood Lead Levels of Primary School Children in Kathmandu Municipality, Nepal

Sherchand O1, Mehta K.D2, Poudel P3, Deo B4, Baral N5

<sup>1</sup>Department of Biochemistry, Nepal Medical College, Jorpati

<sup>2.5</sup>Department of Biochemistry, <sup>3</sup>Department of Pediatric, <sup>4</sup>Department of Psychiatry; B.P. Koirala Institute of Health Sciences, Dharan, Nepal.

**Correspondance:** Dr. Ojaswee Sherchand, **Email**: drojasweesherchand@hotmail.com

#### **Abstract**

**Introduction**: Lead is a toxic metal which has contaminated our environment and created health problems around the globe. Children are vulnerable to lead as the intake per unit body weight is higher and even low levels can cause neurological damage. Nepal lacks data on sources of lead exposure and its health impacts; hence screening of blood lead is mandatory.

**Objectives:** To determine blood lead levels, its associated risk factors and impact on health in primary school children in Kathmandu municipality, Nepal.

**Methods:** A total of 218 school children between 6-16 years age from Kathmandu were included in the study undertaken from November 2012 to June 2013. Consents were taken from guardians and students. Questionnaire was used to acquire data followed by blood sampling. Lead was measured using atomic absorption Spectrophotometer, hemoglobin and serum calcium was measured using commercial kits. Intelligent Quotient was assessed using Seguin form board. Data was analyzed using SPSS version 20.

**Results:** Overall, 63% (137) had detectable blood lead level (BLL) and 54% (117) had BLL  $\geq$  5 ug/dl. 55% were male and 45% female. The median BLL (IQR) was 8(0-34) and 4(0-18) in males and females respectively. The mean hemoglobin and serum calcium was 13.7 $\pm$ 2.4 and 8.1 $\pm$ 1.8 respectively. On risk factors evaluation, the odds of having elevated blood lead level (EBLL) was significantly higher in children living in homes with chipping walls (p value 0.001). Children belonging to families with lower socioeconomic status (p value 0.001) and residing near traffic congested areas (p value 0.007). The median IQ was 73.5(66–91). EBLL showed significant negative correlation with IQ (p value 0.001). EBLL was identified as significant risk factor lowering IQ by 2.35 points per 10 ug/dl rise in blood.

**Conclusion:** Children living in homes with chipping walls, having lower socioeconomic status and residing near traffic congested areas had significantly higher lead level. EBLL showed significant negative correlation with IQ.

Keywords: Blood lead level, elevated blood lead level, Intelligent Quotient, Lead

## Introduction

Lead is a naturally occurring toxic heavy metal without any demonstrated biological benefit. It has contaminated our environment globally due to its extensive use. As a result exposure to lead is inevitable in day to day life.

This continuous exposure has adverse effects on our health, affecting multiple body systems; the neurological, hematological, gastrointestinal, cardiovascular, renal and reproductive systems.<sup>2</sup> Even though its detrimental

effect spares no age group, infants and children are particularly vulnerable to the toxic effects of lead due to their increased oral exploratory behavior, greater gastrointestinal absorption and immature blood-brain barrier.<sup>3</sup> Consequently, in infants and children even relatively low levels of exposure can cause serious and at times, irreversible neurological damage.<sup>4</sup> These growing scientific evidences have compelled scientists to gradually decline the acceptable blood lead level to 5ug/dl in 2012.<sup>5</sup> This may be further lowered in years to come as newer evidences come into light.

Most countries around the globe have conducted several studies<sup>6,7,8,9</sup> about Lead and its impact on health and have formulated strategies to remove exposure sources. However, in Nepal only few studies have been conducted<sup>10, 11</sup> but non addressing children who are most at risk. Globally, children's blood lead levels has declined, after the removal of lead from gasoline<sup>12</sup>, however, the lead levels are still high which necessitates the search for other important sources. Studies conducted on paints in Nepal depicted high lead content implying it as a potential exposure source.<sup>13</sup> Another important source is lead acid batteries due to its growing use especially after the government's encouragement to use solar power in an attempt to save electricity. Improper dumping of such used lead acid batteries can contaminate the surrounding environment with lead.

We have conducted a cross sectional study to measure the blood lead level in school children in Kathmandu municipality, Nepal. We want to establish the baseline blood lead level in children and explore the risk factors including socio-demographic factors and biochemical parameters. We have also evaluated the impact of blood lead level on intellectual performance. However, the most significant objective of our study is to spread public awareness regarding lead toxicity and to awaken our policy makers to impose proper guidelines necessary for health professionals and the general public to prevent lead toxicity.

