LEAD LEVELS IN DENTINE AND CIRCUMPULPAL DENTINE OF DECIDUOUS TEETH OF NORMAL AND LEAD POISONED CHILDREN

IRVING M. SHAPIRO, BRUCE DOBKIN, ORHAN C. TUNCAY AND HERBERT L. NEEDLEMAN

Center for Oral Health Research and School of Dental Medicine, University of Pennsylvania, 4001 Spruce Street, Philadelphia, Pennsylvania 19174; Department of Psychiatry, Harvard Medical School, Boston, Massachusetts and School of Medicine, Temple University, Philadelphia, Pennsylvania (U.S.A.)

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SUMMARY

A method for the preparation of circumpulpal dentine for lead analysis is described. The technique consists of cutting 600-μ-thin sections of deciduous teeth, and, utilizing a vice, fracturing off the circumpulpal dentine from the coronal dentine. Both dentine and circumpulpal dentine of teeth of normal American children and previously identified lead-poisoned children were prepared in this fashion and analyzed for lead. Comparing the lead-poisoned teeth with normal teeth, the results indicate that a 6-fold difference in the mean lead content existed between the two groups. The uptake of lead by the two dentine zones at low levels of lead intake was also examined. Thus, comparing tooth lead levels of normal American and Icelandic children, 2–3 times less lead was found in the teeth of Icelandic children. The sensitivity of the dentinal zones for lead, the simplicity of the preparative method and the ease of collection of the teeth supports the use of teeth as useful indicators of previous lead exposure.

INTRODUCTION

It is now recognized that there are certain limitations to the use of blood lead measurements in the diagnosis of plumbism. These are mainly due to the transitory nature on the lead ion in blood and to the uptake of lead by other tissues of the body. Realizing the limitations of blood analyses, hair and nail have been studied for their ability to bind lead. However, the problem of exogeneous lead contamination has restricted their use in the diagnosis of plumbism.

The practical problems inherent in the use of bone as a lead biopsy tissue inhibits its use as an indicator of lead ingestion. However, it occurred to us that as teeth sequester lead and are not contaminated by external lead sources, then they would be better indicators of both high and low levels of lead exposure than either blood,
hair or nail. With this in mind, we have utilized primary teeth to identify children previously exposed to lead. Employing whole deciduous teeth of children who resided in the "lead belt" of urban Philadelphia, we demonstrated a significant elevation in whole tooth lead levels compared with suburban controls. When the distribution of lead in these teeth was studied by chemical and electron probe analyses, we noted that while lead was present in coronal dentine, it was concentrated in the dentine layer immediately adjacent to the pulp (circumpulpal dentine). It was concluded from these studies that a much clearer insight into the accumulation of lead could be obtained by studying not the whole tooth, but selected areas of dentine. We here describe a method for preparing dentine and circumpulpal dentine for lead analysis and report the lead levels of these areas in normal and lead poisoned children.

EXPERIMENTAL

Eleven children recovered from unequivocal lead poisoning were identified from records of three children's hospital in Philadelphia and Boston and their shed deciduous teeth were collected. Control teeth were collected from children in three second grade classes in suburban Boston primary schools and from children residing in Reykjavik and Akranes in Iceland.

The teeth were cleansed mechanically, embedded in self-curing acrylic and longitudinal sections were prepared on a Bronwill Thin Sectioning Machine (VWR Scientific, Baltimore, Maryland) utilizing two 4-inch radius diamond cutting discs separated from each other by a 600-µ-thick copper spacer. The diamond wheels were water cooled and rotated at 5000 rpm. The sectioning time was 4 min.

About 500 tooth sections were examined microscopically. Between 85 and 95% showed a well delineated zone of secondary dentine. The thickness of this zone ranged from about 150 to 250 µ. Initially, the area of the secondary dentine in each sample was measured microscopically and the lead content of the secondary dentine determined. However, this step was later omitted when it was found that the lead content of the 300-µ zone (the circumpulpal dentine) closely followed that of the secondary dentine.

