The skeleton is the main store of lead in the body, and most research on lead turnover or exchange has been directed towards bone analyses (1). Tooth analyses have been used as an alternative for skeletal lead to identify lead exposure in children (2-4).

Evidence for lead turnover or exchange in calcified tissue in humans is based on 1) relatively crude methods, such as X-ray fluorescence (XRF) measurements on tibia, calcaneus, or phalanges of occupationally exposed workers (5-9); 2) indirect methods based on comparisons of tooth and blood lead levels (3,10,11); and 3) short-term experiments on limited numbers of subjects using radioactive tracers (12) or longer experiments using stable isotopes (13). Rabinowicz (1) used some of the above techniques and calculated that the bulk turnover rates for lead in compact bone were -2%/year and -8%/year for the spine (trabecular bone).

We have shown that in blood of Australian subjects and in environmental samples (diet, air, house dust, gasoline), the $^{206}\text{Pb}/^{204}\text{Pb}$ ratio is <17.0 and the $^{207}\text{Pb}/^{206}\text{Pb}$ ratio is >0.91 (14,15). These ratios are totally different from those prevailing in other countries. Furthermore, we have established that lead in the blood of Australian migrants exchanged rapidly with lead in the Australian environment, especially in the first 3-6 months; even after >12 months residence, approximately 40-70% of lead in their blood was skeletally derived (14). As Australian lead was exchanging with the skeletal lead in the subjects, it was possible that exchange was also occurring in the dentine of teeth. If exchange in teeth was occurring, then there should be a greater contribution of Australian lead to dentine corresponding to residence time in Australia.

Thus, the aims of this pilot study were to determine if exchange of lead occurred in tooth dentine and, if so, at what rate and how this rate compared with that in bone.

Methods

Permanent teeth of Australian migrants were obtained from dental practitioners throughout Sydney. The subjects were mainly from Eastern and Southern Europe. Information regarding country and city of origin, age of subject, and residence time in Australia up until tooth extraction was requested, but not always obtained. Likewise, information regarding residence time in other countries prior to arrival in Australia was not generally available. In one subject (Table 1; Croatia 3 (1546, 1547)) it was possible to analyze trabecular bone attached to the permanent tooth. Deciduous teeth were generally those from children whose mothers were enrolled in a related project; these deciduous teeth were analyzed to provide evidence for the skeletal lead isotopic signature of the mother.

This pilot study was performed in two stages. The initial study of permanent teeth ($n = 18$) and deciduous teeth ($n = 14$) were from subjects from the Commonwealth of Independent States (CIS; the former Soviet Union), the former Yugoslavia, Poland, Bulgaria, and Lebanon. The second stage involved analyses of other permanent teeth ($n = 19$) for subjects from Turkey, Lebanon, Syria, Spain, Greece, Egypt, Italy, Uruguay, the United Kingdom, and Chile.

Deciduous and permanent teeth were analyzed. Deciduous teeth were crowns only, as the roots had been resorbed. The crowns were cut transversely into 1-2-mm thick slices from the incisal and cervical areas using a diamond-impregnated stainless steel disc. The rationale and advantages of using cross-sectional slices were described in Gulson (16).

For permanent teeth, 1-2-mm sections of the outer crown and the root sample were taken from an area approximately 1-2 mm from the root tip (see Fig. 1 in Gulson (16)). In three teeth, the circumpulpal dentine and cementum were removed from slices of the root and compared with contiguous slices in which the circumpulpal dentine and cementum were retained to evaluate the effect of circumpulpal dentine on lead isotopic composition and concentration. In another three teeth, the cementum was removed from slices of root and compared with contiguous slices with retained circumpulpal canal and cementum.

Analytical methods follow those described in Gulson (16). Briefly, from 5 to 50 mg of specially cleaned tooth sections were dissolved in 6 M HCl to which a $^{203}\text{Pb}$ solution of known isotopic composition and concentration was added (the isotope dilution method). Lead was separated by ion exchange chromatography and the isotope

Address correspondence to B.L. Gulson, Graduate School of the Environment, Macquarie University, Sydney, NSW 2109 Australia.

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ratios were measured by thermal ionization mass spectrometry. The precision of the isotopic ratios based on over 1,800 analyses of international standards and natural samples is ± 0.05% (2 Σ) for the \( ^{207}\text{Pb}/^{206}\text{Pb} \) ratio and ± 0.1% for the \( ^{206}\text{Pb}/^{207}\text{Pb} \) ratio. Data for replication of tooth analyses are given in Gulson and Wilson (4) and Gulson (16).

