

Analysis of lead levels in deciduous teeth from children in Clark County, Nevada.

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Abstract

Background: Elevated blood lead levels (EBLL) are declining in the United States, although some population subgroups continue to exhibit significant health disparities. A childhood lead poisoning prevention program was recently started in Nevada, and many efforts have been made to support this program and increase the screening rates.

Methods: To expand the potential pool of children screened for EBLLs, a pilot study was performed to

evaluate lead concentrations in extracted deciduous teeth using Graphite Furnace Atomic Absorption Spectrometry (GFAAS), Inductively Coupled Plasma Mass Spectrometry (ICP-MS), and X-Ray Fluorescence (XRF) analysis.

Results: Lead concentrations as determined by GFAAS and ICP-MS were found to be within normal ranges ($0.585 \text{ ppm} \pm 0.022$) and were similar to previous studies. Hispanic patients exhibited higher lead levels ($0.580 \text{ ppm} \pm 0.032$) than Black ($0.478 \text{ ppm} \pm 0.051$) patients, and were significantly higher than White ($0.275 \text{ ppm} \pm 0.035$) patients ($p < 0.05$). Analysis of a small number of matched saliva samples, however, found no evidence for acute lead poisoning. Although limited by a small initial sample size ($n=22$), this pilot study provides evidence that teeth can be effectively used to reveal lead exposure in pediatric dentistry patients.

Key words: dentistry, teeth, lead levels

Introduction

Although the rates of elevated blood lead levels (EBLLs) in the United States (US) have declined significantly over the past few decades, these declines have not been consistent or uniform (Rischitelli, Nygren, Bougatsos, Freeman, & Helfand, 2006). National surveillance reports from the Centers for Disease Control and Prevention (CDC) have found consistent year-to-year decreases in the incidence of EBLL among children less than 72 months of age, from a high of 7.61% in 1997 to 1.00% in the latest reported data from 2007 (CDC, 2010). Many public health officials attribute these declines to the restriction of lead in gasoline, which began in 1972 and was completed in 1995 (Silbergeld, 1997), as well as the banning of lead in retail home paints in the 1970s and the subsequent abatement programs employed by the Environmental Protection Agency (EPA), the Occupational Safety and Health Administration (OSHA), the Food and Drug Administration (FDA), the Department of Housing and Urban Development (HUD), and the CDC.

Despite the overall decreases in EBLLs, childhood lead exposure among some specific populations has continued to rise despite these measures, as reported by the Government Accountability Office (GAO) (US GAO, 2010). This report found EBLLs were more likely among children from lower socioeconomic status (SES), among minority children, and those living in older housing units, primarily built prior to 1978. These findings are supported by reviews and epidemiologic evidence, which have demonstrated that pediatric lead

poisoning occurs primarily due to ingestion of lead from environmental sources, including paint chips, dust, soil, toys, ceramics, home remedies, and household medication (Gorospe & Gerstenberger, 2008; Landrigan & Todd, 1994; Needleman, 1998).

Over two thirds of Nevada's population (71.9%), and nearly three-quarters of all children aged five years or less (74.0%), reside within Clark County (US Census Bureau, 2010). An evaluation of traditional sources of environmental lead contamination has revealed that within Clark County, approximately one-fifth (18%) of all homes were built prior to 1978, and are likely to have lead-containing paint, paint chips, dust and soil that may be a source of lead exposure for young children (Gorospe & Gerstenberger, 2008). These older homes serve as residences and housing for more than 40,000 low-income, low SES, and minority (including Hispanic) residents (Clark County, 2004). Because these homes are disproportionately occupied by low-income, low-SES, minority families, a large-scale lead screening project was performed between August 2004 and May 2005. This study has revealed that Hispanics accounted for 26% of all EBLs and 86% of all childhood lead-poisoning cases, as defined by the CDC (EBL > 10 μ g/dL) (Gorospe & Gerstenberger, 2008).

