

DO NOT QUOTE ON LIFE

TABLE OF CONTENTS
CHAPTER 10
METABOLISM OF LEAD

<u>Section</u>	<u>Page</u>
10.1 Introduction.....	10-1
10.2 Lead Absorption in Humans and Animals.....	10-1
10.2.1 Respiratory Absorption of Lead.....	10-1
10.2.1.1 Human Studies.....	10-2
10.2.1.2 Animal Studies.....	10-6
10.2.2 Gastrointestinal Absorption of Lead.....	10-7
10.2.2.1 Human Studies.....	10-7
10.2.2.2 Animal Studies.....	10-11
10.2.3 Percutaneous Absorption of Lead.....	10-14
10.2.4 Transplacental Transfer of Lead.....	10-15
10.3 Distribution of Lead in Humans and Animals.....	10-16
10.3.1 Lead in Blood.....	10-18
10.3.2 Lead Levels in Tissues.....	10-18
10.3.2.1 Soft Tissues.....	10-19
10.3.2.2 Mineralizing Tissue.....	10-23
10.3.3 Chelatable Lead.....	10-24
10.3.4 Mathematical Descriptions of Physiological Lead Kinetics...	10-26
10.3.5 Animal Studies.....	10-27
10.4 Lead Excretion and Retention in Humans and Animals.....	10-29
10.4.1 Human Studies.....	10-29
10.4.2 Animal Studies.....	10-34
10.5 Interactions of Lead with Essential Metals and Other Factors.....	10-37
10.5.1 Human Studies.....	10-37
10.5.2 Animal Studies.....	10-40

DRAFT
DO NOT QUOTE OR CITE

PRELIMINARY DRAFT

CONTENTS (Continued)

<u>Section</u>	<u>Page</u>
10.5 (Continued)	
10.5.2.1 Interactions of Lead with Calcium.....	10-41
10.5.2.2 Interactions of Lead with Iron.....	10-45
10.5.2.3 Lead Interactions with Phosphate.....	10-46
10.5.2.4 Interactions of Lead with Vitamin D.....	10-46
10.5.2.5 Interactions of Lead and Lipids.....	10-47
10.5.2.6 Lead Interaction with Protein.....	10-47
10.5.2.7 Interaction of Lead with Milk Components.....	10-48
10.5.2.8 Lead Interactions with Zinc and Copper.....	10-48
10.6 Interrelationships of Lead Exposure, Exposure Indicators and Tissue Lead Burdens.....	10-49
10.6.1 Temporal Characteristics of Internal Indicators of Lead Exposure.....	10-49
10.6.2 Biological Aspects of External Exposure-Internal Indicator Relationships.....	10-50
10.6.3 Internal Indicator-Tissue Lead Relationships.....	10-51
10.7 Metabolism of Lead Alkyls.....	10-55
10.7.1 Absorption of Lead Alkyls in Humans and Animals.....	10-55
10.7.2 Biotransformation and Tissue Distribution of Lead Alkyls..	10-56
10.7.3 Excretion of Lead Alkyls.....	10-58

PRELIMINARY DRAFT

10. METABOLISM OF LEAD

10.1 INTRODUCTION

The absorption, distribution, retention, and excretion of lead in humans and animals and the various factors which mediate the extent of such toxicokinetic processes are discussed in this section. While inorganic lead is the form of the element which has been most heavily studied, organolead compounds are also emitted into the environment and are quite toxic. Hence, these forms are also included for discussion.

Since the preparation of the 1977 Air Quality Criteria Document for lead (AQCD-77), a number of reports have appeared which have proved particularly helpful in both the quantification of the various processes discussed in this section and the assessment of the relative interactive impact of factors such as nutritional status in determination of exposure risk.

10.2 LEAD ABSORPTION IN HUMANS AND ANIMALS

The amounts of lead entering the bloodstream from various routes of absorption are determined not only by the levels of the element in the particular media but also by various physical and chemical parameters characterizing lead. In addition, specific host factors such as age and nutritional status are important, as is interindividual variability. An additional element in all of this, of practical significance in assessment of absorption rates, is the question of whether the subject is in "equilibrium" with respect to a given level of lead exposure.

10.2.1 Respiratory Absorption of Lead

The movement of lead from ambient air to the bloodstream is a two-part process: some fraction of air lead is deposited in the respiratory tract and of this deposited amount, some fraction is subsequently absorbed directly into the bloodstream or otherwise cleared from the respiratory tract. At present, enough data exist to make some quantitative statements about both of these components of respiratory absorption of lead.

In the AQCD-77 for lead, modeling of the deposition and removal of lead from the lungs and the upper respiratory tract by the International Radiological Protection Commission, IRPC (Task Group on Lung Dynamics, 1966), was described. Briefly, the model predicts that 35 percent of lead inhaled from ambient air would be deposited in the airways, with most of this being

PRELIMINARY DRAFT

deposited in the lung. The IRPC model predicts a total deposition of 40-50 percent for 0.5 μm particles and indicates that the absorption rate would vary, depending on the solubility of the particular form.

10.2.1.1 Human Studies--Table 10-1 tabulates the various studies with human subjects which provide data on the deposition of inorganic lead in the respiratory tract. Studies of this type have involved diverse methodology to characterize both the particles being inhaled in terms of size and size ranges, and fractional distribution. The use of radioisotopic or stable lead isotopes to directly or indirectly measure lead deposition and uptake into the bloodstream has been particularly helpful in determining the quantitative features of these processes.

From the studies of Kehoe (1961a,b,c) and their updated evaluation by Gross (1981), as well as data from Chamberlain et al. (1978), Morrow et al. (1980), and Nozaki (1966), it appears that the deposition rate of airborne lead as encountered in the general population is ca. 30-50 percent, with these rates being dependent on particle size and ventilation rates. Ventilation rate is particularly important with submicron particles where Brownian diffusion governs deposition, with the result that a slower breathing rate enhances the frequency of collisions of particles with alveolar wall.

Figure 10-1 reproduces a composite figure of Chamberlain et al. (1978) comparing data, both calculated and experimentally measured, for the relationship of percentage deposition to particle size. With increasing size of particle, the deposition rate decreases to a minimum over the range where Brownian diffusion predominates, followed by an increase in deposition with size ($>0.5 \mu\text{m}$ MMED) as impaction and sedimentation become the main deposition factor.

In contrast to the picture for ambient air or chamber data tabulated in Table 10-1, deposition rates in some occupational settings are associated with relatively large particles. These particles are in a range where relatively higher deposition rates occur; however, much of this will be in the upper respiratory tract, with considerable movement eventually to the gastrointestinal tract by ciliary action and swallowing. Mehani et al. (1966) measured deposition rates in battery workers and those in marine scrap yards and observed deposition rates of 28-70 percent.

PRELIMINARY DRAFT

TABLE 10-1. DEPOSITION OF LEAD IN THE HUMAN RESPIRATORY TRACT

Form	Particle Size	Exposure	Percent Deposition	Reference
Pb ₂ O ₃ aerosols from engine exhaust	0.05 µm median count diameter in 38 studies; 5 subjects exposed to average of 0.9 µm	Chamber studies, 10, 20, or 150 µg/m ³ , 3 h. alternate days, 12 subjects	30-70 percent for mainly 0.05 µm particles with a mean of 48%	Kehoe, 1961; Gross, 1981
Lead "fumes" made in induction furnace	0.05-1.0 µ mean diameter	mouthpiece/aerosol chamber; 10 mg/m ³ , adult subjects	42/0.05 µm 63/1.0 µm	Nozaki, 1966
²⁰³ Pb-labeled Pb ₂ O ₃ aerosol	mean densities of 0.02, 0.04, 0.09 µm	mouthpiece/aerosol chamber, adult subjects	80/0.02 µm 45/0.04 µm 30/0.09 µm	Chamberlain et al., 1978
Ambient air lead near motorway and other urban areas in U.K.	mainly 0.1 µm	2-10 µg/m ³ , adult subjects	60/fresh exhaust 50/other urban area	Chamberlain et al., 1978
²⁰³ Pb-labeled Pb(OH) ₂ or Pb(Cl) ₂ aerosols	both forms at 0.25 µm MMAD	50 liters air; 0.2 µCi/liter; adult subjects	23/chloride 26/hydroxide	Morrow et al., 1980
Lead in workplace air; battery factory and shipbreaking operations	Not determined; defined as fumes, fine dust, or coarse dust	3 adult groups: 23 µg/m ³ - controls 86 µg/m ³ - battery workers 180 µg/m ³ - scrap yard	47/battery workers; 39 shipyard and controls	Mehani et al., 1969

TEH 0530791

10-4

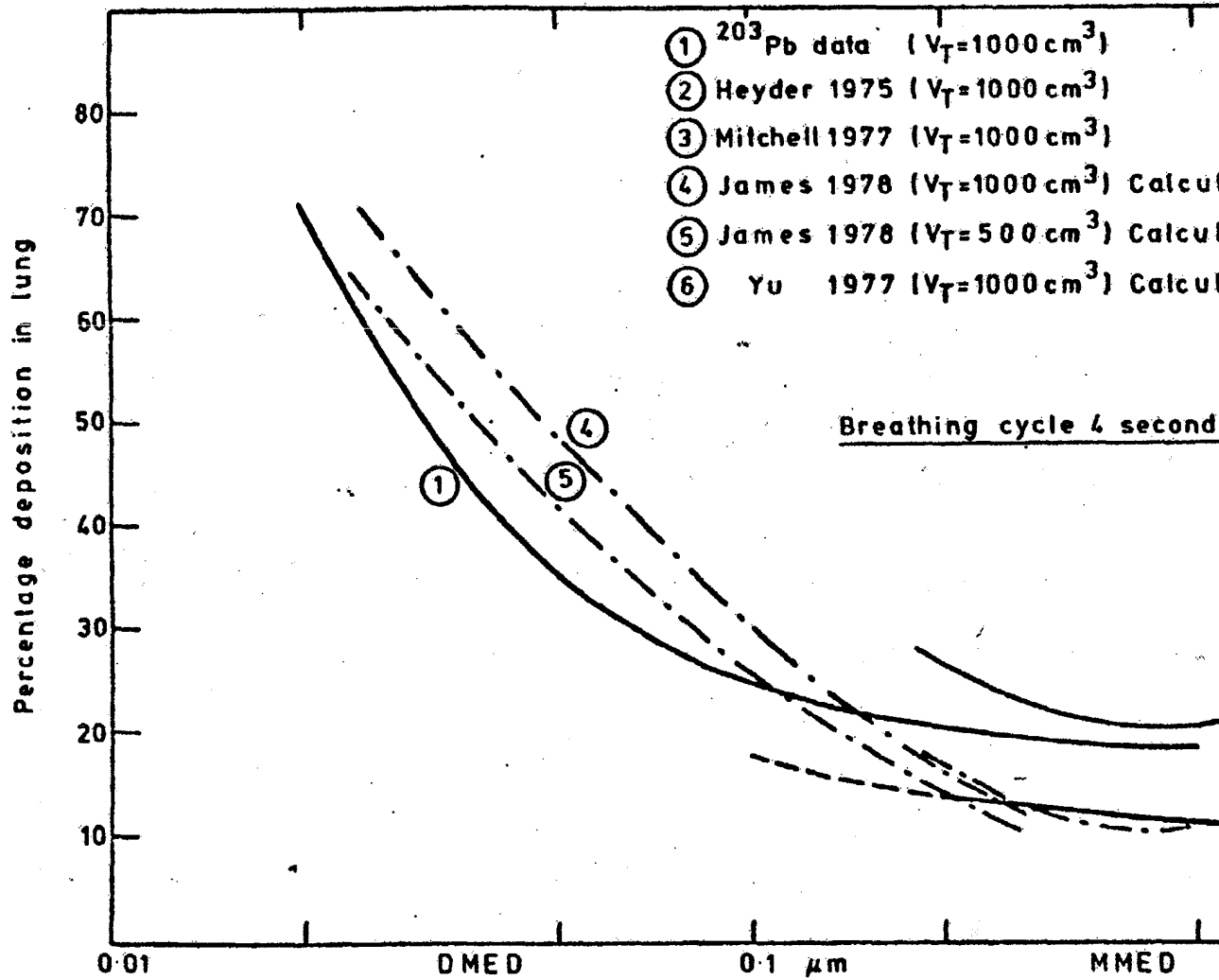


FIGURE 10.1 EFFECT OF PARTICLE SIZE ON LEAD DEPOSITION RATE IN THE LUNG
(From Chamberlain et al., 1978)

DUP050031704

PRELIMINARY DRAFT

Systemic uptake of lead directly from the lower respiratory tract occurs, while much of upper tract deposition involves swallowing and some uptake in the gut. From the radioactive isotope data of Chamberlain et al. (1978) and Morrow et al. (1980) and the stable isotope studies of Rabinowitz et al. (1977), it can be concluded that lead deposited in the lower respiratory tract is quantitatively absorbed.

Chamberlain et al. (1978) used ^{203}Pb -labeled lead in engine exhaust, lead oxide or lead nitrate aerosols in experiments where human subjects inhaled the lead from a chamber through a mouthpiece or in wind tunnel aerosols. By 14 days, ca. 90 percent of the label was removed from the lung. Lead movement into the bloodstream could not be described by a simple exponential function, 20 percent being absorbed within 1 hour and 70 percent within 10 hours.