Gulson and Wilson (4) and Gulson (16) demonstrated the use of the lead isotope technique, combined with the well-established histology of teeth, in evaluating in utero and early childhood lead exposure from slices of deciduous teeth. In this approach, analysis of the enamel provides evidence of in utero exposure. Analysis of dentine provides evidence of exposure during the early childhood years, when hand-to-mouth activity is usually an important contributor to lead body burden, and potentially up to the time of tooth exfoliation (2,3).

In children exposed to lead sources from mining, paint, or gasoline in communities such as the Broken Hill lead mining community, Gulson and Wilson (4) and Gulson (16) showed that the source of lead from the incisal sections was different from the source of lead in the cervical sections of deciduous teeth, reflecting the change in lead from in utero exposure to early childhood.

These data also demonstrated that there was minimal exchange of lead in enamel in contrast to dentine. Using the lead in enamel as a fixed parameter, the changes in dentinal lead concentration and isotopic composition can be calculated simply by difference. In this paper, the differences are calculated by \( (^{207}\text{Pb}/^{206}\text{Pb})_{\text{dentine}} - (^{207}\text{Pb}/^{206}\text{Pb})_{\text{enamel}} = \Delta(^{207}\text{Pb}/^{206}\text{Pb}) \).

The theoretical aspects of the lead isotope method are described in Gulson (16).

**Results and Discussion**

Lead isotopic and concentration data for the present investigation are presented in Tables 1 and 2 and Figure 1.

**Variation in rate of exchange with residence time with tooth type.** To estimate the reproducibility of any changes for a given subject, two teeth from four subjects were measured. Reproducibility was excellent for two sets of molars [Croatia 3 (IS46/IS47) and Yugoslavia 1 (IS28) in Table 1]. In spite of quite different lead concentrations, the change in \( ^{207}\text{Pb}/^{206}\text{Pb} \) is identical in each pair of teeth. In a subject from Spain who had been in Australia for 21 years, the upper lateral incisor (M813/M814) exhibited no change compared with the molar (M815/M816) for which a change of 44 was observed. In a subject from Syria who had been in Australia for 19 years, the

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**Table 1. Lead isotopic and concentration data and isotopic shifts in \( ^{207}\text{Pb}/^{206}\text{Pb} \) between enamel (crown) and dentine (root) for immigrant teeth**