#### **Materials and Methods**

**Subject:** This cross sectional study was conducted in primary school children of Kathmandu Municipality, Nepal from November 2012 to June 2013. Six schools were selected to represent the sociodemographic profile of the entire city (Figure 1). A total of 218 Primary school students between the ages of 6-16 years, were included. Although children aged less than 6 years are most vulnerable to the effects of lead, logistic complexities made examining these younger ages difficult. Ethical clearance was taken from Ethical Review Board of B.P.K.I.H.S, Kathmandu Municipality and Nepal Health Research Council for

conducting the research. The rationale and the objectives of our study were discussed with the school administration committee and guardians. Written informed consent was taken from Guardians/Head Master and children themselves. Educational information regarding harmful effects of lead on health and its impact on the cognitive development of children was delivered to class teachers and students after completion of the sampling. Pamphlets containing information about lead and its potential exposure sources, and emphasis on removing such sources were also distributed to each participant and to the teachers of the school as well as the guardians.

Questionnaire: A semi-structured Questionnaire was taken from students with some assistance from teachers and guardians. Data regarding socio-demographic variables, parental educations and occupations, neighborhood condition such as crowded neighborhood, presence of factories or welding shops, battery recycle plants, use of paint in house and school and condition of walls (chipping), knowledge about the harmful effects of lead, and use of cosmetic that contains high concentration of lead such as surma (a kind of traditional eye-cosmetic). Dietary habits, source of drinking water, and usage of any ceramic utensil were also enquired. Clinical assessment and anthropometric measurements were obtained.

**Blood sample collection**: 5ml blood was drawn from antecubital vein under strict aseptic conditions; 3 ml was placed into standard commercial evacuated tubes containing sodium heparin (Vacutainer®) and 2 ml was placed in glass tubes and allowed to clot for serum separation to measure biochemical parameters. The samples were transported to Biochemistry laboratory, B.P.K.I.H.S maintaining cold chain and stored at -20°C until analysis.

Estimation of BLL: BLL was estimated from blood samples collected in vacutainer. Samples were first subjected to acid digestion and then lead (Pb) levels were measured in these samples. BLL was measured using Flame Atomic Absorption Spectrophotometer (Thermo Scientific brand, China) using an air-acetylene flame for lead (heavy metals). A linear calibration curve for Pb was generated using four calibrator solutions (0ppm, 1ppm, 2ppm, 3ppm). The calibration curve was verified using a control sample (0.3 ppm). Aspiration of standard was done after every ten sample to check the instrument. All necessary precautions were taken to avoid any possible contamination of the sample. Flame AAS was chosen as it is easy to use, fast, relatively low interferences and gives a good performance. Blood lead level  $\geq 5$  ug/dl considered as elevated blood lead level as per Centre for Disease Control U.S.A. 2012. Further stratification of blood lead level was done to evaluate health effects of lead at different levels. (Table 1).

16 Sherchand O et al.,

Measurement of other biochemical parameters: Hemoglobin was measured with cyanomethemoglobin method. A criterion for anemia was established using reference from W.H.O guidelines. <sup>14</sup> Serum total calcium was measured using commercial kits (Accurex autozyme) by arsenazo III method. Normal range of calcium was established using Pediatric reference. <sup>15</sup> Body mass index (BMI) was calculated and stunting and wasting (thinness) were evaluated using WHO standards for that age and sex.

Intelligent Quotient test: IQ test was done by using Seguin form board and measured by applying Vineland Social Maturity Scale (Indian Adaptation) under the guidance of Psychologist from B.P.K.I.H.S. IQ was classified using Current Weschler Intelligence scale for children (WISC IV). Children with mental retardation were further classified as having mild, moderate, severe and profound mental retardation.

Statistical Tools: Descriptive statistics were used to analyze demographic parameters. Data were checked for normal distribution by SPSS and divided into groups based on gender and also divided into two groups based on age using ten years as cut off point. For normally distributed data, mean with standard deviation (SD), Pearson test and Student's T tests were applied. For data not having normal distribution, non-parametric tests were applied. Mann Whitney U test was used to compare BLLs with biochemical parameters as hemoglobin, calcium, and student t test for comparing BLL with IQ. The multivariable linear regression model was fitted to identify independent predictors of BLLs. Pearson correlation was used to determine correlation between BLL and hemoglobin, calcium, IQ. Multivariable logistic regression analysis was performed to explore the associations between these factors and BLL above 5μg/dL (≥5 μg/dL vs. <5 μg/dL). All analyses were done in Statistical Package for the Social Science (SPSS) version 20.