Rabinowitz et al. (1977) administered ^{204}Pb tracer to young adult volunteers and were able to determine by isotope tracer as well as balance data that 14 μg of lead was absorbed by these subjects daily for ambient air lead of 1-2 $\mu\text{g}/\text{m}^3$. Assuming a ventilation rate of 20 m^3 daily, a deposition rate of 50 percent of ambient air (Chamberlain et al., 1978), and a mean air lead level of 1.5 $\mu\text{g}/\text{m}^3$ (2.0 $\mu\text{g}/\text{m}^3$ outside the study unit, 1.0 $\mu\text{g}/\text{m}^3$ inside, as determined by the authors), then 15 μg lead was available for absorption; hence, better than 90 percent of inhaled lead was absorbed daily.

Morrow et al. (1980) followed the systemic uptake of ^{203}Pb -labeled lead in either lead chloride or lead hydroxide aerosols with an average size of 0.25 (± 0.1) μm MMAD, using 17 adult subjects. Half of the deposited fraction of either aerosol was absorbed in 14 hours or less.

The radiolabel data described above are consistent with the results of Hursh and Mercer (1970), who studied the systemic uptake of ^{212}Pb on a carrier aerosol.

Chamberlain and Heard (1981) calculated an absorption rate for particle sizes encountered in workplace air of ca. 47 percent.

Given the relative invariance of absorption rate for deposited lead in the above studies as a function of chemical form of the element (Chamberlain et al., 1978; Morrow et al., 1980), it appears that inhaled lead lodged deep in the respiratory tract is equally absorbed regardless of form.

Supporting evidence for total systemic uptake of deposited lead in humans comes from autopsy tissue analysis for lead content. Barry (1975) found that

PRELIMINARY DRAFT

lead was not accumulated in the lungs of lead workers, consistent with the data of Gross et al. (1975) for non-occupationally exposed subjects.

All of the available data for lead deposition and uptake from the respiratory tract in humans has been obtained with adults, and comparisons with such exposure in children is not quantitatively possible. Although children 2 years of age weigh one-sixth as much as an adult, they inhale 40 percent as much air lead as adults (Barltrop, 1972). James (1978) has also taken into account differences in airway dimensions, adults vs. children, and has estimated that the 10-year-old child has a deposition rate 1.6-2.7-fold higher than the adult on a weight basis.

10.2.1.2 Animal Studies--Experimental animal data for quantitative assessment of lead deposition and absorption rates from the lung and upper respiratory tract are limited. The available information does, however, support the outcome of human data demonstrating that respired lead is extensively and rapidly absorbed.

Morgan and Holmes (1978) exposed adult rats, by nose-only technique, to a ²⁰³Pb-labeled engine exhaust aerosol generated in the same manner as by Chamberlain et al. (1978) over a period of 8 days. Exposure was at a level of 21.9-23.6 nCi label/liter chamber air. Adjusting for label deposition on the animal pelt, 20-25 percent of the label was deposited in the lungs. Deposited lead was extensively taken up in blood, 50 percent within 1 hour, 98 percent within 7 days. The absorption rate kinetic profile was similar to that reported in humans (Chamberlain et al., 1978).

Boudene et al. (1977) exposed rats to ²¹⁰Pb-labeled aerosols at a level of 1 µg label/m³ and 10 µg/m³, the majority of the particles being 0.1-0.5 µm in size. At 1 hour, 30 percent of the label left the lung, 90 percent by 48 hours.

Bianco et al. (1974) used ²¹²Pb aerosol inhaled briefly by dogs and at a submicron size range (≤ 0.2 µm) and found a label clearance half-time from the lung of ca. 14 hours. Greenhalgh et al. (1979) found that direct instillation of ²⁰³Pb-labeled lead nitrate solution into the lung of the rat lead to an uptake of ca. 42 percent within 30 minutes, compared to an uptake rate of 15 percent by 15 minutes in the rabbit. These instillation data are consistent with the report of Pott and Brockhaus (1971), where it was noted that intratracheal instillation of lead in solution (as bromide) or suspension (as

PRELIMINARY DRAFT

oxide) serially over 8 days resulted in systemic lead levels in tissues indistinguishable from injected lead. Randall et al. (1975) found that the relative movement of lead into blood of baboons inhaling a lead oxide (Pb_3O_4) was more rapid and to a greater extent with coarse $1.6 \mu m$ mean diameter) than with fine ($0.8 \mu m$ mean diameter) particles. This suggests that considerable fractions of both size particles were eventually lodged in the gut, where absorption of lead tends to be higher than in other animal species (Pounds et al., 1978), with a more rapid movement of the larger particles to the gut.

10.2.2 Gastrointestinal Absorption of Lead

Gastrointestinal absorption of lead involves uptake of lead from food and beverages as well as non-food materials such as lead deposited in the upper respiratory tract and swallowed eventually, and ingestion of non-food material, primarily in children via normal mouthing activity and pica.

Two issues of concern with lead uptake from the gut are the relative rates of such uptake in developing versus adult organisms, including humans, and the role of bioavailability of lead in affecting such uptake.

10.2.2.1 Human Studies--Based on the long-term metabolic studies with adult volunteers, Kehoe (1961a,b,c) estimated that ca. 10 percent of dietary lead--in food and beverages--is absorbed from the gut of humans. As may be seen in the assessment by Gross (1981), there can be considerable variation among subjects of the various balance parameters. These studies did not take into account the contribution of biliary clearance of lead into the gut, which would have affected measurements for both absorption and total excretion. Chamberlain et al. (1978) have determined that the level of endogenous fecal lead is ca. 50 percent of urinary lead values. Chamberlain et al. (1978) have estimated that 15 percent of dietary lead is absorbed, if the amount of endogenous fecal lead is taken into account.

Subsequent to the Kehoe studies, a number of reports have determined GI absorption using both stable and radioisotopic labeling of dietary lead. Generally, these support the observation that absorption of lead taken with food in the adult human is limited.

Harrison et al. (1969) determined a mean absorption rate of 14 percent for 3 adult subjects ingesting ^{203}Pb -labeled lead in diet, a figure in accord with the results of Hursh and Suomela (1968).

PRELIMINARY DRAFT

Chamberlain et al. (1978) studied the absorption of ^{203}Pb in two forms (as the chloride and as the sulfide) taken with food. The corresponding absorption rates were 6 (sulfide) and 7 (chloride) percent, taking into account endogenous fecal excretion.

Using adult subjects ingesting the stable isotope ^{204}Pb added to diet, Rabinowitz et al. (1974) reported an average gut absorption of 7.7 percent. In a later study, Rabinowitz et al. (1980) measured an absorption rate of 10.3 percent.

A number of recent studies indicate that lead ingested under conditions of fasting is absorbed to a much greater extent than when it is taken with or incorporated into food.

Blake (1976) measured a mean absorption rate of 21 percent when 11 adult subjects ingested ^{203}Pb -labeled lead chloride several hours after breakfast. Chamberlain et al. (1978) found that lead uptake in 6 subjects fed ^{203}Pb as the chloride was 45 percent after a fasting period, compared to 6 percent with food. Heard and Chamberlain (1982) obtained a rate of 63.3 percent using a similar procedure with 8 subjects. Rabinowitz et al. (1980) reported an absorption rate in 5 subjects of 35 percent when ^{204}Pb was ingested after a fasting period of 16 hours. To the extent that lead in beverages is ingested between meals, these isotope studies support the observations of Barltrop (1975) and Gerber and Wei (1974) that beverage lead is absorbed to a greater extent than is food lead.

The relationship of lead bioavailability in the human gut to the chemical/biochemical form of lead can be determined from available data, although interpretation is complicated by the relatively small amounts given and the presence of various components of food in the gut.

Harrison et al. (1969) found no difference in lead absorption from the human gut when lead isotope was given either as the chloride or incorporated into alginate. Chamberlain et al. (1978) found that labeled lead as the chloride or sulfide was absorbed to the same extent when given with food, while the sulfide form was absorbed at a rate of 12 percent compared to 45 percent for the chloride when given under fasting conditions. Rabinowitz et al. (1980) obtained similar absorption rates for the chloride, sulfide or cysteine complex forms when administered with food or under fasting conditions. Heard and Chamberlain (1982) found no difference in absorption

PRELIMINARY DRAFT

rate when isotopic lead, ^{203}Pb , was given with unlabeled liver and kidney or when the label was first incorporated into these organs.

Three studies have focused on the question of differences in gastrointestinal absorption rates between adults and children.

Alexander et al. (1973) carried out 11 balance studies with 8 children, aged 3 months to 8 years. Intake averaged $10.6 \mu\text{g Pb/kg body weight/day}$ (range 5-17) and the mean absorption rate determined from metabolic balance studies was 53 percent. Ziegler et al. (1978) carried out a total of 89 metabolic balance studies with 12 normal infants aged 2 weeks to 2 years. Diets were closely controlled and lead content measured. Two discrete studies were carried out and in the first, 51 balance studies using 9 children furnished a mean absorption rate of 42.7 percent. In the second study, 6 children were involved in 38 balance studies involving dietary lead intake at 3 levels. For all daily intakes of $5 \mu\text{g Pb/kg/day}$ or higher, the mean absorption rate was 42 percent. At low levels of lead intake, data were variable, with some children apparently in negative balance, probably due to the difficulty in controlling low lead intake.

In contrast to these studies, Barltrop and Strehlow (1978) found that children hospitalized as orthopedic or "social" admissions gave highly variable results. Children were 3 weeks to 14 years old and a total of 104 balance studies with 29 children were carried out. Fifteen of the subjects were in net negative balance, the average dietary absorption was -40 percent and when weighted by number of balance studies, -16 percent.

It is difficult to draw a close comparison of these data with those of Ziegler et al. (1978). Subjects were inpatients, represented a much greater age range, and were not defined in terms of mineral nutrition or weight change status. As an urban pediatric group, the children in the latter study may have had higher prior lead exposure so that the "washout" phenomenon (Kehoe, 1961a,b,c; Gross, 1981) may have contributed to the highly variable data across these subjects. The calculated mean daily lead intake in the Barltrop and Strehlow group ($6.5 \mu\text{g/kg/day}$) is lower than those for all but one study group described by Ziegler et al. (1978). In the latter study it appears that data for absorption becomes more variable as the daily lead intake is lowered. Finally, in those children classified as orthopedic admissions, it is not

PRELIMINARY DRAFT

clear that skeletal trauma was without effect on lead equilibrium between bone and other body compartments.

As typified by the results of the NHANES II effort (Mahaffey et al., 1979), children at 2-3 years of age show a small peak in blood lead during childhood, and the question arises whether this observation indicates a biologically intrinsic factor such as increased absorption or retention, compared to older children, or whether this age group possesses some relationship to lead exposure peculiar to this group.

Several studies are relevant to the question. Zielhuis et al. (1978) reported data for blood lead levels in 48 hospitalized Dutch children ranging in age from 2 months to 6 years. Children up to 3 years old had a mean blood lead level of 11.9 $\mu\text{g Pb/dl}$ vs. a value of 15.5 in children aged 4-6 years. A significant positive relationship between child age and Pb-B was calculated ($r = 0.44$, $p < 0.05$). In the Danish survey of Nygaard et al. (1977), a subset of 126 children representing various geographical areas and age groups yielded the following blood lead values by mean age group: children ($N = 8$) having a mean age of 1.8 years had a mean Pb-B of 4.3 $\mu\text{g Pb/dl}$; those with a mean age of 3.7-3.9 had values ranging from 5.6-8.3 $\mu\text{g Pb/dl}$; children 4.6-4.8 years of age had a range of 9.2-10 $\mu\text{g Pb/dl}$. These authors note that the youngest group was kept at a nursery while the older kindergarten children had more interaction with the outside environment. Sartor and Rondia (1981) surveyed two population groups in Belgium, one of which consisted of subsets of children aged 1-4, 5-8, and 9-14 years. Children under the age of 1 had a mean Pb-B of 10.7 $\mu\text{g/dl}$; the 1-4 and 5-8 age groups were comparable, 13.9 and 13.7 $\mu\text{g/dl}$ respectively; and those 9-14 years old had a Pb-B of 17.2 $\mu\text{g/dl}$. In this study, all of the children were hospital patients.

While these European studies suggest that restriction of children in terms of environmental interaction, e.g., hospitalization or nurseries, is associated with an apparently different age-blood lead relationship than the U.S. NHANES II subjects, it remains to be demonstrated that European children in the 2-3 year age group show a similar peak. The issue merits further study.

Oral Intake of Non-Foodstuffs in Young Children

The normal mouthing activity of young children as well as the actual ingestion of non-food items, i.e., pica is a major concern in pediatric lead

PRELIMINARY DRAFT

exposure, particularly in urban areas with deteriorating housing stock and heavy automotive density as well as non-urban areas contiguous to lead production facilities. The magnitude of such potential exposures is discussed in Chapter 7, while an integrated assessment of relative impact on human intake will be assembled in Chapter 13. Such intake is intensified for children with pica and would include paint, dust and dirt.

EPA (1979), using data of Day et al. (1975) and Lepow et al. (1974) has attempted to quantitate the daily intake of soil/dust in young children from such mouthing activity as thumb-sucking and finger-licking. A total of 100 mg/day was obtained for children 2-3 years old, the amount of lead in this ingested quantity varying considerably from site to site. In the report, a gastrointestinal absorption rate of 30 percent was taken for lead in soil and dust. Of relevance to this estimate of absorption rate in children are the animal data discussed in the next section showing that lead in soil or dust and of variable chemical form is as available for absorption as food lead. The in vitro studies relating lead solubility in street dusts as a function of acidity clearly demonstrate that the acidity of the human stomach is adequate to extensively solubilize lead assimilated in this form.

To the extent that ingestion of such material by children is done other than at mealtime, the fasting factor in enhancing lead absorption from the human GI tract (vide supra) must also be considered. Hence, a factor of 30 percent for lead absorption from dusts and soils is not an unreasonable value.