| Country | Tooth section | \( ^{207}\text{Pb}/^{206}\text{Pb} \) | \( ^{206}\text{Pb}/^{207}\text{Pb} \) | Pb (ppm) | \( ^{207}\text{Pb}/^{206}\text{Pb} \) (Δ × 1000) | Tooth type | Age (year) | Years in Australia | N or R* |
|---------|---------------|----------------|----------------|--------|----------------|-------------|------------|-----------------|-------|
| Lebanon 1 | H899 CR | 0.6794 | 17.64 | 2.8 | 138 | PM | NA | NA | N |
| Lebanon 2 | CR | 0.6748 | 17.77 | 1.0 | 15 | LO M | NA | NA | N |
| Lebanon 3 | RT | 0.6763 | 17.73 | 5.4 | | | | | |
| Lebanon 4 | CR | 0.6774 | 17.70 | 1.8 | 108 | PM | NA | NA | N |
| Lebanon 5 | M819 CR | 0.8666 | 18.00 | 4.9 | 549 | LLO 2nd M | 39 | 23 | N |
| Lebanon 6 | M820 RT | 0.9215 | 16.81 | 29.0 | | | | | |
| Lebanon 7 | M821 CR | 0.9236 | 16.73 | 8.2 | 0 | UL 2nd M | 29 | 26 | Total | |
| Lebanon 8 | M822 RT | 0.9241 | 16.72 | 10.7 | | | | | |
| Lebanon 9 | M831 CR | 0.9668 | 17.54 | 1.8 | 97 | UR Cl | NA | 21 | R |
| Lebanon 10 | M832 RT | 0.8771 | 17.72 | 15.1 | | | | | |
| Lebanon 11 | M835 CR | 0.8823 | 17.63 | 2.2 | 126 | UR 1st M | 41 | 1 | N |
| Lebanon 12 | M836 RT | 0.8949 | 17.33 | 7.7 | | | | | |
| Spain 1 | M813 CR | 0.8697 | 17.47 | 11.2 | | | | | |
| Spain 2 | M814 CR | 0.8732 | 17.82 | 4.4 | 0 | UR I | 38 | 21 | None |
| Spain 3 | M815 CR | 0.8749 | 17.78 | 5.1 | 44 | UL M | 38 | 21 | R |
| Spain 4 | M816 CR | 0.8705 | 17.91 | 18.8 | | | | | |
| Spain 5 | M825 CR | 0.8949 | 17.33 | 2.3 | 141 | UR M | 45 | 19 | R |
| Spain 6 | M826 RT | 0.8808 | 17.65 | 21.9 | | | | | |
| Spain 7 | M827 CR | 0.8900 | 17.45 | 2.4 | 60 | UR Cl | 45 | 19 | R |
| Spain 8 | M828 RT | 0.8840 | 17.57 | 8.2 | | | | | |
| Turkey 1 | M829 CR | 0.8796 | 17.68 | 3.1 | 123 | UR 1st M | 45 | 14 | R |
| Turkey 2 | M830 CR | 0.8647 | 17.99 | 10.0 | 145 | LO 1st M | 31 | 8 | N |
| Turkey 3 | M831 CR | 0.8702 | 17.69 | 5.3 | | | | | |
| Bulgaria 1 | H814 CR | 0.8544 | 18.27 | 16.2 | 50 | LO I | NA | NA | N |
| Bulgaria 2 | H815 CR | 0.8555 | 18.20 | 16.9 | 69 | LO I | NA | NA | N |
| Czech 1 | H917 CR | 0.8624 | 18.08 | 24.1 | | | | | |
| Czech 2 | H918 CR | 0.8691 | 17.92 | 20.0 | | | | | |
| Czech 3 | H919 CR | 0.8670 | 17.94 | 4.1 | 20 | NA | NA | NA | N |
| Czech 4 | H920 CR | 0.8690 | 17.92 | 14.5 | | | | | |
| Poland 1 | I823 CR | 0.8720 | 17.84 | 2.5 | 371 | PM | 69 | 43 | N |
| Poland 2 | I824 CR | 0.8733 | 17.79 | 3.4 | 519 | CA | 63 | 44 | N |
| Poland 3 | I824 RT R | 0.9236 | 16.72 | 34.5 | | | | | |
| Poland 4 | I824 RT NR | 0.9252 | 16.70 | 63.0 | | | | | |
| Poland 5 | O737 CR | 0.8544 | 18.22 | 0.8 | 28 | M | 34 | 2 | N |
| Croatia 1 | I544 CR | 0.8694 | 18.39 | 6.5 | 123 | LO M | 55 | 12 | N |
| Croatia 2 | I545 CR | 0.8649 | 18.45 | 6.5 | 118 | LO M | 25 | 4 | N |
| Croatia 3 | I546 CR | 0.8612 | 18.10 | 7.6 | | | | | |
| Croatia 4 | I547 CR | 0.8516 | 18.33 | 1.7 | 30 | U M/1 | 45 | 15 | N |
| Croatia 5 | I548 CR | 0.8546 | 18.26 | 4.1 | | | | | |
| Egypt | M829 CR | 0.8910 | 17.43 | 2.3 | 92 | 2nd M | 50 | 15 | R |
| M830 RT | 0.8818 | 17.63 | 6.3 | | | | | | |

*Continued, next page.*
Abbreviations: N, normal; R, reverse; CR, crown; RT, root; M, molar; PM, premolar; L0, lower; U, upper; CA, canine; I, incisor; CI, central incisor; LLO, left lower; UL, upper left; RLO, right lower; UR, upper right; RT R, circumpulpal material reamed out; RT NR, circumpulpal material not reamed out; NA, information not available.

*Normal means that the dentine lead relative to the enamel lead has shifted towards the Australian isotopic value (in this case -16.7); reverse means that the dentine lead relative to the enamel lead has shifted away from the Australian value.

These data show that there has possibly been total exchange with Australian lead as the subject came to Australia at 3 years of age.

Different teeth from same subject.

Table 1 Continued.