A Tube Cap Vice (Beckman Instruments, Palo Alto, California) was pre-calibrated to a depth of 300 µ. Under a dissecting microscope, the dentine sections were inserted in the vice such that the circumpulpal dentine was rigidly clamped while the coronal dentine and enamel were free (see Figs. 1 and 2). Pressure was then applied to the free edge of the protruding tissue to produce a fracture, at the junction of the free and clamped tissue, which ran along the slice in a direction which was parallel to the long axis of the secondary dentine. The circumpulpal dentine and the residual coronal dentine were dried for 16 h at 60° and then weighed on a micro-balance. The samples were dissolved in 0.1 ml 70% HClO4 (Aristar grade, B.D.H. Ltd., White Plains, New York) and 5 ml of highly purified 1 M sodium acetate subsequently added. The samples were analyzed by Anodic Stripping Voltammetry (Environmental Sciences Associates, Burlington, Massachusetts). The plating time was 10 min, the sweep rate 60 mV/sec-div. at a plating potential of 1.05 V; the current was varied from 100 to 500 µA.
Fig. 1. Photograph of dissecting microscope and vice (V).

RESULTS AND DISCUSSION

Following the procedure described, it was found possible for one trained person to prepare and analyze about 40 teeth a week. With further modifications, partic-
ularly at the sectioning stage, where considerable time is required to section each tooth, there can be little doubt that the deciduous tooth could be utilized for routine large scale lead screening. Even so, the method in its present form permits the dental tissue to be used as investigative tissue for studying retrospective lead uptake at both high and low lead intake levels.

Of the tissues studied, the coronal dentine represents a tissue that calcifies before tooth eruption while the secondary dentine is a zone of dentine that does not commence to form until after eruption takes place. Lead in the secondary dentine zone, therefore, represents the exposure of the body to lead during a fixed and usually known time period, i.e. from eruption to exfoliation.

The variability of the method to utilize deciduous tooth to monitor ingested lead is shown in Table I. Of the teeth analyzed, four were obtained from normal children (subject B and C) and four were from a child who had been treated for lead poisoning (subject A). While variations in the level of lead in dentine and circumpulpal dentine in each of the individual subjects were noted, the magnitude of the variation was small when compared to the mean lead concentrations of normal and lead poisoned teeth. Due to the nature of the experimental system it was not possible to determine

<table>
<thead>
<tr>
<th>Subject</th>
<th>Tooth</th>
<th>Dentine (% Pb)</th>
<th>Circumpulpal dentine (% Pb)</th>
</tr>
</thead>
<tbody>
<tr>
<td>A (known lead poisoned)</td>
<td>Incisor</td>
<td>102.9</td>
<td>389.2</td>
</tr>
<tr>
<td></td>
<td>Canine</td>
<td>141.6</td>
<td>508.9</td>
</tr>
<tr>
<td></td>
<td>Canine</td>
<td>121.2</td>
<td>798.4</td>
</tr>
<tr>
<td></td>
<td>Canine</td>
<td>91.2</td>
<td>653.2</td>
</tr>
<tr>
<td></td>
<td>Molar</td>
<td>226.9</td>
<td>793.3</td>
</tr>
<tr>
<td></td>
<td>201.7</td>
<td>665.8</td>
<td></td>
</tr>
<tr>
<td>B (normal)</td>
<td>Incisor</td>
<td>7.1</td>
<td>—</td>
</tr>
<tr>
<td></td>
<td>Incisor</td>
<td>12.4</td>
<td>49.1</td>
</tr>
<tr>
<td>C (normal)</td>
<td>Incisor</td>
<td>6.9</td>
<td>39.5</td>
</tr>
<tr>
<td></td>
<td>Incisor</td>
<td>3.5</td>
<td>18.7</td>
</tr>
</tbody>
</table>
whether the variation was due to the preparative and analytical procedures employed or to a differential uptake of lead by the teeth.

Table II indicates the mean lead concentration of dentine and circumpulpal dentine of teeth of lead-poisoned and normal children. The 7-fold difference in mean circumpulpal dentine lead from (American) control and poisoned children is considerably greater than that described for whole tooth analysis\(^1,4\) and underlines the importance of selected zonal analysis of dentine. The data also suggest that the tooth is responsive to lead accumulation at low as well as high levels of lead exposure. Cumulation of dentinal lead by the control groups indicates that although these teeth contain low concentrations of lead, a 2-3-fold difference exists between American and Icelandic teeth. While detailed information concerning the origin of lead in these teeth is lacking, the data imply that a significant difference exists between the two populations in the level of intake of this toxic element.

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REFERENCES