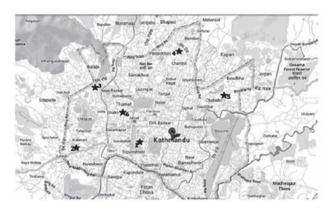


Figure 1: Map of Kathmandu, the number alongside the star indicates the location of school selected for study.

**Results:** The median BLL with (IQR) were 8(0-34) and 4(0-18) in males and females respectively. BLL was significantly higher in male gender (p value 0.02). The mean hemoglobin with SD was  $13.7\pm2.4$  and  $13.7\pm2.5$  in males and females respectively. Mean serum calcium was 7.9±1.4 in males and 8.2±2.0 in females. In age distribution, the younger age group ( $\leq 10$  years) had median BLL with IQR of 6(0-37) and older age group (>10 years) had median BLL with IQR of 5 (0-21). Mean hemoglobin of 13.6±2.4 and 13.8±2.4 was detected in younger and older age group respectively. Mean serum calcium was 8.4±1.7 in younger and significantly higher than in older age group (p value 0.03). The mean Body Mass Index (BMI) were 15.5  $\pm$ 2.3 in males and 15.8  $\pm$  2.6 in females. The summary of these parameters are summarized in Table 2. Analysis of nutritional parameters showed moderate to severe stunting in 41% male and 34% female children. In body mass Index (BMI) for age assessment, 12% had acute malnutrition and 5% had severe acute malnutrition. Total of 18% male and 16% female had moderate to severe acute malnutrition, summarized in Table-3. Comparison of average BMI of male and female with WHO standards showed most fell below WHO standards, given in figure 2 and figure 3.

Table no 1 Stratification of BLL

Level	BLL interval(ug/dl)
1	0-5
2	5-10
3	10-15
4	15-20
5	20- 59
6	60-69
7	70-79
8	≥80

Table 2 Biochemical Parameters of Children under Study

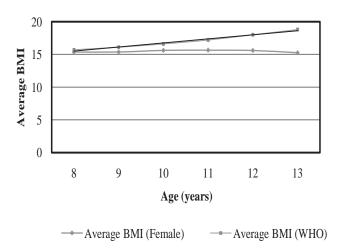
Variable	Number	BLL (ug/dl)	Hb (gm %)	Serum calcium	IQ	BMI
				(mg/dl)		
Age (years)	n(%)	Median(IQR)	$Mean \pm SD$	$Mean \pm SD$	Median(IQR)	$Mean \pm SD$
≤10	87(40%)	6 (0-37)	$13.6 \pm 2.4$	$8.4 \pm 1.7*$	76(66-98)	$15.3 \pm 2.4$
>10	131(60%)	5 (0-21)	$13.8 \pm 2.4$	$7.9 \pm 1.7$	73(65-89)	$15.8 \pm 2.4$
Sex						
Male	120(55%)	8 (0-34)**	$13.7 \pm 2.4$	$7.9 \pm 1.4$	73(66-91)	$15.5 \pm 2.2$
Female	98(45%)	4 (0-18)	$13.7 \pm 2.5$	$8.2 \pm 2.0$	74(65-91)	$15.8 \pm 2.6$

<sup>\*</sup> p< 0.05 Independent T test, \*\* p< 0.05 Man-Whitney U test; abbreviation: Hb is hemoglobin and IQ is Intelligent Quotient.

Table 3 Nutritional Parameters in sample population with Reference to WHO standards

Nutritional Parameters	Total n(%)	No of Children		Reference
BMI (z score)	218	Male n (%)	Female n (%)	
Median ±2 SD	180(83%)	98(82%)	82(84%)	Normal
< - 2SD to - 3SD	27(12%)	16(13%)	11(11%)	Mod thinness
< - 3SD	11(5%)	6(5%)	5(5%)	Severe thinness
Height for Age (z-score)				
Median ±2 SD	136(62%)	71(59%)	65(66%)	Normal
< - 2SD to - 3SD	57(26%)	33(28%)	24(24%)	Mod stunting
< - 3SD	25(11%)	16(13%)	9(9%)	Severe stunting

Abbreviations: SD is standard deviation, Mod is moderate



35
30
25
20
15
10
6 7 8 9 10 11 12 13 14 15 16
Age (years)

Average BMI (Male) — Average BMI (WHO)

Figure 2 Line comparing average BMI of female children with the WHO standard for age.