| Country | Tooth | Pb206Pb/207Pb | Pb206Pb/204Pb | Pb (ppm) | 207Pb/206Pb (λ × 1000) | Tooth type | Age (year) | Years in Australia | N | R*
|---------|-------|----------------|----------------|---------|-------------------------|------------|-------------|-------------------|---|---
| Greece | M800 CR | 0.8602 | 18.17 | 5.0 | 35 | UR 3rd M | 53 | 28 | R*
|        | M810 RT | 0.8567 | 18.23 | 24.3 | | | | | |
| United Kingdom | O753 CR | 0.9067 | 17.12 | 1.7 | 170 | UL I | 30 | 26 | N
|        | O754 RT | 0.8237 | 16.72 | 5.9 | | | | | |
| Uruguay | O751 CR | 0.8640 | 18.05 | 6.9 | 139 | UR I | 46 | 11 | N
|        | O752 RT | 0.8779 | 17.78 | 22.2 | | | | | |
| Italy 1 | O755 CR | 0.8591 | 18.21 | 4.2 | | | | | |
|        | O756 RT NR | 0.8797 | 17.78 | 28.5 | 206 | | | | |
|        | O757 RT R | 0.8801 | 17.73 | 29.0 | 210 | | | | |
| Italy 2 | O758 CR | 0.8670 | 18.00 | 1.6 | | | | | |
|        | O759 RT NR | 0.8771 | 17.76 | 19.8 | 101 | | | | |
|        | O760 RT R | 0.8739 | 17.85 | 19.1 | 69 | | | | |
| Chile | O761 CR | 0.8779 | 17.74 | 5.5 | | | | | |
|        | O762 NR | 0.8706 | 17.91 | 24.0 | 73 | | | | |
|        | O763 R | 0.8696 | 17.91 | 20.7 | 83 | | | | |

Figure 1. Change in 207Pb/206Pb ratio in dentine relative to enamel versus the length of time the subject has resided in Australia. Note the increase in the change with time as the dentine approaches the Australian lead value. Normal means that the dentine lead relative to the enamel lead has shifted towards the Australian isotopic value (in this case -16.7); reverse means that the dentine lead relative to the enamel lead has shifted away from the Australian value.

The data for the bone sample and that for a tooth with a value of change of 549 (λ) are excluded from the regression analysis.

molar exhibited more than twice the change observed in the upper central incisor. The difference in change between the Spanish and Syrian teeth may be a function of lead concentration as the root dentine of the central incisor of the Spanish subject contained more than three times that in the Syrian incisor, as discussed below.

Tooth type. In adults, tooth type was not usually considered to be important when evaluating lead accumulation (17,18-20), although it was attributed significance in some investigations (21-23). Tooth type does not appear to be of concern in the evaluation of isotopic compositions in deciduous teeth (4,10); however, in this study, tooth type does appear important in some cases. For example, in two cases, the subjects arrived in Australia when they were about 3-4 years old and have resided there for 26 years [Lebanon 6 (M821/M822) and the United Kingdom (O753/O754)]. The Lebanese tooth was a second molar and there was no isotopic difference between the enamel and dentine. In contrast, the U.K. tooth was an incisor and the dentine showed significant exchange with Australian lead (Table 1). Other than the higher lead concentration in the Lebanese tooth, the explanation for the difference in exchange rate may be the time of formation of the teeth. In the U.K. case, the crown (enamel) in the permanent incisor was already formed and the subject had been exposed to U.K. lead, by 4 years of age, whereas the crown of the molar in the Lebanese subject was only just forming (or not even so) at the age of 3 years.

Enamel and dentine. For permanent teeth in this study, enamel always contained lower lead concentrations compared with dentine in the roots. The ratio of dentine Pb/enamel Pb varies from 1.2 to 18.5, but most are in the range 1-8. For deciduous teeth the ratio varies from 1.5 to 4.1. There is a significant correlation of enamel and dentine lead (R² 0.40, p=0.0001; Table 2).

Removal of circumpulpal dentine. Rabinowitz et al. (21,10,11) identified a correlation between tooth and blood lead concentrations in deciduous teeth using a wedge sampling approach whereby a wedge of coronal dentine and the circumpulpal canal was analyzed. Part of the correlation may have arisen from the inclusion of the pulpal canal, whose lead probably reflects that of more recent origin, such as in blood leads. To evaluate the contribution from the circumpulpal canal to the root analysis, slices of root were analyzed with the canal and cementum intact and compared with contiguous slices from which the circumpulpal canal and cementum had been removed. Samples in which the circumpulpal dentine has been removed exhibited a poorer correlation with the rate of exchange with residence time of dentine lead and Australian lead than those with intact circumpulpal dentine.

Lead concentrations in sections with the canal and cementum removed were approximately half those with the canal intact, and the isotopic composition exhibited a slightly higher proportion of Australian lead (Poland 2, Croatia 5, and Yugoslavia 1 in Table 1). These data are consistent with earlier data obtained on deciduous teeth (4).