These data suggest a great need to improve the number of children tested for lead, including minorities and those from lower-income neighborhoods, to improve data collection, monitoring, and on-going prevention and abatement programs in Southern Nevada. Research within minority and Hispanic populations of the area has demonstrated atypical sources of lead poisoning. These sources include lead-contaminated candies and home remedies (Gerstenberger, Savage, Sellers, Zupnik, & Gorospe, 2007; Gorospe & Gerstenberger, 2008).

Lead exposures may have a wide range of effects in children and can affect many organ systems, including the renal and nervous system – leading to neuropsychological deficits, delayed development, diminished intelligence, altered behaviors, seizures, coma, and even death (Landrigan & Todd, 1994). To improve overall childhood health and development, the CDC has called for the elimination of all health disparities by the year 2010 through the national Healthy People objectives (Hogan, Njoroge, Durant, & Ferre, 2001). Despite the development of the CDC's Childhood Lead Poisoning and Prevention Program (CLPPP), an estimated 310,000 children in the US under five years of age have EBLs, and

more than 31,000 were identified in 2007 alone (CDC, 2010; Warniment, Tsang & Galazka, 2010). These statistics have prompted many states to develop plans for determining the prevalence of EBL, as well as reducing, preventing, and eliminating childhood lead poisoning. This includes Nevada, which was awarded a CDC grant through the Southern Nevada Health District (SNHD) to screen for childhood lead poisoning and implement prevention programs (Gerstenberger, Savage, Sellers, Zupnik, & Gorospe, 2007).

Although these efforts have led to some initial success in providing screening and education programs, many families, children, and homes have yet to be screened. Also, a significant proportion of these families may not be receiving routine medical care due to lack of funds or insurance, and may be missed by traditional, office-based screening and education programs. Many families report not having visited a physician or other health care provider within the past year, even for routine exams or standard vaccinations (UNLV SDM, 2010). In addition, these studies have demonstrated that despite the presence of screening programs, many minority and Hispanic populations are reluctant to submit to blood-based lead testing (Gerstenberger, Savage, Sellers, Zupnik, & Gorospe, 2007; Gorospe & Gerstenberger, 2008). Based upon these findings, researchers have been evaluating other biological materials which can accurately assess accumulated lead exposures in children.

Previous studies have demonstrated that lead accumulation can be determined from analysis of enamel from deciduous (primary) teeth by various means, including graphite furnace atomic absorption spectrometry (GFAAS), Inductively Coupled Plasma Mass Spectrometry (ICP-MS), and X-ray fluorescence (XRF) (Arruda-Neto et al., 2009; Costa de Almeida et al., 2007; de Almeida, de Souza Guerra, Tanus-Santos, Barbosa, & Gerlach, 2008; de Souza Guerra et al., 2010; Gomes et al., 2004; Polido Kaneshiro Olympio et al., 2010). Similarly, other studies have successfully demonstrated that lead accumulation can also be quantified through analysis of whole, extracted deciduous teeth using these same analysis methods, greatly reducing the required technology and infrastructure for these studies (Hernández-Guerrero, Jiménez-Farfán, Belmont, Ledesma-Montes, & Baez, 2004; Malara, Kwapulinski, & Malara, 2006; Tvinnereim, Eide, & Riise, 2000).

The primary goal of this pilot study was to screen for lead exposure using deciduous teeth extracted from

children seeking routine treatment in the University of Nevada, Las Vegas – School of Dental Medicine (UNLV-SDM) Pediatric Dentistry clinic. UNLV-SDM has a large patient population, serving more than 87,000 unique Medicaid patients, as well as other low-income and uninsured patients – who frequently report UNLV-SDM as the only medical provider they have visited within the past twelve months (UNLV SDM, 2010). The UNLV-SDM patient clinic may therefore represent an additional, potential site for providing lead screening and education to children seeking care for routine dental care or painful dental complications.

Methods

Human Subjects

The protocol for this study titled “Evaluation of Metal Levels (Specifically Targeting Lead) In Extracted Primary Teeth Among Pediatric Patients” was filed, amended, and approved by the University of Nevada Las Vegas (UNLV) Office of Research Integrity – Human Subjects (OPRS#1002-3362) on March 25, 2010. In brief, parents and subjects were recruited by members of the UNLV-SDM Pediatric Clinic during their child’s dental visit. Informed Consent / Parent Permission was required and was conducted onsite, as well as an Assent to Participate in Research for children over the age of seven who were capable of reading.