Paint chip ingestion by children with pica has been estimated in the NAS report on lead poisoning in children (NAS, 1976) to be considerable. In the case of paint chips, EPA (1979) estimated an absorption rate as high as 17 percent. This value may be compared to animal data in the next section which indicates that lead in old paint films can undergo significant absorption in animals.

10.2.2.2 Animal Studies--Lead absorption from the GI tract of various adult experimental animal species appears to resemble that for the adult human, on the order of 1-15 percent in most cases.

Kostial and Kello (1979), Kostial et al. (1978), and Kostial et al. (1971) reported a value of 1 percent or less in adult rats maintained on commercial rat chow. These studies were carried out using radioisotopic tracers. Similarly, Barltrop and Meek (1975) reported an absorption rate of 4

PRELIMINARY DRAFT

percent in control diets while Aungst et al. (1981) found the value to range from 0.9 to 6.9 percent, depending on the level of lead given in the diet. In these rat studies, lead was given with food. Quarterman and Morrison (1978) administered ^{203}Pb label in small amounts of food to adult rats and found an uptake rate of ca. 2 percent at 4 months of age.

Pounds et al. (1978) obtained a value of 26.4 percent with 4 adult Rhesus monkeys given ^{210}Pb by gastric intubation. The higher rate, relative to the rat, may reflect various states of fasting at time of intubation or differences in dietary composition (vide infra), two factors which affect rates of absorption.

As seen with human subjects (vide supra), fasting appears to enhance the rate of lead uptake in experimental animals. Gerber and Wei (1974) found that fasting markedly enhanced gut uptake of lead in rats. Forbes and Reina (1972) found that lead dosing by gastric intubation of rats yielded an absorption rate of 16 percent, higher than other data for the rat. It is likely that intubation is done at time periods when there is little food in the gut. The data of Pounds et al. (1978) as described above may also suggest a problem with giving lead by gastric intubation or with water compared to mixing with food.

Bioavailability of lead in the GI tract of experimental animals has been the subject of a number of reports. The design of these studies differ, reflecting how "bioavailability" is defined by different investigators. In some cases, the dietary matrix is kept constant or nearly so while the chemical or physical form of the lead is varied. By contrast, other data describe the effect of changes in bioavailability as the basic diet matrix was changed. The latter case is complicated by the simultaneous operation of lead-nutrient interactive relationships, which are described in a separate section within this chapter.

Allcroft (1950) observed comparable effects when calves were fed lead in the form of the phosphate, oxide, basic carbonate ($\text{PbCO}_3 \cdot \text{Pb}(\text{OH})_2$), or incorporated into wet or dry paint. By contrast, lead sulfide in the form of finely ground galena ore was less toxic. Criteria for relative effect included kidney and blood lead levels and survival rate over time.

In the rat, Barltrop and Meek (1975) carried out a comparative absorption study using lead in the form of the acetate as the reference substance. The

PRELIMINARY DRAFT

carbonate and thallate were absorbed to a greater extent while the sulfide, chromate, naphthenate, and octoate were 44-67 percent of the reference agent. Gage and Litchfield (1968, 1969) found that lead naphthenate and chromate can undergo considerable absorption from the rat gut when incorporated into dried paint films, although less than when given with other vehicles. Yu et al. (1978) found that lead in the form of the acetate or as a phospholipid complex was equally absorbed from the GI tract of both adult and young rats at a level of 300 ppm. Uptake was assessed by weight change, tissue levels of lead, and urinary aminolevulinic acid levels.

In a study of interest for the problem of lead bioavailability in soils and dusts, particularly to exposed children, Dacre and Ter Haar (1977) compared the effects of lead as the acetate to lead contained in roadside and house paint soil, at a level of ca. 50 ppm, in commercial rat chow. Uptake of lead was indexed by weight change, tissue lead content, and inhibition of ALA-D activity. There was no significant difference in any of these parameters across the 3 groups, suggesting that neither the geochemical matrix in the soils or the various chemical forms--basic carbonate in paint soil, and the oxide, carbonate, and basic carbonate in roadside soil--affect lead uptake.

These data are consistent with the behavior of lead in dusts to acid extraction as reported by Day et al. (1979), Harrison (1979), and Duggan and Williams (1977). In the Day et al. study, street dust samples from England and New Zealand were extracted with hydrochloric acid over the pH range of 0-5. At gastric acidity, pH of 1, ca. 90 percent of the dust lead was solubilized. Harrison (1979) noted that at this same acidity, up to 77 percent of Lancaster, England, street dust lead was soluble, while an average 60 percent solubility was seen in London dust samples (Duggan and Williams, 1977). Since gastric solubilization must occur for lead in these media to be absorbed, the above data are useful in determining relative risk.

Kostial and Kello (1979) compared the relative absorption of ^{203}Pb from the gut of rats maintained on commercial rat chow vs. rats fed such "human" diets as baby foods, porcine liver, bread, and cow's milk. Absorption in the latter cases varied from 3-20 percent, compared to <1.0 percent with rat chow. This range of uptake for the non-chow diet compares closely with that reported for human subjects (vide supra). Similarly, Jugo et al. (1975a) observed that

PRELIMINARY DRAFT

rats maintained on fruit diets had an absorption rate of 18-20 percent. It would appear, then, that the generally observed lower absorption of lead in adult rat vs. adult human (see earlier discussion) is less reflective of a species than a dietary difference.

Barltrop and Meek (1979) studied the relationship of particle size of lead in two forms--as the metal or as lead octoate or chromate in powdered paint films--to the extent of gut absorption in the rat and found that there was an inverse relationship of uptake and particle size for both forms.

A number of studies have documented that the developing animal absorbs a relatively greater fraction of ingested lead than the adult groups, supporting those studies that have shown this age dependency in humans. For example, the adult rat absorbs ca. 1 percent lead or less when contained in diet, vs. a corresponding value 40-50 times greater in the rat pup (Kostial et al., 1971, 1978; Forbes and Reina, 1972). In the rat, this difference persists through weaning (Forbes and Reina, 1972), at which point uptake resembles that of adults. Part of this difference can be ascribed to the nature of the diet (mother's milk vs. regular diet), although it should be noted that the extent of absorption enhancement with milk vs. rat chow in the adult rat (Kello and Kostial, 1973) falls short of what is seen in the neonate. An undeveloped, less selective intestinal barrier may also exist in the rat neonate.

In non-human primates, Munro et al. (1975) observed that infant monkeys absorbed 65-85 percent via the gut vs. 4 percent in adults. Similarly, Pounds et al. (1978) noted that juvenile Rhesus monkeys absorbed ca 50 percent more lead than adults.

The question of the relationship of level of lead intake through the GI tract and rate of lead absorption was addressed by Aungst et al. (1981), who exposed adult and suckling rats to doses of lead by intubation over the range 1-100 mg Pb/kg or by variable concentrations in drinking water. With both age groups and both forms of oral exposure, lead absorption as a percentage of dose decreased, suggesting a saturation phenomenon for lead transport across the gut wall.

10.2.3 Percutaneous Absorption of Lead

Absorption of inorganic lead compounds through the skin appears to be considerably less significant than the respiratory and gastrointestinal routes of uptake. This is in contrast to the observations for lead alkyls and other

PRELIMINARY DRAFT

organic derivatives (AQCD-77). Uptake of alkyl lead through the skin is further discussed in Section 10.7.

Rastogi and Claussen (1976) found that cutaneous or subcutaneous administration of lead naphthenate in rat skin was associated with higher tissue levels and more severe toxic effects than was the case for lead acetate. Laug and Kuntze (1948) applied lead as the acetate, orthoarsenate, oleate, and the ethyl alkyl to rat skin and determined that the greatest levels of kidney lead were associated with the alkyl contact.

Moore et al. (1980) studied the percutaneous absorption of ^{203}Pb -labeled lead acetate in cosmetic preparations using 8 adult volunteers. Applied in wet or dry forms, absorption was indexed by blood, urine, and whole body counting. Absorption rates ranged from 0-0.3 percent, with the highest values obtained when the application sites were scratched. These workers estimated that the usual use of these preparations would result in an absorption of ca. 0.06 percent.

10.2.4 Transplacental Transfer of Lead

Uptake of lead by the human and animal fetus occurs readily, based on such indices of uptake as fetal tissue lead measurements and, in the human, cord blood lead levels. Barltrop (1969) and Horiuchi et al. (1959) demonstrated by fetal tissue analysis that placental transfer in the human occurs by the 12th week of gestation, with increasing fetal lead uptake throughout development. Highest levels occur in bone, kidney, and liver, followed by blood, brain, and heart.

Cord blood contains significant amounts of lead, generally correlating with maternal blood values and being slightly but significantly lower than their mothers' in concentration. (Scanlon, 1971; Harris and Halley, 1972; Gershanik et al., 1974; Buchet et al., 1978; Alexander and Delves, 1981; Rabinowitz and Needleman, 1982).

A cross-sectional study of maternal blood lead carried out by Alexander and Delves (1981) showed that a significant decrease in maternal blood lead occurs throughout pregnancy, a decrease greater than the dilution effect of the concurrent increase in plasma volume. Hence, during pregnancy there is either an increasing deposition of lead in placental or fetal tissue or an increased loss of body lead via other routes. Increasing uptake by the fetus during gestation as demonstrated by Barltrop (1969) suggests that the former

PRELIMINARY DRAFT

explanation is a likely one. Hunter (1978) found that summer-born children showed a trend to higher blood lead than those born in the spring, suggesting increased fetal uptake in the summer due to increases in circulating maternal lead. This observation was confirmed in the report of Rabinowitz and Needleman (1982).

Ryu et al. (1978) and Singh et al. (1978) both reported that infants born to women having a history of lead exposure has significantly elevated blood lead values at birth.

10.3 DISTRIBUTION OF LEAD IN HUMANS AND ANIMALS

A quantitative understanding of the sequence of changes in levels of lead in various body pools and tissues is essential in interpreting measured levels of lead with respect to past exposure as well as present and future risks of toxicity. This section discusses the distribution kinetics of lead in various portions of the body--blood, soft tissues, calcified tissues, and the "chelatable" or toxicologically active body burden--as a function of such parameters as exposure history and age.

A given quantity of lead taken up from the GI tract or the respiratory tract into the bloodstream is initially distributed in accordance with the rate of delivery by blood to the various organs and systems. Lead is then redistributed to organs and systems in proportion to their respective affinities for the element. With consistent exposure for an extended period, a near steady-state of intercompartmental distribution is achieved.

Fluctuations in the near steady-state will occur whenever short-term lead exposures are superimposed on a long-term uptake pattern. Furthermore, the steady-state description is imperfect because on a very short (hourly) time scale, intake is not constant. Lead intake with meals and changes in ambient air lead--outside to inside and vice versa--will cause quick changes in exposure levels which may be viewed as short-term alternations in the small, labile lead pool. Metabolic stress could remobilize and redistribute body stores, although documentation of the extent to which this happens is very limited (Chisolm and Harrison, 1956).

10.3.1 Lead in Blood

Viewed from different time scales, lead in whole blood may be seen as residing in several distinct, interconnected pools. More than 99 percent of blood lead is associated with the erythrocytes (De Silva, 1981; Everson and

PRELIMINARY DRAFT

Patterson, 1980; Manton and Cook, 1979) under typical conditions, but it is the very small fraction of lead transported in plasma and extracellular fluid which provides the lead available to the various body organs (Baloh, 1974).

Most of the erythrocyte lead is bound with the cell, although toxicity of the element to the erythrocyte (Raghavan et al., 1981) is mainly associated with membrane lead content. Within erythrocytes from non-exposed subjects, lead is mainly bound to hemoglobin, in particular HbA₂, which binds ca. 50 percent of cell lead although it comprises only 1-2 percent of total hemoglobin (Brueger et al., 1973). A further 5 percent is bound to a 10,000-dalton molecular weight fraction, about 20 percent to a much heavier molecule, and about 25 percent is considered "free" or bound to lower-weight molecules (Ong and Lee, 1980a; Raghavan and Gonick, 1977). Raghavan et al. (1980) have observed that among workers exposed to lead, those who develop signs of toxicity at relatively low blood lead levels seem to have a diminished binding of intracellular lead with the 10,000-dalton fraction, suggesting an impaired biosynthesis of a protective species. According to Ong and Lee (1980b), fetal hemoglobin has a higher affinity for lead than adult hemoglobin. Whole blood lead in daily equilibrium with other compartments was found to have a mean life of 35 days (25-day half-life) and a total content of 1.9 mg, based on studies with a small number of subjects (Rabinowitz et al., 1976). Chamberlain et al. (1978) established a similar half-time for lead-203 in blood when volunteers were given the label by ingestion, inhalation, or injection.

Alterations in blood lead levels in response to abrupt changes in exposure apparently occur over somewhat different periods, depending on whether the direction of change is greater or less. With increased lead intake, blood lead achieves a new value in ca. 60 days (Griffin et al., 1975; Tola et al., 1973), while a decrease may involve a longer period of time, depending on the magnitude of the past higher exposure (O'Flaherty et al., 1982; Rabinowitz et al. 1977; Gross, 1981). With age, there appears to be a modest increase in blood lead, Awad et al. (1981) reporting an increase of 1 µg for each 14 years of age.

In the latter case, particularly with occupational exposure, it appears that the time for re-establishing near steady-state is more dependent upon the extent of lead resorption from bone and the total quantity deposited, extending the "washout" interval.