Removal of cementum. Slices of root with intact circumpulpal canal and cementum had relatively similar isotopic compositions and lead concentrations to contiguous slices from which the cementum was removed (Italian and Chilean subjects in Table 1). From these and the above analy-
The rate of exchange with residence time in dentine of European and Australian lead is estimated from the slope of the regression line in Figure 1, relative to a value in European lead of 0.862 and that in Australian lead of 0.922. The estimated rate of exchange with residence time or turnover of lead in dentine is -1 ± 0.3%/year. Perhaps, fortuitously, this is similar to the rate estimated for lead turnover in cortical bone by Rabinowitz (3), or it may reflect the similarity in processes that occur in lead in bone and dentine, notwithstanding the absence of osteoclasts and osteoblasts in dentine. In the lead isotope study of deciduous teeth from Broken Hill children, Gulson (16) estimated that lead was added to dentine at a rate of approximately 2–3%/year.

**Rate of exchange with residence time in trabecular bone**. Croatia 3 (1547) lived in Australia for 15 years prior to tooth extraction. There was little change in isotopic composition between enamel and dentine over this time, but there has been almost total exchange with Australian lead in the bone (Table 1). Although based on only one analysis, the estimated rate of exchange with residence time of lead in trabecular bone relative to enamel is ~6%, approximately six times that of dentine/enamel. This value is similar to the lead turnover of ~8% in trabecular bone estimated by Rabinowitz (3) using a quite different approach. The almost complete isotopic exchange in socket bone (trabecular bone) lead with Australian lead for subject 1547 over a 15-year period is consistent with half-lives of 7–13 years estimated from measurements on tibia/calcaneus/phalange by the XRF method and blood lead analyses on occupationally exposed subjects (5–9).

**A two-way exchange of lead in dentine**. In the initial study, all analyses showed a normal change in the isotopic composition of the dentine. That is, there was a positive shift in the 207Pb/206Pb ratio from the country of origin, manifested by the isotopic composition in the enamel, towards the Australia values. Negative changes (reverse data in Table 1) signify interactions with other sources of lead. When the study was expanded, 9 of 19 enamel/dentine pairs exhibited a reversal of this trend. At this stage, no explanation for the reversal is obvious apart from lack of information about previous residence history, occupational/hobby exposure, or their diets in Australia. Diet is not considered a major contributor to the reverse isotopic changes in dentine because analyses of teeth and bones from Australian residents indicate that the 206Pb/207Pb ratio has been <17.0 (or 207Pb/206Pb >0.91) for decades. This long-term low value is also indicated by the data for Lebanon 6 (M821/M822) who arrived in Australia at 3 years of age and thus experienced 26 years of exposure to Australian lead. Any trace of his maternal skeletal lead has been replaced with Australian lead. Furthermore, it is only over the last few years that the Australian diet has 206Pb/207Pb values >17.0 (15).

Alternative explanations for the reverse isotopic trends in dentine may be that the lead is mobilized from an endogenous source, such as cortical bone, and introduced to the dentine via the dental tubules, or that they reflect a two-way migration of lead in dentine.

Based on epidemiological data, Steenhout (23) argued for a one-way exchange of lead in tooth dentine but, given the structure of teeth and that collagen could theoretically act as an ion exchange medium, there is no reason to preclude a two-way exchange of lead. In evaluating models of tooth lead kinetics, Rabinowitz et al. (11) found that tooth lead/blood lead data were compatible with a model that allows lead to be slowly removed from dentine, a conclusion also reached by Gulson and Wilson (4) and Gulson (16). Bercovitz and Lauffer (20) were able to procure paired trabecular bone and tooth samples from 97 Israeli subjects 9–81 years of age. They found higher concentrations of lead in bone than in teeth until the age of 50; after 50 years of age the reverse was noted. These changes were attributed to the release of lead from trabecular bone, which is detectable after age 35.

**Rate of exchange with residence time and age**. There are variable correlations of rate of exchange with residence time in the 207Pb/206Pb ratio and the age of the subject. For all data, the R² = 0.07 and p = 0.19 (n = 25), but with the exclusion of the extreme data point for Lebanon 5 (M819), the correlation assumes significance (R² = 0.17, p = 0.05, n = 24; Table 2).

### Conclusions

Results of this study of permanent teeth support the earlier investigation of deciduous teeth in demonstrating that, relative to enamel, lead can be added to dentine. In contrast to deciduous teeth and the initial part of this study, reversals were observed in the predicted trends of lead added to dentine. These reversals may arise from changes in environmental sources, from endogenous mobilization of lead from the skeleton, or from a two-way exchange of lead in dentine. A systematic study of (anterior) teeth from several subjects, for example, from two countries, who have lived in Australia for >5 years (preferably >10 years), would
provide evidence of lead turnover in teeth. A small sample of socket bone would provide even more convincing data on lead turnover in teeth and bone.

Given the limited amount of data for bone lead turnover in children, an investigation of teeth from children who have lived >1 year in Australia may provide evidence of lead turnover in a period of rapid skeletal development.

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