Figure 3 Line comparing average BMI of male children with the WHO standard for age.

Sherchand O et al.,

Risk factors for lead exposure: Ten risk factors were analyzed; they were anemia, hypocalcemia, undernourished status (<2 z score/age as per WHO), chipping paints at home, distance of home from heavy traffic roads(within 1 km), factory located near residence, source of drinking water, male gender, age≤ 10 years, low socioeconomic status of family. Among these chipped paints at home (p value 0.001), male gender (p value 0.03), lower socioeconomic status (p value 0.001), anemia (p value 0.01) and living near traffic congested roads (p value 0.007) were significantly associated risk factor for elevated blood lead level (EBLL). When the odds ratios were adjusted, chipping paints was found to be the most significant risk factor (p value 0.0001). It was also found that children living near factories, undernourished children, children

suffering from hypocalcemia were found to have higher BLL. These findings are summarized in Table 4.

Intelligent Quotient: The median IQ was 74 with IQR 66-91. On evaluating risk factor associated with low IQ, we found anemia, EBLL, undernourishment, lower socioeconomic status increased the risk for decreasing IQ. EBLL showed significant negative correlation with IQ scores (p value 0.001), (figure 4). Risk association was significant for EBLL compared to other risk factors (p value 0.01). Through our study we found an increase in BLL by 10ug/dl was associated with decrease in IQ by 2.35 points. We further stratified blood lead level to identify threshold affecting IQ, but we found the effect was the same at all levels of lead.

Table 4 Risk factors for Elevated blood lead level among 218 students in univariate (crude) and multivariate model (adjusted).

Variable	Total	EBLL	Crude OR	P-value	Adjusted	P-	
	n (%)	n=117(54%)	(95%CI)		OR (95%CI)	value	
Children's Characteristics							
Age(≤10 years)	87(40%)	48 (55%)	1.1(0.6-1.9)	0.7			
Male gender	120(55%)	72 (60%)	1.7(1.0-3.0)	0.03	1.6(0.8-2.9)	0.1	
Hypocalcemia	149(68%)	81(54%)	1 (0.6 - 1.9)	0.7			
Anemia	38(17%)	27(71%)	2.4(1.1- 5.2)	0.01	1.8(0.8-4)	0.1	
Under nourished	38(17%)	22(58%)	1.2(0.6- 2.5)	0.5%			
Chipped paints	39(18%)	30(77%)	3.5(1.6- 7.8)	0.001	7.7(2.5-24)	0.0001	
Factory near residence	41(19%)	25 (61%)	1.4(0.7- 2.8)	0.2			
Residence near heavy traffic road	191(88%)	109 (57%)	3.1(1.3–7.5)	0.007	8.1(2.3-28)	0.001	
Water source	162(74%)	89(55%)	1.2(0.7- 2.2)	0.5			
Lower Socioeconomic status	118(54%)	75(64%)	2.4(1.4- 4.2)	0.001	2(1.2-3.8)	0.01	

OR is odds ratio

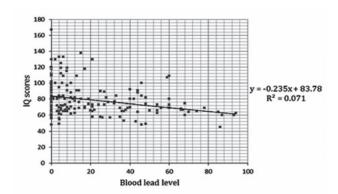


Figure 4 Negative correlation between Blood Lead levels and IQ

## **Discussion**

Our study indicates that despite the elimination of leaded gasoline since 2000, children in Kathmandu municipality are still exposed to high levels of lead. The median blood lead level of school children of Kathmandu was 5.5ug/dl. Overall, 63% of children had detectable blood lead level, 54% children had lead level above CDC cutoff point (BLL ≥5 ug/dl) and 40% children had lead level above 10 ug/dl. This indicates a serious health threat to our children from lead exposure. We found that children living in areas near heavy traffic congestion, homes where wall paints were chipping and belonged to families with lower socioeconomic status had significantly higher blood lead. Amongst these chipping paints was found to be the most significant risk factor (p value 0.0001).