Protocol

In brief, if the extraction of a deciduous tooth was deemed medically necessary and parental consent was obtained (as well as the Assent to Participate for patients who were over seven years of age), each specimen was labeled and each tooth assigned a unique, randomly-generated number to prevent research bias. The specimens were stored in lead-free plastic tubes with approximately 5mL of distilled water at room temperature. Specimens were then transferred to the UNLV Environmental and Occupation Health Laboratory for analysis. The condition and appearance of each specimen, as well as the tooth number –using the Primary Universal

Numbering System - Primary Teeth (Allan, 1969), were recorded before cleaning and analysis.

The UNLV-SDM Pediatric Clinic regularly performs childhood tooth extractions each week, which allowed for the selection of only deciduous teeth that were whole, minimally decayed, or slightly damaged. Sex and age of participants, as well as the position of the tooth and tooth type were included in the statistical analysis. However, a potential technical limitation to this study included the variation of mineral composition in different tooth types within any one individual.

Graphite Furnace Atomic Absorption Spectrometry (GFAAS): Lead (Pb) analysis

Some samples (n=10) were analyzed by GFAAS. The removal of all external sources of contamination included the lysis of any attached tissue (gingival, gum) in 3% hydrogen peroxide, followed by sonication in a 2% detergent solution to remove oil residue, a brief rinse in 2% nitric acid, and a final rinse with distilled water (Millipore). Teeth were then dried in a 65°C oven for 12 hours. After recording the dry weight of each specimen, each tooth was added to a 50 mL metals-free polypropylene tube with 45 to 50 mL of 10% nitric acid. A reagent blank and National Institute of Standards and Technology (NIST) bone meal (Standard Reference Material 1486) was included with each sample set for quality control.

All digested solutions were brought to a standard volume of 50 mL and stored at room temperature until analysis on the Perkin Elmer Graphite Furnace AAnalyst 600. In brief, a standard curve for each sample run was created by the instrument from a 50 ppb sample created from a Perkin Elmer 1,000 ppm lead standard solution (Table 1). The optical alignment of the furnace and conditioning of graphite tube were performed before the first run of the day. All samples were analyzed in triplicate with the mean measured concentrations used for calculations.

Table 1. GFAAS Program for the Analysis of Deciduous Teeth

Step	Temperature (°C)	Ramp Time (s)	Hold Time (s)	Gas Flow (mL/min)
Dry 1	110	1	30	250
Dry 2	130	15	30	250
Pyrolysis	850	15	20	250
Atomize	1600	0	5	0
Clean-out	2450	1	3	250

Injection Temperature= 20°C
 Minimum Correlation Coefficient= 0.995000

Inductively Coupled Plasma-Mass Spectrometry (ICP-MS): Lead (Pb) analysis

The remaining samples (n=12) were analyzed commercially using Inductively Coupled Plasma Mass Spectrometry (ICP-MS) performed by Exova (Santa Fe Springs, CA). The deciduous teeth, and two patient-matched saliva samples, were analyzed by ICP-MS at Exova (Santa Fe Springs, California). Cleaning and pretreatment of deciduous teeth prior to digestion is the same as samples prepared for GFAAS analysis. Each deciduous tooth was digested in 1 mL of nitric acid and heated to 110°C on a heating block for one hour. During the digestion, 0.5 mL of 30% hydrogen peroxide was added to the solution. The sample completely dissolved. Prior to analysis the sample solutions were brought to a standard weight of 10g with nanopure water. Saliva samples were digested by adding 5 g of a solution of 1% nitric acid and 3% hydrochloric acid to 0.5g of saliva. Quality control measures included a laboratory fortified blank, matrix spike recovery, and internal standard. Spike recoveries ranged from 95% to 101%.