PRELIMINARY DRAFT

Lead levels in newborn children are similar to but somewhat lower than those of their mothers, 8.3 vs. 10.4 µg/dl (Buchet et al., 1978), and 11.0 vs. 12.4 µg/dl (Alexander and Delves, 1981). Alexander and Delves (1981) also reported that maternal blood lead levels decrease throughout pregnancy, such decreases being greater than the expected dilution via the concurrent increase in plasma volume. These data are consistent with increasing fetal uptake during gestation (Barltrop, 1969). Increased tissue retention may also be a factor.

Levels of lead in blood are sex-related, adult women invariably showing lower levels than adult males (e.g., Mahaffey et al., 1979). Of interest in this regard is the study of Stuik et al. (1974) showing lower Pb-B response than men for an equivalent level of lead intake.

The small but biologically significant lead pool in blood plasma has proven technically difficult to measure and only recently have reliable values become available (see Chapter 9). Chamberlain et al. (1978) found that injected ²⁰³Pb was removed from plasma--and, by inference, extracellular fluid--with a half-life of less than 1 hour. These data support the observation of De Silva (1981) that oral lead is rapidly cleared from plasma. Ong and Lee (1980a) found that ²⁰³Pb, in their *in vitro* studies, is virtually all bound to albumin and only trace amounts to high-weight globulins. It is not possible to state which binding form constitutes an "active" fraction for movement to tissues.

Although Rosen et al. (1974) reported that plasma lead was invariant across a range of whole blood levels, the findings of Everson and Patterson (1980), De Silva (1981), and Cavalleri et al. (1978) indicate that there is an equilibrium between red cell and plasma, such that levels in plasma rise with levels in whole blood. This is consistent with the data of Clarkson and Kench (1958) that lead in the red cell is relatively labile to exchange, and a logical prerequisite for a dose-effect relationship in various organs. Ong and Lee (1980c), furthermore, found that plasma calcium is capable of displacing RBC membrane lead, suggesting that plasma Ca is a factor in the cell-plasma lead equilibrium.

10.3.2 Lead Levels in Tissues

Of necessity, various relationships of tissue lead to exposure and toxicity in humans generally must be obtained from autopsy samples, although

PRELIMINARY DRAFT

in some studies, biopsy data have been described. There is, then, the inherent question of how such samples adequately represent lead behavior in the living population, particularly in cases where death was preceded by prolonged illness or disease states. Also, victims of fatal accidents are not well characterized as to exposure status, and are usually described as having no "known" lead exposures. Finally, these studies are, of necessity, cross-sectional in design. Some important aspects of the available data include the relative distribution of lead between soft and calcifying tissue, the effect of age and development on lead content of soft and mineral tissue, and the relationship between total and "active" lead burdens in the body.

10.3.2.1 Soft Tissues--In adults, after age 20, most soft tissues do not show age-related changes in lead levels, in contrast to the case with bone (Barry, 1975, 1981; Schroeder and Tipton, 1968; Butt et al., 1964). Kidney cortex shows increases in lead with age, associated with formation of lead nuclear inclusion bodies (Indraprosit et al., 1974). Based on these rates of accumulation, the total body burden may be divided into pools which behave differently: the largest and kinetically slowest pool is the skeleton, which accumulates lead with age; and the much more labile lead pool in soft tissue.

Soft tissue levels generally stabilize in early adult life and show a turnover rate similar to blood, sufficient to prevent accumulation except in the renal cortex, owing to formation of lead-containing nuclear inclusion bodies (Cramer et al., 1974; Indraprosit et al., 1974). The data of Gross et al. (1975) and Barry (1975) indicate that aortic levels appear to rise with age, although this may reflect entrapment of lead in atherosclerotic deposits.

Biliary and pancreatic secretions, while presumably reflecting some of the organ levels, have tracer lead concentrations distinct from either blood or bone pools (Rabinowitz et al., 1973).

For relative levels of lead in soft tissue, the reports of Barry (1975, 1981), Gross (1975), and Horiuchi et al. (1958) indicate that soft tissue lead content generally is below 0.5 parts per million wet weight, with higher values for aorta and kidney cortex. The higher values in aorta presumably reflect lead in plaque deposits, while higher kidney levels may be associated with the presence of lead-accumulating tubular cell nuclear inclusions.

The relatively constant lead concentration in lung tissue across age groups suggests no accumulation of respired lead and is consistent with data

PRELIMINARY DRAFT

for deposition and absorption (see previous section). Brain tissue was generally under 0.2 ppm wet weight and appeared to show no change with increasing age. Since these data were collected by cross-sectional study, age-related changes in the low levels of lead in brain would have been difficult to discern. Barry (1975) found that tissues in a small group of samples from subjects with known or suspected occupational exposure showed higher lead levels in aorta, liver, brain, skin, pancreas, and prostate.

Levels of lead in whole brain are less illuminating to the issue of sensitivity of certain regions within the organ to toxic effects of lead than is regional analysis. The relative distribution of lead across brain regions has been reported from various laboratories and the relevant data for humans and animals are set forth in Table 10-2.

The data of Grandjean (1978) and Niklowitz and Mandybur (1975) for human adults and that of Okazaki et al. (1963) for autopsy samples from young children dying of lead poisoning are relatively consistent in showing that lead is selectively accumulated in the hippocampus. The correlation of lead level with that of potassium suggests that uptake of lead is greater in cellulated areas. The involvement of the cerebellum in lead encephalopathy in children (see Section 12.4) and in adult intoxication with occupational exposure indicates that both relative sensitivity of brain regions to lead as well as relative uptake are factors in lead neuropathology.

In adult rats, the hippocampus also shows selective uptake of lead into hippocampus (Fjerdingstad et al., 1974; Danscher et al., 1975) and the amygdala (Danscher et al., 1975). By contrast, control or lead-exposed neonate rats show greatest uptake of lead into cerebellum, followed by cerebral cortex, then brainstem plus hippocampus. Hence, there is a developmental difference in relative lead distribution in the rat with or without increased lead exposure (Klein and Koch, 1981).

In studies of young dogs, unexposed animals showed highest levels in the cerebellum, while lead exposure was associated with selective uptake into gray matter, cerebellar level being relatively low. Unlike the young rat, then, the distribution of lead in brain regions appears to be dose-dependent (Stowe et al., 1973).

Barry (1975, 1981) compared lead levels in soft tissues of children vs. adults. Tissue lead of infants under 1 year old was generally lower than in

PRELIMINARY DRAFT

TABLE 10-2. REGIONAL DISTRIBUTION OF LEAD IN HUMANS AND ANIMALS

Species	Exposure Status	Relative Distribution	Reference
<u>Humans</u>			
Adult Males	Unexposed	Hippocampus $\cong$ amygdala > medulla oblongata > half brain > optic tract $\cong$ corpus callosum. Pb correlated with K.	Grandjean, 1978
Children	fatal lead poisoning	hippocampus > frontal cortex >> occipital white matter, pons	Okazaki et al., 1963
Child, 2 y. old	fatal lead poisoning	cortical gray matter > basal ganglion > cortical white matter	Klein et al., 1970
Adults 10-21	3 subjects unexposed 1 subject with lead poisoning as child	hippocampus > cerebellum $\cong$ temporal lobes > frontal cortex in 3 unexposed subjects; temporal lobes > frontal cortex > hippocampus > cerebellum > in case with prior exposure	Niklowitz and Mandybur, 1975
<u>Animals</u>			
Adult rats	Unexposed	hippocampus > amygdala >> whole brain	Danscher et al., 1975
Adult rats	Unexposed	Hippocampus has 50 percent of brain lead with a 4:1 ratio of hippocampus:whole brain	Fjendingstad et al., 1974

TEH 0530808

DUP050031721

PRELIMINARY DRAFT

TABLE 10-2 (cont.)

Species	Exposure Status	Relative Distribution	Reference
Neonatal rats	Controls and daily i.p. injection, 5.0 or 7.5 mg/kg	In both treated and control animals: cerebellum > cerebral cortex > brainstem + hippocampus	Klein and Koch, 1981
Young dogs	Controls and dietary exposure, 100 ppm; 12 weeks of exposure	Controls: cerebellum $\approx$ medulla > caudate > occipital gray > frontal gray Exposed: occipital gray > frontal gray $\approx$ caudate > occipital white $\approx$ thalamus > medulla > cerebellum	Stowe et al., 1973

PRELIMINARY DRAFT

older children, having a mean of 0.3 ppm, while children aged 1-16 years had values which were comparable to adult women. In the Barry (1981) study, the absolute concentration of lead in brain cortex or the brain cortex: blood lead ratios did not appear to be different in infants or older children compared to adults. Such direct comparisons do not account for relative tissue mass changes with age, but this factor is comparatively less with soft tissue than with the skeletal system (see Excretion and Retention section).

Subcellular distribution of lead in soft tissue is not uniform, with relatively high amounts of lead being sequestered in mitochondria and nucleus. Cramer et al. (1974) studied renal biopsy tissue in lead workers having exposure of variable duration and observed lead-binding nuclear inclusion bodies in renal proximal tubules subjects having short exposure, with all showing mitochondrial changes. A considerable body of animal data (see Animal Studies) document the selective uptake of lead into these organelles. Pounds and Wright (1982) describe these organellar pools in kinetic terms as having half-lives of relatively short duration in cultured rat hepatocytes, while McLachlin et al. (1980) found that rat kidney epithelial cells form lead-sequestering nuclear inclusions within 24 hours.

10.3.2.2 Mineralizing Tissue--Biopsy and autopsy data have shown that lead becomes localized and accumulates in human calcified tissues: bones and teeth. The accumulation begins in fetal development (Barltrop, 1969; Horiuchi et al., 1959).

Total lead content in bone may exceed 200 mg in men aged 60-70 years, but in women, the accumulation is somewhat lower. Various investigators (Barry, 1975; Horiguchi and Utsonomiya, 1973; Schroeder and Tipton, 1968; Horiuchi et al., 1959) have documented that ca. 95 percent of total body lead is lodged in bone. These reports not only establish the affinity of bone for lead, but also provide evidence that lead increases in bone until 50-60 years, a later fall-off reflecting some combination of diet and mineral metabolism changes. Tracer data show accumulation in both trabecular and compact bone (Rabinowitz et al., 1976).

In adults, bone lead is the most inert pool as well as the largest, and accumulation can serve to maintain elevated blood lead levels years after past, particularly occupational, exposure has ended. This accounts for the observation that duration of exposure correlates with the rate of reduction of blood lead after termination of exposure (O'Flaherty et al., 1982).

PRELIMINARY DRAFT

The relative proportion of body lead lodged in bone is reported to be lower in children than in adults, although concentrations of lead in bone increase more rapidly than in soft tissue during childhood (Barry, 1975, 1981). In 23 children, bone lead was 9 mg, or 73 percent of total body burden vs. a figure of 94 percent in adults. Expression of lead in bone in terms of concentration across age groups, however, does not accommodate the "dilution" factor, which is quite large for the skeletal system in children (see Excretion and Retention section).

The isotope kinetic data of Rabinowitz et al. (1976) and Holtzman (1978) indicate biological half-times of lead in bone of the order of several decades, although it appears that there are two bone compartments, one of which is a repository for relatively labile lead (Rabinowitz et al., 1977).

Tooth lead levels also increase with age at a rate proportional to exposure (Steenhout and Pourtois, 1981), and are also roughly proportional to blood lead levels in man (Winnecke, 1981) and experimental animals (Kaplan et al., 1980).

Dentine lead is perhaps the most responsive component of dentition to lead exposure since it is laid down from the time of eruption until the tooth is shed. Needleman and Shapiro (1974) have documented the utility of dentine lead as an indicator of the degree of subject exposure. Fremlin and Edmonds (1980), using alpha particle excitation and micro-autoradiography, have shown dentine zones of lead enrichment related to abrupt changes in exposure.

The rate of lead deposition in dentition appears to vary with the type of tooth, being highest in the central incisors and lowest in molars, a difference which must be taken into account when using tooth lead data for exposure assessment, particularly for low levels of lead exposure (Mackie et al., 1977; Delves et al., 1982).

10.3.3 Chelatable Lead

Relatively mobile lead in organs and systems is potentially more "active" toxicologically in terms of being available to sites of action. Hence, diffusible, mobilizable, or exchangeable lead may each be a more significant predictor of imminent toxicity or recent exposure than total body or whole blood burdens. In reality, however, these would be quite difficult assays.

In this regard, "chelatable" urinary lead has been shown to provide an index of this mobile portion of total body burden and chelation challenge is

PRELIMINARY DRAFT

now viewed as the ultimate probe of undue body burden in children and adults (CDC, 1978; WHO, 1977; Chisolm and Barltrop, 1979; Chisolm et al., 1976; Saenger et al., 1982; Hanson et al., 1981). In general, the amount of plumburesis associated with chelant challenge is related to the dose and the schedule of administration.

A quantitative description of inputs to the fraction of body lead that is chelatable from various body compartments is difficult to fully define, but it very likely includes a sizable, fairly mobile compartment within bone as well as soft tissues, based on: 1) the fact that the amount of lead mobilized by chelation is age dependent (Araki, 1973; Araki and Ushio, 1982) in non-exposed adults while blood and soft tissue lead levels are not (Barry, 1975), indicating a lead pool labile to chelation but kinetically distinct from soft tissue; 2) the studies of chelatable lead in animals (Hammond, 1971, 1973) suggesting removal of some bone lead fraction and the response of explanted fetal rat bone lead to chelants (Rosen and Markowitz, 1980); 3) the tracer modeling estimates of Rabinowitz et al. (1977) suggesting a mobile bone compartment; and 4) the complex, non-linear relationship of lead intake by air, food, and water (see next chapter) to blood lead, as well as the exponential relationship of chelatable lead to blood lead (Chisolm et al., 1976).