The paints used in Nepal have demonstrated high lead contents<sup>16</sup> beyond the limit set by Global Alliance to Eliminate Lead in Paint (GAELP). GAELP recommended a limit of 90 ppm [0.009%] in paint and many countries around the globe have established paint policies to meet this standard. Nepal has yet to take a stern step for eliminating lead from paints.

Among gender comparison boys had significantly higher BLL than girls. This might be explained by the fact that boys spend more time engaged in outdoor activities and have greater hand-to-mouth activities than girls as a result are subjected to greater lead exposure. However, we can only postulate such reason as we did not gather information on physical activities in our cohort. Furthermore this association of gender and lead can be influenced by other factors. A recent study showed that physical activity and race/ethnicity can influence the association of gender and BLL<sup>17</sup>.

We also examined nutritional status of children and found undernourished children were more vulnerable to have elevated BLL. We found children with hypocalcemia and anemia had higher odds of having EBLL. Studies suggest that transport of lead across G.I. mucosa utilizes the transporters available for essential metals. Anemia specially iron deficiency increases divalent metal transporter 1 (DMT 1) expression to absorb iron, but this transporter is also utilized by lead for transport into intestinal lumen. Similarly, both calcium and lead utilize same transporter in the intestine. Reports from many experimental studies show that absorption of lead from gastrointestinal tract is inversely related to calcium content of diet.

A study conducted in Nepal few years back had shown that drinking water sources near heavy traffic areas were found to have high lead level. <sup>19</sup> However, in our study we found the odds of contaminated drinking water leading to elevated BLL though present was not significant. The currant WHO standard for the lead content of drinking water is 10ug/dl. At present, the principle source of lead in drinking water is lead solders used in joints of pipes and water mains, lead from such fittings leach into water especially when water has acidic pH.

We also found children belonging to families with lower socio-economic status had significantly higher BLL. Socioeconomic conditions can affect lives in many ways; poor people generally work in areas of poor hazard control where potential sources of lead exposure are present and their children play in such environment without any protection. Besides, the lack of clean drinking water and nutritious food result in malnutrition which can enhance toxicity by heavy metals as lead. Moreover, poor people often rely on household remedies and folk practices to treat children when they fall ill. World health Organization has stated that the burden of lead toxicity in highest in low income countries.

Our study revealed significant negative correlation between BLL and IQ. We found an increase in BLL by 10ug/dl was associated with decrease in IQ by 2.35 points. Inorganic lead is indeed a widespread neurotoxin. Most literatures suggest that a unit increase in blood lead from 0 to 10  $\mu\text{g/dl}$  range is more detrimental to children's intellectual functioning than an equivalent increase within the 10 to 20  $\mu\text{g/dl}$  range. However, in our study threshold relationship between BLL and IQ could not be established as BLL affected IQ at all levels.

**Limitations:** Examining all grades or all schools of Kathmandu municipality was not feasible because of time and financial constraints. Our study was limited to school children in Kathmandu, BLL of children in other parts of Nepal may differ. Finally, our study did not measure lead in potential exposure sources, such as soil, dust and air.

20 Sherchand O et al.,

### **Conclusion**

This is the first study done in Kathmandu Municipality to determine blood lead level in children. The data generated through our study established that paints used in Nepal are one of the most significant risk factors of lead exposure. This emphasizes the urgency to plan stern steps by our policy makers to minimize lead content in paints and formulate prevention strategies to overcome present exposure. Furthermore, to ensure success, public awareness about lead, management of existing cases of lead poisoning and assignment of monitoring units need to established.

# **Acknowledgments**

We would also like to acknowledge the schools, their teachers and students for participating in our study. We would like to thank B.P. Koirala Institute of Health Sciences for allowing us to conduct this study. We express our sincerest gratitude to University Grant Commission, Sanothimi for funding our study. We are thankful to SEAM-N laboratory, Biratnagar for allowing us to measure blood lead.

We would also like to acknowledge Mr. Surrendra Uranw (PhD fellow, Kalanet Project, B.P. Koirala Institute of Health Sciences, Dharan) and Mr. Rajeshwor Mehta (Technologist in Man-Mohan Cardiothoracic Surgery, Institute of Medicine, Maharajgung, Kathmandu) and Miss Rekha Dakal (Technician, Institute of Medicine, Maharajgung, Kathmandu) and Mr. Subodh Gupta (Technician, B.P. Koirala Institute of Health Sciences, Dharan) for their invaluable help.