X-Ray Fluorescence (XRF): Lead (Pb) analysis

Determination of lead composition for some samples was performed using a Niton XRFR Analyzer at the UNLV Environmental and Occupation Health Laboratory. The removal of all external sources of contamination from the extracted deciduous teeth included the lysis of any attached tissue (gingival, gum) in 30% hydrogen peroxide, followed by sonication in a 2% detergent solution to remove oil residue, and a final rinse with distilled water (Millipore). Whole teeth were then dried in a 60°C oven for six hours. After recording the dry weight of each specimen, each tooth was analyzed by XRF and limit of detection (LOD) for each sample reported.

Saliva Collection Protocol

In brief, some patients who agreed to participate were also asked to donate a saliva sample for analysis, which would be matched with the extracted deciduous tooth sample. An increasing number of clinical tests now routinely use saliva instead of blood, not only because of reduced risks and the ease of collection, but also for the number, frequency, and volume of samples that can be collected from any one individual (Bergdahl & Skerfving, 2008). Although teeth can reveal past exposure to environmental pollutants and contaminants (including lead), new evidence has demonstrated that saliva and other non-invasively collected bioindicators can provide relevant biological information about current lead levels in tissues or organs (Bergdahl & Skerfving, 2008). No current protocols involving phlebotomy or finger-stick tests were approved or available for this initial pilot study.

Patients who agreed to donate the matched saliva sample were then asked to chew on a small piece of paraffin wax for one minute and then to expectorate. Participants were given a small saliva collection container, 50 mL sterile polypropylene tube, Fisher Scientific (Fair Lawn, NJ). Each saliva sample was assigned the matching unique, randomly-generated number to the extracted tooth sample in order to facilitate research comparison and analysis. Samples were stored on ice until transport to a biomedical laboratory for analysis.

Statistics

Mean concentrations of lead from the study sample, and sample subgroups were calculated and reported, as was standard error (S.E.). The *t*-test is a commonly employed statistical procedure used to infer whether differences exist between the means of two population samples or subgroups. The differences between sample population subgroups were therefore measured using a *t* distribution, $\alpha = 0.05$. All samples were analyzed using two-tailed *t*-tests as departure from normality can make more of a difference in a one-tailed than in a two-tailed *t*-test. As long as the sample size is even moderate (>20), quite severe departures from normality make little practical difference in the conclusions reached from these analyses (Hays, 1994).

However, analyses involving multiple two sample *t*-tests have a higher probability of Type I error, leading to false rejection of the null hypothesis, H_0 . To confirm the effects observed from these experiments and minimize the possibility of Type I error, further analysis of the data was facilitated using ANOVA with SPSS (Chicago, IL) to more accurately assess relationships and statistical significance among and between groups.

Correlation

Analysis of simple linear correlation considers the relationship between two variables. Based upon this understanding, Pearson's correlations were performed to analyze the strength and direction of association between individual patient ages (independent variable) and measurable lead concentrations (dependent variable). Pearson's R or correlation coefficients measure the strength of linear relationships and were interpreted using the following:

- -1.0 to -0.7 strong negative association
- -0.7 to -0.3 negative association
- -0.3 to +0.3 little or no association
- +0.3 to +0.7 positive association
- +0.7 to +1.0 strong positive association

Regression

If there is a logical relationship that implies functional dependence of one variable on another, linear regression can help to determine the magnitude of the effect of one variable on another (Hays, 1994). Based upon this understanding, linear regression (R^2) was calculated to determine the relative contribution of the independent variable (age) to the dependent variable (lead concentration) assessed. Finally, statistical power (p) was calculated to determine the applicability of these results to other study populations ($\alpha = 0.05$; number of predictors not including regression constant = 1).

Results

A total of fifty nine (59) samples were collected from the UNLV-SDM pediatric patient clinic between March and October, 2010. The patients were randomly selected for participation in this pilot study from a pool of pediatric patients who were treated on one of ten clinic sampling dates. Several samples were discarded because of their poor physical condition or other confounding factors, such as the presence of amalgam restorations or extensive root resorption, yielding a final sample of teeth from 22 unique patients, which were subsequently screened for the presence of lead (Pb). The results of the GFAAS and ICP-MS analyses revealed an average lead concentration of $0.585 \text{ ppm} \pm 0.022$ (S.E.) from these samples (Table 2).