The logarithmic relationship of chelatable to blood lead in children (Chisolm et al., 1976) is consistent with the studies of Saenger et al. (1982), who reported that levels of mobilizable lead in "asymptomatic" children with moderate elevations in blood lead were quite similar in many cases to those values obtained in children with signs of overt toxicity. Hansen et al. (1981) report that lead workers challenged with CaNa_2EDTA show 24-hour urine lead levels that in many cases exceed the accepted limit levels even though blood lead was only moderately elevated in many of those workers. The action level corresponded, on the regression curve, to a blood value of 35 $\mu\text{g/dl}$.

Several reports provide insight into the behavior of labile lead pools in children treated with chelating agents over varying periods of time. Treatment regimens using CaNa_2EDTA or $\text{CaNa}_2\text{EDTA} + \text{BAL}$ for up to 5 days invariably are associated with "rebound" in blood lead, ascribed to a redistribution of lead among mobile lead compartments (Chisolm and Barltrop,

PRELIMINARY DRAFT

1979). Marcus (1982) reported that 41 children given oral D-penicillamine for 3 months showed a significant drop in blood lead (mean initial value of 53.2 $\mu\text{g/dl}$) by 2 weeks, a slight rise which was within measurement error with a peak at 4 weeks, and a fall to 6 weeks, followed by no further change at a blood lead of 36 $\mu\text{g/dl}$. Hence, there was a near steady-state at an elevated level for 10 of the 12 weeks with continued treatment. The author states that effort was made to limit further lead intake as much as possible. From these reports, it appears that a re-equilibration does occur, varying in characteristics with type and duration of chelation. The rebound seen in short-term treatment with CaNa_2EDTA or $\text{CaNa}_2\text{EDTA-BAL}$, although attributed to soft tissue, could well include shift of lead from a larger mobile bone compartment to soft tissues and blood. The apparent steady state between the blood lead pool and other compartments that is achieved in the face of plumburesis, induced by D-penicillamine (Marcus, 1982), suggests a rather sizable labile body pool which, in quantitative terms, would appear to exceed soft tissue alone.

10.3.4 Mathematical Descriptions of Physiological Lead Kinetics

In order to account for observed kinetic data and make predictive statements, a variety of mathematical models have been suggested, including those describing "steady state".

Tracer experiments have suggested compartmental models of lead turnover based on a central blood pool (Holtzman, 1978; Rabinowitz et al., 1976; and Batschelet et al., 1979). These have hypothesized well-mixed, interconnected pools and utilized coupled differential equations with linear exponential solutions to predict blood and tissue lead exchange rates. Were lead to be retained in these pools in accordance with a power-law distribution of residence times, rather than being uniform, a semi-markov model is more appropriate (Marcus, 1979).

Lead pools with turnover quicker than whole blood, on the order of minutes, have been detected within isolated cells (Pounds and Wright, 1982). Evidence of an extracellular lead pool in humans exists in observations of lead plasma (De Silva, 1981) or urine (Rabinowitz et al., 1974) after oral lead exposure as well as from lead-203 studies with injection, ingestion, and inhalation (Chamberlain and Heard, 1981). No single model has been developed which utilizes what has been learned of lead behavior in these highly labile pools which exist around and within permanent and concentrated sites.

PRELIMINARY DRAFT

Existing steady-state models are also deficient, not only because they are based on small numbers of subjects but also because there may be a dose dependency for some of the interpool transfer coefficients. In this case, a non-linear dose-response model would be more appropriate when considering changes in blood lead levels. For example, the blood lead/air lead relationship (Hammond et al., 1981) as well as that for diet (Department of Environment, 1982) and tap drinking water (Sherlock et al., 1982) are all non-linear in mathematical form. In addition, alterations in nutritional status or the onset of metabolic stresses can complicate steady-state relationships.

The above discussions of both the non-linear relationship of intake to the blood lead pool and the non-linear relationship of chelatable, or toxicologically active, lead to blood levels logically indicate that intake at elevated levels can add substantially to this chelatable pool and be substantially unrecognized in blood lead measurements.

10.3.5 Animal Studies

The relevant questions to be asked of animal data are those which cannot be readily or fully satisfied in human subjects: (1) what is the effect of exposure level on relative distribution within the body at specific time points? (2) what is the relationship of age or developmental stage on relative distribution of lead to organs and systems, particularly the nervous system? (3) what are the relationships of physiological stress and nutritional status to the redistribution kinetics? and (4) can one better define the relationship of chelatable lead to such indicator lead pools as blood?

Administration of a single dose of lead to rats produces high initial lead concentrations in soft tissues, which then fall rapidly as the result of excretion and transfer to bone (Hammond, 1971), while the relative distribution of lead was seen to be relatively independent of the dose.

Castellino and Aloj (1964) reported that single dose exposure of rats to lead was associated with a fairly constant ratio of red cell to plasma, a rapid distribution to tissues and relatively higher uptake in liver, kidney, and particularly bone. Lead loss from organs and tissues follow first-order kinetics except for bone.

PRELIMINARY DRAFT

The data of Morgan et al. (1977), Castellino and Aloj (1964), and Keller and Doherty (1980a) document that the skeletal system in rats and mice is the kinetically rate-limiting step in whole-body lead clearance.

Subcellular distribution studies involving either tissue fractionation after in vivo lead exposure or in vitro data document that lead is preferentially sequestered in the nucleus (Castellino and Aloj, 1964; Goyer et al., 1970) and mitochondrial fractions (Castellino and Aloj, 1964; Barltrop et al., 1974) of cells from lead-exposed animals. Lead enrichment in the mitochondria is consistent with the high sensitivity of this organelle to the toxic effects of lead.

The neonate animal can be characterized as retaining proportionately higher levels of tissue lead compared to the adult (Goldstein et al., 1974; Momcilovic and Kostial, 1974; Mykkänen et al., 1979; Klein and Koch, 1981) and showing slow decay of brain lead levels while other tissue levels show significant decreases over time. This appears to be the result of enhanced entry by lead due to a poorly developed brain barrier system in the developing animals as well as enhanced body retention in the young animals. The effects of such changes as metabolic stress and nutritional status may be noted in the literature.

Keller and Doherty (1980b, 1980c) have documented that tissue redistribution of lead, specifically bone lead mobilization, occurs in lactating female mice, both lead and calcium transfer occurring from mother to pups. Changes in lead movement from body compartments, particularly bone, with changes in nutrition are described in the Interactions section of this chapter.

In studies with rats which are relevant both to the issue of chelatable lead vs. lead indicators in humans and to the relative lability of lead in the young vs. the adult, Jugo et al. (1975b) and Jugo (1980) studied the relative chelatability of lead in neonate vs. adult rats and the relative lability in the erythrocyte. Challenge of young rats by metal chelants yields proportionately lower levels of urinary lead than in the adult, which has been ascribed to tighter binding of lead in the young animal (Jugo et al., 1975). In a related observation, the chelatable fraction of lead bound to erythrocytes of young animals given ^{203}Pb was ca. 3-fold greater than in the adult rat (Jugo, 1980), although the relative fraction of dose in the cells

PRELIMINARY DRAFT

was higher in the suckling rat. The difference in the suckling rat erythrocyte in terms of binding of lead and relative content compared to the adult may be compared to the observation of Ong and Lee (1980b) that human fetal hemoglobin binds lead more avidly than does the mature form.

10.4 LEAD EXCRETION AND RETENTION IN HUMANS AND ANIMALS

Dietary lead in humans and animals which is not absorbed passes through the GI tract and is eliminated with feces, as is that fraction of air lead which is swallowed and not absorbed. Lead absorbed into the blood stream and not retained undergoes excretion through the renal and gastrointestinal tracts, the latter by biliary clearance. The relative amounts appearing in urine and feces appear to be a function of such factors as species, age, and differences in dosing. Of significance in this discussion is the age or developmental stage dependency of lead excretion and retention.

10.4.1 Human Studies

Booker et al. (1969) found that ^{212}Pb injected into 2 adult volunteers led to initial appearance of label first in urine (4.4 percent of dose in 24 h.), then in both urine and feces in ca. equal amounts.

By use of the stable isotope ^{204}Pb , Rabinowitz et al. (1973) reported that urinary and fecal excretion of the label amounted to 38 and 8 $\mu\text{g/day}$ in adult subjects, accounting for 76 and 16 percent, respectively of the measured recovery. Fecal excretion was ca. twice that of all the remaining modes of excretion: hair, sweat, and nails (8 percent).

Perhaps the most detailed study of lead excretion in adult humans is that of the Harwell, England, group (Chamberlain et al., 1978), who used ^{203}Pb administered by injection, inhalation, and ingestion. Following injection of the label or oral intake, the relative amounts in urine (Pb-U) and feces (Pb-F_e, endogenous fecal lead) were comparable for the two administration routes, endogenous fecal lead being 50 percent of that in urine, or a 2:1 ratio of urinary/fecal lead, after allowing for increased transit time of fecal lead through the GI tract.

Based on the metabolic balance and isotope excretion data of Kehoe (1961a,b,c), Rabinowitz et al. (1976) and Chamberlain et al. (1978), as well as some recalculations of the Kehoe and Rabinowitz data by Chamberlain et al., it appears that short-term lead excretion amounts to 50-60 percent of the absorbed fraction, the balance moving mainly to bone with some subsequent

PRELIMINARY DRAFT

fraction, ca. half, of this stored amount eventually being excreted. The rapidly excreted fraction was determined by Chamberlain et al. (1978) to have a half-time of excretion of ca. 19 days. This is consistent with the estimates of Rabinowitz et al. (1976), who expressed clearance in terms of mean-times. Mean-times are multiplied by $\ln 2$ (0.693) to arrive at half-times. The similarity of blood ^{203}Pb half-times with that of body excretion seen by Chamberlain et al. (1978) indicate a steady rate of clearance from the body.

The age dependency of lead excretion rates in humans has not been well studied, all of the above lead excretion data involving adults. Table 10-3 combines available data from adults and infants for purposes of comparison. Intake, urine, fecal, and endogenous fecal lead data from 2 studies involving adults and one report with infants are used. For consistency in the adult data, 70 kg is taken as average adult weight, and a $\text{Pb-f}_e/\text{Pb-U}$ value of 0.5 used. Lead intake, absorption, and excretion is expressed as $\mu\text{g Pb/kg/day}$. For the Ziegler et al. (1978) data with infants, endogenous fecal lead excretion is calculated using the adult ratio as well as the extrapolated value of $1.5 \mu\text{g Pb/kg/day}$. The respiratory intake value for the infants is an upper value ($0.2 \mu\text{g Pb/m}^3$) since Ziegler et al. found air lead to be $<0.2 \mu\text{g/m}^3$. In comparison with the two representative adult groups, infants appear to have a lower total excretion rate, although the relative excretion of endogenous fecal lead may be higher than for adults.

Lead is accumulated in the human body with age, mainly in bone, up to ca. 60 years of age, when decrease occurs with changes in intake as well as in bone mineral metabolism. Total accumulation by 60 years of age ranges up to ca. 200 mg (see review of Barry, 1978), although occupational exposure can raise this figure several-fold (Barry, 1975). Holtzman (1978) has reviewed the available literature on studies of lead retention in bone. In normally exposed humans a biological half-time of ca. 17 years has been calculated, while data for uranium miners yield a range of 1320-7000 days. Chamberlain et al. (1978) have estimated life-time averaged daily retention at $9.5 \mu\text{g}$ using data of Barry (1975). Within shorter time frames, however, it should be noted that retention can vary considerably due to such factors as disruption in the individual's equilibrium with lead intake at a given level of exposure, the differences between children and adults, and in elderly subjects the presence of osteoporosis (Gross and Pfitzer, 1974).

PRELIMINARY DRAFT

TABLE 10-3. RELATIVE EXCRETION AND RETENTION
RATES^a IN ADULTS AND INFANTS

	Children ^b	Adult Group A ^c	Adult Group B ^d
Dietary intake (µg/kg)	10.76	3.63	3.86
Fraction absorbed ^e	0.46 (0.55) ^f	0.15 ^g	0.15 ^g
Diet lead absorbed (µg/kg)	4.95 (5.92)	0.54	0.58
Air lead absorbed (µg/kg)	0.20	0.21	0.11
Total absorbed lead (µg/kg)	5.15 (6.12)	0.75	0.68
Daily urinary Pb (µg/kg)	1.00	0.47	0.34
Ratio: urinary/absorbed Pb	0.19 (0.16)	0.62	0.50
Endogenous fecal Pb	0.5 (1.56) ^h	0.24 ⁱ	0.17 ⁱ
Total excreted Pb	1.50 (2.56)	0.71	0.51
Ratio: total excreted/ absorbed Pb	0.29 (0.42)	0.92	0.75
Fraction of intake retained	0.34 (0.33)	0.01	0.04

^a µg/kg/day

^b Ziegler et al., 1978

^c Rabinowitz et al., 1977

^d Thompson, 1971, and estimates of Chamberlain et al., 1978

^e corrected for endogenous fecal Pb; $Pb-F_e = 0.5 \times Pb-U$

^f corrected for endogenous fecal Pb at extrapolated value from Ziegler et al., 1978

^g corrected for $Pb-F_e$

^h extrapolated value for end fecal Pb of 1.56

ⁱ for a ratio of 0.5, $Pb-F_e/Pb$

PRELIMINARY DRAFT

Lead labelling experiments, such as that of Chamberlain et al. (1978) indicate a short-term or initial retention of ca. 40-50 percent of the fraction absorbed, bone taking up much of this amount. It is difficult to determine how much lead resorption from bone will eventually occur using labeled lead, given the extremely small fraction of labeled to unlabeled lead, i.e., label dilution, which would exist. Based on the estimates of Kehoe (1961a,b,c), the Gross (1981) evaluation of the Kehoe studies, Rabinowitz et al. (1976), the Chamberlain et al. (1978) assessments of the aforementioned reports, and the data of Thompson (1971), ca. 25 percent of the lead absorbed daily undergoes long-term bone storage.