Conflict of interest: None declared.

# References

- 1. Singh, R., Gautam, N., Mishra, A., & Gupta, R. (2011). Heavy metals and living systems: An overview. Indian Journal of Pharmacology, 43(3), 246–253. doi:10.4103/0253-7613.81505
- 2. Gillis et al.: Analysis of lead toxicity in human cells. BMC Genomics 2012 13:344.
- CDC. Infant Lead Poisoning Associated with Use of Tiro, an Eye Cosmetic from Nigeria- Boston, Massachusetts, 2011. MMWR. August 3, 2012/ 61(30):574-576
- Agency for Toxic Substances and Disease Registry. 1999. Toxicological profile for lead. Atlanta: US Department of Health and Human Services, Public Health Service.

- Centers for Disease Control and Prevention. Appendix C.1, the lead laboratory. In: Screening young children for lead poisoning: guidance for state and local public health officials. Atlanta, GA: U.S. Department of Health and Human Services; 1997.
- Yang JS, Kang SK, Park IJ, Rhee KY, Moon YH, Sohn DH. (1996) Lead concentrations in blood among the general population of Korea. International Archives of Occupational and Environmental Health, 68(3):199– 202.
- Mushak P, Davis JM, Crocetti AF, Grant LD. Prenatal and postnatal effects of low-level lead exposure: integrated summary of a report to the U.S. Congress on childhood lead poisoning. Environ Res 50:11–36 (1989).
- Lacasana M, Romieu I, Sanin LH, Palazuelos E, Hernandez-Avila M. Blood lead levels and calcium intake in Mexico City children under five years of age. International Journal of Environmental Health Research 2000;10:331-40.
- Manser WW, Khan MA. Trace element studies on Karachi Populations, Part I: Normal ranges for blood copper, zinc and magnesium for adults. Journal of the Pakistan Medical Association 1989;39:43-9.
- K.C A B. (2010, July 16) Solar Batteries Poisoning Silent Springs in Nepal. New Spotlight. Vol: 04 No. 04. Retrieved from http://www.spotlightnepal.com/News/ Article/-Solar-Batteries-Poisoning-Silent-Springs-in-Nepal.
- 11. Sakata S, Shimizu S, Ogoshi K, Hirai K, Ohno Y, Kishi T, Sherchand JB et al (2007). Inverse relationship between serum erythropoietin and blood lead concentrations in Kathmandu tricycle taxi drivers. Int Arch Occup Environ Health. 80(4):342-5.
- 12. Attina, T. M., & Trasande, L. (2013). Economic costs of childhood lead exposure in low-and middle-income countries. Environmental health perspectives, 121(9), 1097-1102.
- Gottesfeld P, Pokhrel D, Pokhrel AK. (2014). Lead in new paints in Nepal. Environment Research. 132:70-5. doi: 10.1016/j.envres.2014.03.036. Epub 2014 Apr 16. PMID: 24742730 [PubMed - indexed for MEDLINE]

 Iron deficiency anaemia: assessment, prevention, and control. A guide for programme managers. Geneva, World Health Organization, 2001 (WHO/NHD/01.3).

- 15. https://www.healthcare.uiowa.edu/path\_handbook/ Appendix/Heme/PEDIATRIC\_NORMALS.html
- Gottesfeld, P., Pokhrel, D., & Pokhrel, A. K. (2014).
   Lead in new paints in Nepal. Environmental research, 132, 70-75.
- 17. Liu J, Ai Y, McCauley L, Pinto-Martin J, Yan C, Shen X, et al. Blood lead levels and associated sociodemographic factors among preschool children in the South Eastern region of China. Paediatr Perinat Epidemiol. 2012 Jan;26(1):61-9.
- Bannon DI, Abounader R, Lees PS & Bressler JP (2003). Effect of DMT1 knockdown on iron, cadmium, and lead uptake in Caco-2 cells. Am J Physiol Cell Physiol 284, C44–C50. Abstract/FREE Full Text
- 19. Shrestha R A KBK. The Study of Lead Pollution in Street Dust and Drinking Water Along Arniko Rajmarg of Nepal. J Nepal Chem Soc. 2008/2009;23.
- Lanphear, B. P., Dietrich, K., Auinger, P., & Cox, C. (2000). Cognitive deficits associated with blood lead concentrations <10 microg/dL in US children and adolescents. Public Health Reports, 115(6), 521–529.</li>