Table 2. Analysis of UNLV-SDM study samples for lead using GFAAS and ICP-MS.

Sample ID	Screening method	Pb (ppm)	Tooth	Sex	Age	Race
1133	GFAAS	0.225	Molar	M	3	Hispanic
1133	GFAAS	0.225	Molar	M	3	Hispanic
4202	GFAAS	0.346	Molar	F	8	Hispanic
3677	GFAAS	0.385	Molar	F	7	White
5084	GFAAS	0.322	Molar	F	5	Hispanic
4771	GFAAS	1.282	Molar	F	6	Hispanic
7565	GFAAS	1.289	Molar	F	11	Hispanic
8428	GFAAS	1.232	Molar	F	8	Hispanic
5684	GFAAS	0.565	Molar	F	5	Black
5510	GFAAS	0.730	Molar	M	7	Hispanic
9651	GFAAS	0.17	Incisor	F	10	White
8304	ICP-MS	0.260	Incisor	F	5	Hispanic
7319	ICP-MS	0.450	Incisor	M	5	Hispanic
2466	ICP-MS	0.10	Molar	M	10	Other
2305	ICP-MS	0.27	Molar	M	3	Hispanic
2356	ICP-MS	0.27	Incisor	F	5	White
7653	ICP-MS	0.30	Molar	F	5	Black
2239	ICP-MS	0.57	Incisor	F	7	Black

Table 2
Continued

2788	ICP-MS	0.40	Molar	M	10	Hispanic
2986	ICP-MS	0.35	Molar	M	12	Hispanic
2075	ICP-MS	0.27	Incisor	M	11	Hispanic
2051	ICP-MS	0.33	Molar	M	13	Hispanic
2887	ICP-MS	1.60	Molar	F	10	Hispanic
		Average (ppm)	S.E.	n	Age range	<i>p</i> -value
Total		0.585	0.022	22	3 - 13	
Sorted by Sex						
	Females	0.702	0.038	13	5 - 11	<i>p</i> = 0.14
	Males	0.428	0.031	9	3 - 13	
Sorted by Race						
	Hispanic	0.580	0.032	15	3- 13	
	Black	0.478	0.051	3	5 - 7	<i>p</i> = 0.36
	White	0.275	0.035	3	5 - 10	<i>p</i> = 0.02*
Sorted by Tooth						
	Molar	0.608	0.029	16	3 - 13	
	Incisor	0.331	0.025	6	5 - 11	<i>p</i> = 0.07

The average lead concentration in teeth extracted from females (0.702 ppm \pm 0.038) was measurably higher than the average for males (0.428 ppm \pm 0.031) and the entire sample (0.585 ppm \pm 0.022), but was not statistically significant (p = 0.14 or > 0.05). Similarly, small differences were observed between lead concentrations from different tooth types, such as molars (0.608 ppm \pm 0.029) and incisors (0.331 \pm 0.025), although these differences were not sufficient to reach statistical significance (p = 0.07 or > 0.05). Lead concentrations among Hispanic patients (0.580 ppm \pm 0.032), however were significantly higher than those from White patients (0.275 ppm \pm 0.035) (p = 0.02 or < 0.05), but not Black patients (0.478 ppm \pm 0.051) (p = 0.36 or > 0.05).

The analysis for lead concentration revealed the differences and patterns between lead concentrations from different genders, tooth types and patients of different racial backgrounds (Figure 1). Although the observed differences between the genders and the tooth types were not statistically significant, a small subset of these samples with higher values appeared to coincide with the highest values observed among some Hispanic patients. When these data were cross-referenced with the individual data points from Table 2, this revealed four samples with much higher lead concentrations (Pb > 1.20 ppm), each collected from Hispanic female patients. In addition to being Hispanic and female, these four patients were slightly older (8.75 years) than the entire sample (7.54 years), although this difference was not statistically significant (p = 0.386).