The above estimates relate to either adults or long-term retention over most of an individual's lifetime. Studies with children and the developing animal (see Animal Studies below) indicate lead retention in childhood can be higher than in adults. By means of metabolic balance studies, Ziegler et al. (1978) obtained a retention figure, as percentage of total intake, of 31.5 percent for infants, while the study of Alexander et al. (1973) provided an estimate of 18 percent. Corrected retention data for both total and absorbed intake for the pediatric subjects of Ziegler et al. (1978) are noted in Table 10.2, using the two values for endogenous fecal excretion as noted. Barltrop and Strehlow (1978) calculated a net negative lead retention in their children, but problems in comparison of this report with the others was noted above. Given the increased retention of lead in children relative to adults as well as the greater rate of lead intake on a body weight basis, increased uptake in soft tissues and/or bone is indicated.

Barry (1975, 1981) measured the lead content of soft and mineral tissue in a small group of autopsy samples from children 16 years and under and noted that average soft tissue values are comparable to those in female adults while mean bone lead values are lower than in adults, suggesting that bone in children has less retention capacity for lead than adults. It should be noted, however, that "dilution" of bone lead will occur because of the significant growth rate of the skeletal system through childhood. Trotter and Hixon (1974) studies changes in skeletal mass, density, and mineral content as a function of age, and noted that skeletal mass increases exponentially in children until the early teens, increases less until the early 20s, levels off in adulthood, and then slowly decreases. From infancy to the late teens, bone

PRELIMINARY DRAFT

mass increases up to 40-fold. Barry (1975) noted an approximate doubling in bone lead concentration over this approximate interval, indicating that total skeletal lead had actually increased 80-fold, and obtained a mean total bone lead content up to 16 years of ca. 8 mg, compared to a value of ca. 18 mg estimated from both the bone concentrations in Barry's study at different ages, and the bone growth data of Trotter and Hixon (1974). In a latter study (Barry, 1981), autopsy samples from infants and children between 1-9 years old showed an approximate 3.5-fold increase in mean bone values across the 3 bone types studied, compared to a skeletal mass increase from 0-6 mos. to 3-13 years old of greater than 10-fold, for an estimated total lead ratio of ca. 35-fold. Five reports (see Barry, 1981) noted age vs. tissue lead relationships indicating that overall bone lead levels in infants and children were less than in adults while 4 reports observed comparable levels in children and adults.

If one estimates total daily retention of lead in the infants studied by Ziegler et al. (1978), using a mean body weight of ca. 10 kg and the corrected retention rate in Table 10.3, one obtains a total daily retention of ca. 40 μ g Pb. By contrast, the total reported or estimated skeletal lead as accumulated between 2-14 years is 8-18 mg (vide supra), which averages out to a daily long-term retention of 2.0-4.5 μ g/day or 6-13 percent of total retention. It may be the case that lead retention is highest in infants, up to ca. 2 years of age (the subjects of the Ziegler et al. study), decreasing in older children. The mean retention in the Alexander et al. (1973) study was 18 percent, ca. half that seen by Ziegler et al. (1978), the difference possibly owing to the greater age range in the former study.

Of interest in this regard is the fact that "normal" blood lead levels in children either parallel (adult males) or are ca. 30 percent greater (adult females) than adults (Chamberlain et al., 1978), indicating that (1) the soft tissue lead pool in very young children is not greatly elevated and thus, (2) that there is a huge labile lead pool in bone which is still kinetically quite distinct from soft tissue lead, or that (3) in young children, blood lead is a much less reliable indicator of greatly elevated soft tissue or labile bone lead than is the case with the adult. Barry (1981) found that soft tissue lead levels were comparable in infants $\leq$ 1 year old, children 1-5 and 6-9 years old.

PRELIMINARY DRAFT

Given the implications of the above discussion, that retention of lead in the young child is higher than in adults and possibly older children, while at the same time their skeletal system is less effective for long-term lead sequestration, the very young child is at greatly elevated risk to a toxicologically "active" lead burden compared to older individuals. A more detailed discussion of this is given in Chapter 13.

10.4.2 Animal Studies

In rats and other experimental animals, both urinary and fecal excretion appear to be important routes of lead removal from the organism, the relative partitioning between the two modes being species and dose dependent.

Morgan et al. (1977), using ^{203}Pb injected into adult rats, noted that lead initially appears in urine, followed by equivalent elimination by both routes, and by 5 days, is proportionately higher in feces. Castellino and Aloj (1964), using ^{210}Pb , observed that fecal excretion was ca. twice that of urine, 35.7 vs. 15.9 percent, by 14 days. In the report of Klaasen and Shoeman (1974), relative excretion by the two routes was seen to be dose-dependent up to 1.0 mg/kg, being much higher by biliary clearance into the gut. At 3.0 mg/kg, ca. 90 percent of the excreted amount was measured in feces. The relatively higher proportion appearing in feces in the studies of Castellino and Aloj (1964) and Klaasen and Shoeman (1974), compared to the results of Morgan et al. (1978), is possibly due to carrier dosing used, the latter workers using carrier-free injections. Hence, it appears that increasing dose does favor biliary excretion, as noted by Klaasen and Shoeman (1974).

With regard to species differences, Klaasen and Shoeman (1974) found that the relative amount of biliary clearance was ca. 2 percent in dogs versus rats, while rabbits showed 50 percent of the rate of the rat at equivalent dosing. These data for the dog are in contrast to the results of Lloyd et al. (1975), who observed 75 percent of the excreted lead eliminated through biliary clearance. It should be noted that the latter workers used carrier-free label while the other investigation used injections with carrier at 3.0 mg Pb/kg levels. In mice, Keller and Doherty (1980a) observed that the cumulative excretion rate of ^{210}Pb in urine was 25-50 percent that in feces. In nonhuman primates, Cohen (1970) observed that baboons excreted lead at the rate of 40 percent in feces and 60 percent in urine. Pounds et al. (1978)

PRELIMINARY DRAFT

noted that the Rhesus monkey showed 30 percent of lead lost by renal excretion and 70 percent in feces. This may also be reflecting a carrier dosing difference.

The extent of total lead excretion in experimental animals, usually given labeled lead orally or parenterally, varies, in part owing to the time frames for post-exposure observation.

In the adult rat, Morgan et al. (1977) found that 62 percent of the injected label, ^{203}Pb , was excreted by 6 days. By 8 days, 66 percent of injected ^{203}Pb was eliminated in the adult rat study of Momcilovic and Kostial (1974), while the ^{210}Pb excretion data of Castellino and Aloj (1964) for the adult rat showed 52 percent excreted by 14 days. Similar data were obtained by Klaasen and Shoeman (1974). Lloyd et al. (1975) found that dogs excreted 52 percent of injected lead label by 21 days, 83 percent by 1 year, and 87 percent by 2 years. In adult mice (Keller and Doherty, 1980a), 62 percent of injected lead label was eliminated by 50 days. In the nonhuman primate, Pounds et al. (1978) measured ca. 18 percent excretion in adult Rhesus monkeys by 4 days.

Kinetic studies of lead elimination in experimental animals indicate that excretion is described by 2 or more components. In the rat, Morgan et al. (1977) estimated, from the elimination data of Momcilovic and Kostial (1974), that the excretion curve obeys a 2-component exponential expression with half-times of 21 and 280 hours. In dogs, Lloyd et al. (1975) found that excretion could be described by 3 components, i.e., a sum of exponentials with half-times of 12 days, 184 days, and 4951 days. Keller and Doherty (1980) reported that the half-time of whole-body clearance of injected ^{203}Pb consisted of an initial rapid component and a much slower terminal component, the latter component in the adult mouse having a half-time of 110 days.

The excretion rate dependency on dosing level was investigated in several studies. Although Castellino and Aloj (1964) saw no difference in total excretion rate when label was injected with 7 or 100 μg of carrier, Klaasen and Shoeman (1974) did observe that excretion rate by biliary tract was dose dependent at 0.1, 1.0, and 3.0 mg Pb/kg, but urine values were not provided for obtaining estimates of total excretion. Momcilovic and Kostial (1974) saw increased rate of excretion into urine over the added carrier range of 0.1 to 2.0 μg Pb with no change in fecal excretion. In the report of Aungst et al.

PRELIMINARY DRAFT

(1981) there was no change in excretion rate in the rat over the injected lead dosing range of 1.0 to 15.0 mg/kg. It thus appears that rat urinary excretion rates are dose-dependent over a narrow range, <7 µg, while elimination of lead through biliary clearance is dose-dependent up to an exposure level of 3 mg Pb/kg.

Lead movement from lactating animals to their offspring via milk constitutes both a route of excretion for the mother and a route of exposure to lead for the young. Investigations directed at this phenomenon have involved both prior-plus-ongoing maternal lead exposure during lactation and the effects of just prior treatment. Keller and Doherty (1980b) exposed 2 groups of female rats to ²¹⁰Pb-labeled lead: one group for 105 days prior to mating; the second prior to and during gestation and nursing. During lactation, there was an overall loss of lead from the bodies of the lactating females compared to controls while the femur ash weights were inversely related to level of lead excretion, indicating that such enhancement is related to bone mineral metabolism. Lead transfer via milk was ca. 3 percent of maternal body burden, increasing with continued lead exposure during lactation. Lorenzo et al. (1977) found that blood lead in nursing rabbits given injected lead peaks rather rapidly (ca. 1 hour) while milk lead shows a continuous increase for ca. 8 days, at which point its concentration of lead is 8-fold higher than blood, indicating that lead transfer to milk can occur against a concentration gradient in blood. Momcilovic (1978) and Kostial and Momcilovic (1974) observed that transfer of ²⁰³Pb in the late stage of lactation readily occurs in the rat, with higher overall excretion of lead in nursing vs. control females. Furthermore, it appeared that the rate of lead movement to milk was dose-dependent over the added lead carrier range of 0.2-2.0 µg Pb.

The relative retention of lead in developing vs. adult animals has been investigated in several studies using rats, mice, and nonhuman primates.

Momcilovic and Kostial (1974) compared the kinetics of lead distribution in suckling vs. adult rats after injection of ²⁰³Pb. Over an 8-day interval, 85 percent of the label was retained in the suckling rat compared to 34 percent in the adult. Keller and Doherty (1980a) compared the levels of ²¹⁰Pb in 10-day-old mice with the adult, noting from the clearance half-times (vide supra) that lead retention was greater in the suckling animals than in the

PRELIMINARY DRAFT

adults. In both adult and young mice, the rate of long-term retention was governed by the rate of release of lead from bone, indicating that in the mouse, skeletal lead retention in the young is greater than in the adult. With infant and adult monkeys exposed to ^{210}Pb orally, Pounds et al. (1978) observed that at 23 days, the corresponding amounts of initial dose retained were 92.7 and 81.7 percent, respectively.

The studies of Rader and coworkers (1981; 1982, in press) are of particular interest in that they not only demonstrate that young experimental animals continue to show greater retention of lead in tissue when exposure occurs after weaning but also document that such retention occurs in terms of either uniform exposure (Rader et al., 1981) or uniform dosing (Rader et al., 1982) when compared to adult animals. With uniform exposure, 30-day old rats given lead in drinking water showed significantly higher lead levels in blood and higher percentages of dose in brain, femur, and kidney as well as higher indices of hematopoietic impairment vs. adult animals (ALA-U,EP). As a percentage of dose retained, tissues in the young animals were ca. 2-3-fold higher. In part, the difference is due to a higher ingestion rate of lead. However, in the uniform dosing study where this was not the case, an increased retention of lead still prevailed, the amount of lead in brain being ca. 50 percent higher in young vs. adult animals. Comparison of values in terms of percent retained is more meaningful for such comparisons, since the factor of changes in organ mass (vide supra) is accounted for.

Delayed excretion in the young animal may reflect an immature excretory system or tighter binding of lead in various body compartments.

10.5 INTERACTIONS OF LEAD WITH ESSENTIAL METALS AND OTHER FACTORS

Deleterious agents, particularly toxic metals such as lead, do not express their toxicokinetic or toxicological behavior in a physiological vacuum, but rather are affected by interactions of the agent with a variety of biochemical factors such as nutrients. Growing recognition of this phenomenon and its implications for lead toxicity in humans have prompted a number of studies, many of them relatively recent, which address both the scope and mechanistic nature of such interactive behavior.

10.5.1 Human Studies

In humans the interactive behavior of lead and various nutritional factors is appropriately viewed as being particularly significant with

PRELIMINARY DRAFT

children, since this age group is not only particularly sensitive to lead's effects but also represents the time of greatest flux in relative nutrient status.

Such interactions occur against a backdrop of rather widespread deficiencies in a number of nutritional components in children. While such deficiencies are more pronounced in lower income groups, they do exist across all socioeconomic strata. Mahaffey and Michaelson (1980) have summarized the 3 nutritional status surveys carried out in the United States for infants and young children: The Preschool Nutrition Survey; The Ten State Nutrition Survey; and the Health Assessment and Nutritional Evaluation Survey (NHANES I). The most recent body of data in this connection is the NHANES II study (Mahaffey et al., 1979), dietary information for which is to be reported. In the older surveys, iron deficiency was the most common nutritional deficit in children under 2 years of age, particularly children from low-income groups. Reduced vitamin C intake was noted in about one-third of the children, while sizable numbers of them had significantly reduced intakes of calcium. Owen and Lippmann (1977) reviewed the regional surveys of low-income groups within Hispanic, white, and black populations. In these groups, iron deficiency was a common finding, while low intakes of calcium and vitamins A and C were observed regularly. Hambidge (1977) concluded that zinc intake in low-income groups is generally inadequate, relative to recommended daily allowances.