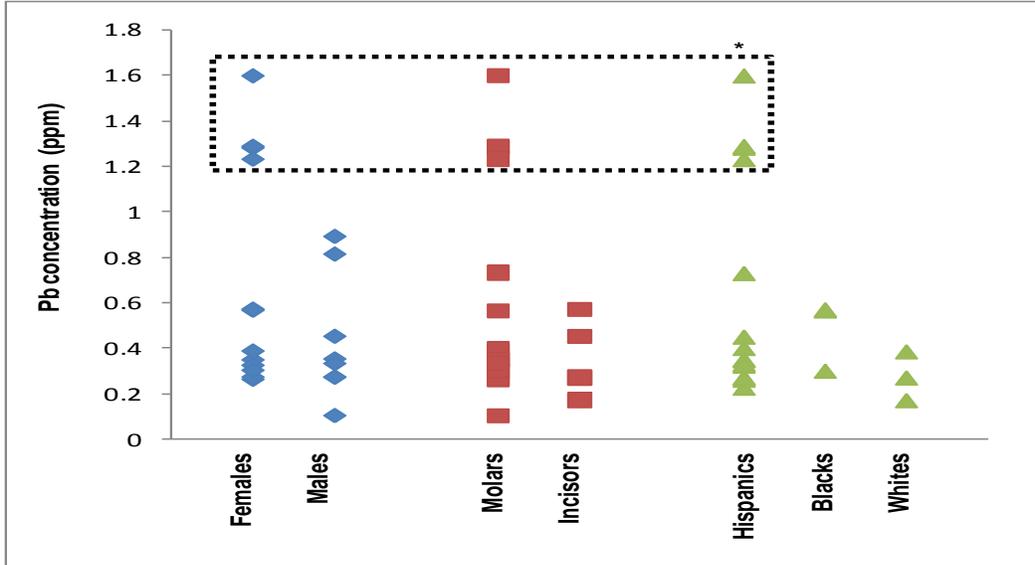


Figure 1. Comparison of lead concentrations in patients by gender, tooth type, and ethnicity.

No statistically significant differences were found between genders or tooth types. Hispanics, however, had higher average lead levels ($0.580 \text{ ppm} \pm 0.032$) than Black ($0.478 \text{ ppm} \pm 0.051$) or White ($0.275 \text{ ppm} \pm 0.035$) patients ($p < 0.05$). In addition, the four samples with the highest lead concentrations ($\text{Pb} > 1.20 \text{ ppm}$), were collected from slightly older Hispanic female patients (average = 8.75 years).

However, to investigate the possible effects of age on lead concentrations found among these samples due to accumulated exposures over time, samples from

patients of similar ages were plotted against the mean concentration of lead from those samples (Figure 2). These results revealed a strong, positive (linear, age-dependent) relationship with older patients exhibiting higher average lead concentrations than younger patients ($R = 0.852$). Linear regression revealed that age was a significant factor for predicting lead concentrations ($R^2 = 0.7259$) among patients from this sample. For example, three and five year old patients (0.245 and 0.362 ppm , respectively) had significantly lower lead levels than seven (0.567 ppm), ten (0.5675 ppm) or eleven (0.7795 ppm) year olds ($p > 0.05$).

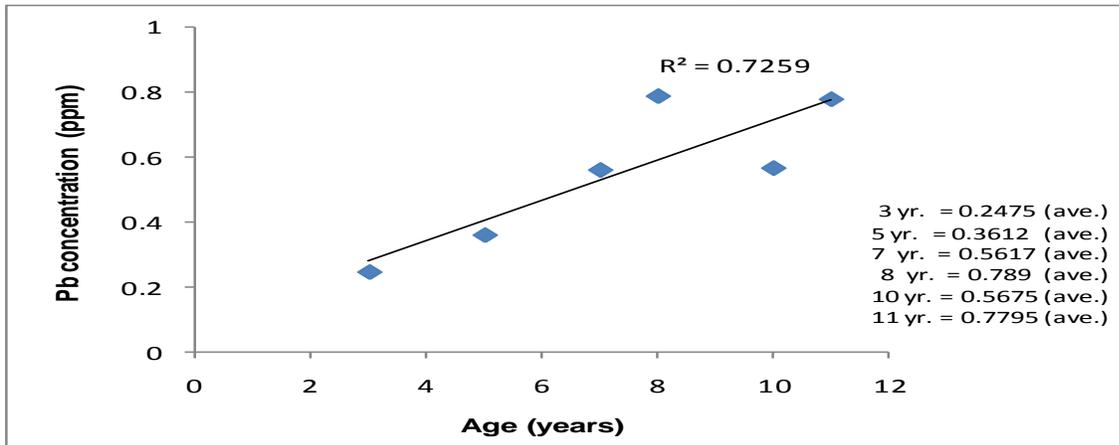


Figure 2. Average lead concentrations as a function of age.

A strong, positive, age-dependent relationship was observed ($R = 0.852$). Regression analysis confirmed age was a significant factor for predicting lead

concentrations ($R^2=0.7259$) among patients from the sample ($p>.05$).

To more accurately assess the validity and sensitivity of these results, several samples were selected at random to be screened for intra-sample and method-specific variability (Table 3). A total of three patients had multiple teeth of similar types (molars) extracted on the same day, which were then screened for lead content. These results revealed similar concentrations of lead from different teeth extracted from the same patient (Table 3, *Intrapersonal comparisons*). To evaluate whether these patients had current, circulating lead levels (acute lead

poisoning), which would not be revealed through accumulated lead deposits from extracted teeth, some patients were asked to provide saliva samples for comparison. Two subjects and their parents gave consent for this analysis and the results demonstrated that although there were measurable concentrations of lead found in both of the analyzed tooth samples (0.260 and 0.450 ppm, respectively), lead levels in saliva were below the ICP-MS detection limits (< 0.005) (Table 3, *Biometric comparisons*).

Table 3. Intra-sample comparisons from UNLV-SDM study

Sample ID	Screening method	Pb (ppm)	Tooth	Sex	Age	Race
<i>Intrapersonal comparisons</i>						
2051A	ICP-MS	0.33	Molar (I)	M	13	Hispanic
2051B	ICP-MS	0.35	Molar (J)			
2986A	ICP-MS	0.32	Molar (A)	M	12	Hispanic
2986B	ICP-MS	0.35	Molar (J)			
2305A	ICP-MS	0.44	Molar (B)	M	3	Hispanic
2305C	ICP-MS	0.27	Molar (I)			
<i>Biometric comparisons</i>						
8304	ICP-MS	0.260	Incisor (D)	F	5	Hispanic
8304*	ICP-MS	<0.005	Saliva			
7319	ICP-MS	0.450	Incisor (G)	M	5	Hispanic
7319*	ICP-MS	<0.001	Saliva			
<i>Screening method comparison</i>						
9651	GFAAS	0.17	Incisor (E)	F	10	White
9651	XRF	<13.3 (LOD)	Incisor (E)			

In addition, a small subset of the sample was processed using a different lead sampling method, XRF analysis. A subset of six samples were screened

using this procedure and the resulting analysis revealed lead concentrations less than the limit of detection (LOD), within the range of 12.4 and 14.1

ppm (Average 13.116 ppm \pm 0.022 S.E.). Due to the reduced sensitivity from this type of analysis, one sample was screened using both the GFAAS (0.17 ppm) and XRF analysis methods ($<$ 13.3 ppm LOD), which clearly demonstrated the increased sensitivity and lower detection limit of the GFAAS and ICP-MS as confirmatory methods.

Discussion

This pilot study screened for environmental lead exposure from deciduous teeth of children at the UNLV-SDM Pediatric Dentistry Clinic using GFAAS and ICP-MS analysis. These data suggest a strong, positive association ($R^2 = 0.7259$) between accumulated lead exposures and race. More specifically, lead exposures were highest in at least some Hispanic children, a group previously demonstrated to be at elevated risk for lead exposure from atypical sources of lead poisoning, including lead-contaminated candies and home remedies specific to Clark County, Nevada (Gerstenberger, Savage, Sellers, Zupnik, & Gorospe, 2007; Gorospe & Gerstenberger, 2008).