Available data from a number of reports document the association of lead absorption with suboptimal nutritional status. Mahaffey et al. (1976) reported in summary form their studies showing that children with blood lead greater than 40 $\mu\text{g Pb/dl}$ had significantly lower ($p < 0.01$) intake of phosphorus and calcium compared to a control group, while iron intake in the two groups was comparable. This effort involved children 1-4 years old from an inner-city, low-income population with close matching for all parameters except the blood lead level. Sorrell et al. (1977), in their nutritional assessment of 1- to 4-year-old children with a range of blood lead levels, observed that blood lead content was inversely correlated with calcium intake, while children with blood lead levels $> 60 \mu\text{g/dl}$ had significantly lower intakes of calcium ($p < 0.001$) and vitamin D.

Rosen et al. (1981) found that children with elevated blood lead (33-120 $\mu\text{g Pb/dl}$) had significantly lower serum concentrations of the vitamin D

PRELIMINARY DRAFT

metabolite 1, 25-(OH) $_2$ D ($p < 0.001$) compared to age-matched controls, and showed a negative correlation of serum 1, 25-(OH) $_2$ D with lead over the range of blood leads measured. These observations and animal data (Barton et al., 1978a, see Animal Studies below) may suggest an increasingly adverse interactive cycle of 1, 25-(OH) $_2$ D, lead, and calcium in which lead reduces biosynthesis of the vitamin D metabolite, which leads to reduced stimulation of calcium binding protein (CaBP) and less uptake of calcium from the gut but greater uptake of lead, the increasing uptake of lead further reducing metabolite levels. Barton et al. (1978a) isolated 2 mucosal proteins in rat intestine, one of which bound mainly lead and was not vitamin D-stimulated; the second bound mainly calcium and was under vitamin control. The authors suggest direct site binding competition between lead and calcium in these proteins. Hunter (1978) investigated the possible interactive role of seasonal vitamin D biosynthesis in adults and children since it is a common observation that lead poisoning is seen more often in summer than in other seasons (See Hunter, 1977 for review). In children, seasonality accounts for 16 percent of explained variance of blood lead in black children, 12 percent in Hispanics, and 4 percent in whites. More recently, it has been documented that there is no seasonal variation in circulating levels of 1, 25-di(OH) vitamin D, the metabolite which affects the rate of lead absorption from the GI tract (Chesney et al., 1981). These results suggest that seasonality is related to changes in exposure.

Johnson and Tenuta (1979) determined that calcium intake was negatively correlated ($r = -0.327$, $p < 0.05$) with blood lead in 43 children aged 1-6 years. The high lead group also consumed less zinc than children with lower blood levels. Yip et al. (1981) found that 43 children with elevated blood lead (>30 $\mu\text{g/dl}$) and EP (>35 $\mu\text{g/dl}$) had an increased prevalence of iron deficiency as these two parameters increased. Children classed as CDC Ib and II had a 79 percent deficiency rate, while those in Class III were all iron-deficient. Chisolm (1981) demonstrated an inverse relationship between "chelatable" iron and chelatable body lead levels as indexed by urinary ALA levels in 66 children with elevated blood lead.

Watson et al. (1980) reported that adult subjects who were iron deficient (determined from serum ferritin measurement) showed a lead absorption rate 2-3 times greater than subjects who were iron replete.

PRELIMINARY DRAFT

In a group of 13 children, Markowitz and Rosen (1981) reported that the mean serum zinc levels in children with plumbism was significantly below the values seen in normal children. Chelation therapy reduces the mean level even further.

Chisolm (1981) reported that there was an inverse relationship between ALA-U and the amount of "chelatable" or systemically active zinc in 66 children challenged with EDTA and with blood lead levels ranging from 45-60 μg Pb/dl. These 2 studies suggest that zinc status is probably as important an interactive modifier of lead toxicity as is either calcium or iron.

The role of nutrient in lead absorption has been reported in several metabolic balance studies for both adults and children. Ziegler et al. (1978), in their investigations of lead absorption and retention in infants, observed that lead retention was inversely correlated with calcium intake, expressed as either intake percentage ($r = -0.284$, $p < 0.01$) or on a weight basis ($r = -0.279$, $p < 0.01$). Of interest is the fact that the range of calcium intake measured was within the range deemed adequate for infants and toddlers by the National Research Council. These data also support the premise that severe deficiency need not be present to see an interactive relationship. Using adults, Heard and Chamberlain (1982) monitored the uptake of ^{203}Pb from the gut in 8 subjects as a function of the amounts of dietary calcium and phosphorus. Without supplementation with either of these minerals in fasting subjects, the label absorption rate was ca. 60 percent compared to 10 percent with 200 mg calcium plus 140 mg phosphorus, the amounts present in an average meal. Calcium alone reduced uptake by a factor of 1.3, phosphorus alone by 1.2, but both together yielded a reduction factor of 6. These workers suggest that insoluble calcium phosphate is formed and co-precipitates any lead present. This interpretation is supported by animal data (see below).

10.5.2 Animal Studies

Reports of lead-nutrient interactions in experimental animals have generally described such relationships in terms of a single nutrient, using relative absorption or tissue retention in the animal to index the effect. Much of the recent data is concerned with the impact of dietary levels of calcium, iron, phosphorus, and vitamin D. Furthermore, some investigators have attempted to elucidate the site(s) of interaction as well as the

PRELIMINARY DRAFT

mechanism(s) governing the interactions. Lead's interactions involve effect of nutrient on modulating lead uptake and effects as well as lead's effect on nutrients, but the focus of this discussion is on the former. These studies are tabulated in Table 10-4.

10.5.2.1 Interactions of Lead With Calcium--The early report of Sobel et al. (1940) noted that variation of dietary calcium and other nutrients affected the uptake of lead by bone and blood in animals. Subsequent studies by Mahaffey-Six and Goyer (1970) in the rat demonstrated that a considerable reduction in dietary calcium was necessary (to 0.1 percent from 0.7 percent), at which level blood lead was increased 4-fold, kidney lead content was elevated 23-fold, and relative toxicity (Mahaffey et al., 1973) was increased. The relative changes in calcium necessary to alter lead's effects in the rat appear to be greater than seen by Ziegler et al. (1978) in young children, indicating either species differences in terms of sensitivity of basic dietary differences as to levels of all interactive nutrients. These observations in the rat have been confirmed in the studies of Kostial et al. (1971), Quarterman and Morrison (1975), Barltrop and Khoo (1975), and Barton et al. (1978a). The inverse relationship between dietary calcium and lead uptake has also been noted in the pig (Hsu et al., 1975), horse (Willoughby et al., 1972), lamb (Morrison et al., 1977), and domestic fowl (Berg et al., 1980).

The mechanism(s) governing lead's interaction with calcium operate at both the gut wall and within body compartments. Barton et al. (1978a), using everted duodenal sac preparations in the rat, reported that: (1) interactions at the gut wall require the presence of intubated calcium to affect lead label absorption, pre-existing calcium deficiency in the animal and no added calcium having no effect on lead transport; (2) animals having calcium deficiency show increased retention of lead rather than absorption (confirmed by Quarterman et al., 1974); and (3) lead transport may be mediated by two mucosal proteins, one of which is high molecular weight, has a high proportion of bound lead and is affected in extent of lead binding with changes in lead uptake. The second protein binds mainly calcium and is vitamin D dependent.

Smith et al. (1978) found that lead is taken up at a different site in the duodenum of rats than calcium but uptake does occur at the site of phosphate uptake, suggesting a complex interaction of phosphorus, calcium, and

TABLE 10-4. EFFECT OF NUTRITIONAL FACTORS ON LEAD UPTAKE IN ANIMALS

Factor	Species	Index of Effect	Interactive Effect	Reference
Calcium	Rat	Lead in tissues and effect severity at low levels of dietary calcium	Low dietary calcium (0.1%) increases lead absorption and severity of effects	Mahaffey-Six and Goyer, 1970 Mahaffey et al., 1973
Calcium	Pig	Lead in tissues at low levels of dietary calcium	Increased absorption of lead with low dietary calcium	Hsu et al., 1975
Calcium	Horse	Lead in tissues at low levels of dietary calcium	Increased absorption of lead with low dietary calcium	Willoughby et al., 1972
Calcium	Lamb	Lead in tissue at low levels of dietary calcium	Increased absorption of lead with low dietary calcium	Morrison et al., 1977
Calcium	Rat	Lead retention	Retention increased in calcium deficiency	Barton et al., 1978a
Iron	Rat	Tissue levels and relative toxicity of lead	Iron deficiency increases lead absorption and toxicity	Mahaffey-Six and Goyer, 1972
Iron	Rat	Lead absorption in everted duodenal sac preparation	Reduction in intubated iron increases lead absorption; increased levels decrease lead uptake	Barton et al., 1978b
Iron	Mouse	Lead retention	Iron deficiency has no effect on lead retention	Hamilton, 1978

PRELIMINARY DRAFT

TABLE 10-4. (continued)

Factor	Species	Index of Effect	Interactive Effect	Reference
Iron	Rat	In utero or milk transfer of lead in pregnant or lactating rats	Iron deficiency increases both in utero and milk transfer of lead to sucklings	Cerklewski, 1980
Phosphorus	Rat	Lead uptake in tissues	Reduced P increased ²⁰³ Pb uptake 2.7-fold vs. controls	Barltrop and Khoo, 1975
Phosphorus	Rat	Lead retention	Low dietary P enhances lead retention; no effect on lead resorption in bone	Quarterman and Morrison, 1975
Phosphorus	Rat	Lead retention	Low dietary P enhances both lead retention and deposition in bone	Barton and Conrad, 1981
Vitamin D	Rat	Lead absorption using everted sac techniques	Increasing vitamin D increases intubated lead absorption	Smith et al., 1978
Vitamin D	Rat	Lead absorption using everted sac techniques	Both low and excess levels of vitamin D increase lead uptake by affecting motility	Barton et al., 1980
Lipid	Rat	Lead absorption	Increases in lipid (corn oil) content up to 40 percent enhances lead absorption	Barltrop and Khoo, 1975
Protein	Rat	Lead uptake by tissues	Both low and high protein in diet increase lead absorption	Barltrop and Khoo, 1975

PRELIMINARY DRAFT

TABLE 10-4. (continued)

Factor	Species	Index of Effect	Interactive Effect	Reference
Protein	Rat	Body lead retention	Low dietary protein either reduces or does not affect retention in various tissues	Quarterman et al., 1978b
Protein	Rat	Tissue levels of lead	Casein in diet increases lead uptake compared to soybean meal	Anders et al., 1982
Milk Components	Rat	Lead absorption	Lactose-hydrolyzed milk does not increase lead absorption, but ordinary milk does	Bell and Spickett, 1981
Milk components	Rat	Lead absorption	Lactose in diet enhances lead absorption compared to glucose	Bushnell and DeLuca, 1981
Zinc/Copper	Rat	Lead absorption	Low zinc in diets increases lead absorption	Cerklewski and Forbes, 1976 El-Gazzar et al., 1978
Zinc/Copper	Rat	Lead transfer in utero and in milk during lactation	Low-zinc diet of mother increases lead transfer in <u>utero</u> and in <u>maternal milk</u>	Cerklewski et al., 1979
Zinc/Copper	Rat	Lead absorption	Low copper in diet increases lead absorption	Klauder et al., 1973 Klauder and Petering, 1975

PRELIMINARY DRAFT

lead. This is consistent with data of Barltrop and Khoo (1975) for rats and the human studies of Heard and Chamberlain (1982), showing that the combined action of the two mineral nutrients is greater than the sum of either's effects.

Mykkänen and Wassermann (1981) observed that lead uptake in the intestine of the chick occurs in 2 phases: a rapid uptake (5 minutes) followed by a rate-limiting slow transfer of lead into blood. Conrad and Barton (1978) have observed a similar process in the rat. Hence, there is either a saturation process occurring, i.e., carrier-mediated transport, or simply lead precipitation in the lumen. In the former case, calcium interact to saturate the carrier proteins as isolated by Barton et al. (1978a), or may precipitate lead in the lumen by initial formation of calcium phosphate.

Quarterman and coworkers (1978a) observed that calcium supplementation of diet above normal also resulted in increased body retention of lead in the rat. Since both deficiency (Barton et al., 1978a) and excess in calcium intake enhance retention, two sites of influence on retention are suggested. Goyer (1978) has suggested that body retention of lead in calcium deficiency, i.e., reduced excretion rate, may be due to impairment of action of the kidney, while Quarterman et al. (1978) suggest that excess calcium suppresses calcium resorption from bone, hence also reducing lead release.

10.5.2.2 Interactions of Lead With Iron--Mahaffey-Six and Goyer (1972) reported iron-deficient rats had increased tissue levels of lead and manifested greater toxicity compared to control animals. This uptake change was seen with but minor change in hematocrit, indicating a primary change in lead absorption over the time of the study. Barton et al. (1978b) found that dietary restriction of iron, using ^{210}Pb and everted sac preparations in the rat, led to enhanced absorption of iron while iron loading suppressed the extent of lead uptake, using normal intake levels of iron. This suggests receptor binding competition at a common site, consistent with the isolation by these workers of 2 iron-binding mucosa fractions. While iron level of diet affects lead absorption, the effect of changes in lead content in the gut on iron absorption is not clear. Barton et al. (1978b) and Dobbins et al. (1978) observed no effect of lead in the gut on iron absorption in the rat, while Flanagan and coworkers (1979) reported that lead reduced iron absorption in mice.