Although the steady and continuous decline in the incidence of elevated blood levels among children in the US has been viewed as a positive outcome by public health professionals, there remain significant geographic, ethnic, and demographic sub-groups that continue to experience health disparities and childhood lead poisoning (CDC, 2010; Rischitelli, Nygren, Bougatsos, Freeman, & Helfand, 2006). To address these on-going health disparities, the CDC has implemented the CLPPP to screen for, evaluate the incidence of, reduce, and ultimately prevent environmental lead exposure among children in the US. Many states and metropolitan areas, including Clark County, Nevada are implementing more comprehensive lead screening and prevention programs to facilitate these efforts.

These results are comparable to other findings, such as a recent study in Brazil that found lead concentrations from extracted teeth were lower among non-exposed, control populations (0.91 ppm \pm 0.12) than among exposed (elevated lead concentration or ELC) populations (1.28 ppm \pm 0.14) (Arruda Neto et al., 2009). The results of the current study revealed that average, overall lead concentrations were slightly lower (0.585 ppm \pm 0.022), but that at least four samples from Hispanic females had comparable lead levels to the ELC populations observed in the previous study (Pb $>$ 1.20 ppm).

Despite these similarities, other studies from Brazil have found much higher lead concentrations in teeth from children in exposed and non-exposed areas (Costa de Almeida et al., 2007; Gomes et al., 2004). More specifically, these studies found lead content in teeth from non-exposed children were between 118 and 106 ppm, while exposed children had average lead concentrations of 169 and 786 ppm, respectively. Although these results may seem striking due to their much higher lead levels, it is important to remember that these studies were conducted in areas where industrial or other large-scale production activities may occur and environmental protection standards may have been limited.

Although these results had statistical power ($P = 0.88$) to generalize the applicability of these findings, there were a number of limitations that must be considered. First among these concerns is the limited sample size for this initial pilot study, which may not have been large enough to be representative of the UNLV-SDM Pediatric Clinic population or the larger population of Clark County, Nevada. However, finding four patients with elevated lead burdens from such a limited sample size strongly suggests that continuation of this study may help to uncover many more patients in need of this type screening. The relative ease of obtaining and testing extracted deciduous teeth as a bioindicator for lead exposure suggests this may represent an effective alternative method for testing children.

These results provide a compelling rationale for continuation and expansion of this project, given that the UNLV-SDM Pediatric Clinic performs an average of 60 unique patient tooth extractions per week. Given the relatively low percentage of children in Clark County, NV screened through the Southern Nevada Health District's childhood lead poisoning and prevention programs (Gerstenberger, Savage, Sellers, Zupnik, & Gorospe, 2007), the implications of this study are clear and unambiguous. This potential pool of pediatric dental patients may be able to increase the screening potential of the CDC and SNHD lead screening programs to include more than 3,000 previously unscreened children with Clark County, Nevada each year. Moreover, this project is congruent with the overall mission of the UNLV-SDM Clinic, which strives to serve the local community by treating and educating patients that are primarily low-income, uninsured, under-insured, or Medicaid dependent. Recent estimates suggest that nearly 56,000 children in Clark County, Nevada currently reside in uninsured and lower-income

households and are more likely to live in conditions of poverty (Rothweiler, Cabb & Gerstenberger, 2007).

In addition to these benefits, continuation of this project may permit the incorporation of health promotion efforts consistent with the CLPPP in a clinic population, which is the target population for this program. In the event that an elevated lead concentration is identified from an extracted tooth, the patient can be referred to the CLPPP to have a capillary (finger stick) test completed for minimal or no cost at the SNHD, which is located across the street from the UNLV-SDM Clinic. This protocol is consistent with the criteria set forth by the federal government regarding lead screening, thus representing a novel conduit for screening and referring new patients.

The continuation and expansion of this project to measure and evaluate lead from extracted teeth would further the efforts of the CDC and the SNHD to reach these children, as well as providing novel sites for lead education, screening, prevention and intervention efforts in collaboration with the School of Community Health Sciences and Public Health. Future planned research projects will involve testing the feasibility of other lead detection methods, including previously employed tooth biopsy methods, as well as expanding the evaluation of other non-invasive biomaterials, such as salivary fluids, to provide information about current, circulating lead levels and acute lead poisoning.

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