PRELIMINARY DRAFT

In the mouse, Hamilton (1978) found that body retention of ^{203}Pb was unaffected by iron deficiency, using intraperitoneal administration of the label, while gastric intubation did lead to increased retention. Animals with adequate iron showed no changes in lead retention at intubation levels of 0.01 to 10 nmoles. Cerklewski (1980) observed that lead transfer both in utero and in milk to nursing rats was enhanced when dams were maintained on low iron diets compared to controls from gestation through lactation.

10.5.2.3 Lead Interactions With Phosphate--The early studies of Shelling (1932), Grant et al. (1938), and Sobel et al. (1940) documented that dietary phosphate influenced the extent of lead toxicity and tissue retention of lead in animals, low levels enhancing those parameters while excess intake retarded the effects.

Barltrop and Khoo (1975) reported that reduced phosphate increased the uptake of ^{203}Pb ca. 2.7-fold compared to controls. Quarterman and Morrison (1975) found that low dietary phosphate enhanced lead retention in rats but had no effect on skeletal lead mobilization nor was injected lead label affected by such restriction. In a related study (Quarterman et al., 1978a), it was found that doubling of the nutrient over normal levels resulted in lowering of lead absorption by ca. half. Barton and Conrad (1981) found that reduced dietary phosphorus increased the retention of labeled lead and deposition in bone, in contrast to the results of Quarterman and Morrison (1975). Increasing the intraluminal level of phosphorus reduced lead absorption, possibly by increasing intraluminal precipitation of lead as the mixed lead/calcium phosphate. In the report of Smith et al. (1978) it was found that lead uptake occurs at the same site as phosphate, suggesting that lead absorption may be more related to phosphate than calcium transport.

10.5.2.4 Interactions of Lead With Vitamin D--Several studies had earlier indicated that a positive relationship may exist between dietary vitamin D and lead uptake, resulting in either greater manifestations of lead toxicity or extent of lead uptake (Sobel et al., 1938, 1940).

Using the everted sac technique and testing with ^{210}Pb , Smith et al. (1978) observed that increasing levels of intubated vitamin D in the rat resulted in increased absorption of the label, uptake occurring at the distal end of the rat duodenum, the site of phosphorus uptake and greatest stimulation by the vitamin. Barton et al. (1980) used ^{210}Pb to monitor lead

PRELIMINARY DRAFT

absorption in the rat under conditions of normal, deficient, and excess amounts of dietary vitamin D. Lead absorption is increased with either low or excess vitamin D, apparently due to increased retention time of fecal mass containing the lead because of alteration of intestinal motility rather than by direct enhancement of mucosal uptake rate. Hart and Smith (1981) reported that vitamin D repletion of rat diet enhanced lead absorption (^{210}Pb) in the rat, while also enhancing femur and kidney lead uptake when the label was given by injection.

10.5.2.5 Interactions of Lead and Lipids--Barltrop and Khoo (1975) observed that varying of the lipid content of rat diet, from 5 up to 40 percent (corn oil) resulted in an increase of lead in blood 13.6-fold higher compared to the normal level with concomitant increase in lead levels in kidney, femur, and carcass. Reduction of dietary lipid below the 5 percent control figure was without effect on lead absorption rate. As an extension of this earlier work, Barltrop (1982, in press) has noted that the chemical composition of the lipid is a significant factor in affecting lead absorption. Study of triglycerides of saturated and unsaturated fatty acids showed that the polyunsaturate, trilinolein, increased lead absorption by 80 percent in rats, when given as 5 or 10 percent loadings in diet, compared to the monounsaturate triolein or any of the saturates in the series tricaproin to tristearin.

10.5.2.6 Lead Interaction With Protein--Quarterman et al. (1978b) have drawn attention to one of the inherent difficulties of measuring lead-protein interactions, the effect of protein on both growth and the toxicokinetic parameters of lead. Der et al. (1974) found that reduction of dietary protein, from 20 to 4 percent, led to increased uptake of lead in rat tissues, but the ca. 6-fold reduction in body weight over the interval of the study makes it difficult to draw any firm conclusions. Barltrop and Khoo (1975), using ^{203}Pb , found that lead uptake into rat tissue could be enhanced with either suboptimal or excess levels of protein in diet. Quarterman et al. (1978b) reported that retention of labeled lead in rats maintained on a synthetic diet containing ca. 7 percent protein was either unaffected or reduced compared to controls, depending on tissues taken for study.

It appears that not only levels of protein but also the type of protein affect tissue levels of lead. Anders et al. (1982) found that rats maintained on either of 2 synthetic diets varying only as to having casein or soybean

PRELIMINARY DRAFT

meal as protein source showed significantly higher lead levels in the casein group.

10.5.2.7 Interactions of Lead With Milk Components--For many years, milk was recommended prophylactically in lead poisoning among lead workers (Stephens and Waldron, 1975), although recent data suggest that milk may actually enhance lead uptake.

Kello and Kostial (1973) found that rats maintained on milk diets absorbed a greater amount of ^{203}Pb than those having access to commercial rat chow. This was ascribed to relatively lower levels of certain nutrients in milk compared to the rat chow. These observations were confirmed by Bell and Spickett (1981), who also observed that lactose-hydrolyzed milk was less effective than the ordinary form in promoting lead absorption, suggesting that lactose may be the enhancing principle. Bushnell and DeLuca (1981) demonstrated that lactose significantly increased lead (^{210}Pb) absorption and tissue retention by weanling rats by comparing diets identical in all respects except for carbohydrate source. These results provide one rationale for why nursing mammals tend to absorb greater quantities of lead than the adult, since lactose is the major carbohydrate source in suckling rats and is known to enhance the uptake of many essential metals.

10.5.2.8 Lead Interactions With Zinc and Copper--The studies of Cerklewski and Forbes (1976) and El-Gazzar et al. (1978) document the effect of zinc-deficient diets in promoting lead absorption in the rat, while repletion with zinc reduced lead uptake. The interaction continues within the body, particularly with respect to ALA-D activity (see Chapter 11). In a study of zinc-lead interactions in female rats during gestation and lactation, Cerklewski (1979) observed that zinc-deficient diets resulted in more transfer of lead through milk to the pups as well as reduced litter body weights.

Klauder et al. (1973) reported that low dietary copper enhanced lead absorption in rats fed a relatively high lead diet (5000 ppm). These observations were confirmed by Klauder and Petering (1975) at a level of 500 ppm lead in diet, these workers subsequently observing that reduced copper enhanced the hematological effects of lead (Klauder and Petering, 1977), and that both copper and iron deficiencies must be corrected to restore hemoglobin levels to normal.

10.6 INTERRELATIONSHIPS OF LEAD EXPOSURE, EXPOSURE INDICATORS AND TISSUE LEAD BURDENS

Information presented so far in this chapter sets forth the quantitative and qualitative aspects of lead toxicokinetics, including the compartmental modeling of lead distribution in vivo, and leads up to the critical issue of the various interrelationships of lead toxicokinetics to lead exposure, toxicant levels in indicators of such exposure, and exposure-target tissue burdens of lead.

Chapter 11 (Section 11.4, 5, 6) discusses the various experimental and epidemiological studies relating the relative impact of various routes of lead exposure on blood lead levels in human subjects, including the description of mathematical models for such relationships. In these sections, the basic question asked is: what is the mathematical relationship of lead in air, food, water, etc. to lead in blood? This question, then, is descriptive and does not address either the biological basis of the observed relationships or implications for adverse health risk in the sequence: external lead → lead in some physiological indicator → lead in target tissues.

For purposes of discussion, this section separately considers a) the temporal characteristics of physiological indicators of lead exposure, b) the biological aspects of the relationship of external exposure to internal indicators of exposure, and c) internal indicator-tissue lead relationships, including both steady-state lead exposure and abrupt changes in lead exposure.

The relationship of internal indicators of body lead, such as blood lead, to biological indicators such as EP or urinary ALA is discussed in Chapter 13, since any comparative assessment of the latter should follow the chapter on biological effects, Chapter 12.

10.6.1 Temporal Characteristics of Internal Indicators of Lead Exposure

The biological half-time for blood lead or the non-retained fraction of body lead is relatively short (see Sections 10.3 and 10.4) so that a given blood or urine lead value will reflect rather recent exposure. In cases where lead exposure can be reliably assumed to have been at a given level, a blood lead value is more useful than in cases where some intermittent, high level of exposure may have occurred. The former most often occurs with occupational exposure, while the latter is of concern with young children.

PRELIMINARY DRAFT

Accessible mineralizing tissue, such as shed teeth, extend the time frame for assessing lead exposure from weeks and several months to years (Section 10.3), since dentition continues to accumulate lead up to the time of shedding or extraction. Levels of lead in teeth increase with age in proportion to exposure (Steenhout and Pourtois, 1981). Furthermore, tooth levels are proportional to blood lead levels in humans (Winnecke, 1981) and animals (Kaplan et al., 1980). The technique of Fremlin and Edmonds (1980), employing micro-autoradiography of irradiated teeth permits the identification of dentine zones relatively high in lead content, thus allowing the disclosure of periods of past, abrupt increases in lead intake.

While levels of lead in shed dentition are more valuable than blood lead in assessing exposure at more remote time points, such information is retrospective in nature and would not be of use for monitoring current exposure. In this case, serial blood lead measurements must be employed. With the development of methodology for in-situ measurement of tooth lead in children, as described in Chapter 9, serial in-situ tooth analysis in this fashion in tandem with serial blood lead determinations would provide comparative data for determination of both time-concordant blood/tooth relationships as well as which measure is the better indicator of ongoing exposure. Given the limitations of an indicator such as blood lead in reflecting lead uptake in target organs, as discussed below, it may well be the case that the rate of accumulation of lead in teeth, measured in-situ, is a better index of ongoing tissue lead uptake than blood lead. This aspect merits much further study.

10.6.2 Biological Aspects of External Exposure-Internal Indicator Relationships

Information provided in Chapter 11 as well as the critique of Hammond et al. (1981) indicate that the relationship of levels of lead in air, food, and water to lead in blood is curvilinear, with the result that as "baseline" blood lead rises, i.e., as one moves up the curve, the relative change in the dependent variable, blood lead, per unit change of lead in some intake medium (such as air) becomes smaller. Conversely, as one proceeds down the curve with reduction in "baseline" lead, the corresponding change in blood lead becomes larger. One assumption in this "single medium" approach is that the baseline is not integrally related to the level of lead in the particular

PRELIMINARY DRAFT

medium being studied. This assumption is not necessarily appropriate in the case of air vs. food lead, nor, in the case of young children, air lead vs. total oral intake of the element.

Hammond et al. (1981) have noted that the shape of the blood lead curves seen in human subjects is similar to that discernible in certain experimental animal studies with dogs, rats, and rabbits (Azar et al., 1973; Prpic-Majic et al., 1973). More recently, Kimmel et al. (1980) reported values of blood lead in adult female rats exposed to lead at 4 levels in drinking water for 6-7 weeks. It may be seen, in comparing blood lead values with dosing levels, that the dose-blood lead curve is curvilinear. Over the dosing range 5-250 ppm in water, the blood lead range was 8.5 to 31 $\mu\text{g/dl}$. Similarly, rats exposed to lead in utero, through weaning, and up to 9 months of age at the dosing range used in the Kimmel et al. study (Grant et al., 1980)--whereby dams received 0.5 to 250 ppm through weaning of pups; weanlings then given the same levels of lead in drinking water--showed a blood lead range of 5-67 $\mu\text{g/dl}$. In all of the above studies, lead in the various dosing groups is assumed to be near or at equilibrium within the various body compartments.

The biological basis of the curvilinear relationship of blood lead to lead intake does not appear to be due to changes in absorption or excretion rates of the element with changes in exposure level. In other words, decrease in the blood lead-medium lead "ratio" with increase of blood lead cannot be taken to mean reduced absorption or enhanced excretion of lead, i.e., reduced relative uptake rate of lead into target tissues. In the study of Prpic-Majic et al. (1973) referred to above, dosing was by injection so that GI absorption rate of lead was not a factor. In the report of Azar et al. (1973), the reported values for urinary lead across the dosing groups indicated that excretion rate for the 10, 50, 100, and 500 ppm dietary lead groups was fairly constant. As suggested by Hammond et al. (1981), the shape of the blood lead curves in the context of external exposure is likely related to the tissue distribution of lead. Other evidence supporting this is the relationship of blood lead to chelatable lead and that of tissue burden to dosing level as discussed below.

10.6.3 Internal Indicator-Tissue Lead Relationships

In living human subjects, it is not possible to directly determine tissue burdens of lead (or relate these levels to adverse effects associated with

PRELIMINARY DRAFT

target tissue) as a function of lead intake. Instead, measurement of lead in an accessible indicator such as blood, along with determination of some biological indicator of impairment, e.g., ALA-U or EP, is used.

Evidence continues to accumulate in both the clinical and experimental animal literature that blood lead has limitations in reflecting both the amounts of lead in target tissues and the temporal changes in tissue lead with changes in exposure.

Perhaps the best example of the problem is the data concerning the relationship of blood lead to chelatable lead (see Section 10.3.3). Presently, measurement of the plumburesis associated with challenge by a single dose of a chelating agent such as CaNa_2EDTA is considered the best measure of the mobile, potentially toxic fraction of body lead in both children and adults (Chisolm et al., 1976; CDC, 1978; Chisolm and Barltrop, 1979; Hansen et al., 1981).

Chisolm et al. (1976) have documented that the relationship of blood lead to chelatable lead is curvilinear, such that a given incremental increase in blood lead is associated with an increasingly larger increment of mobilizable lead.

The problems associated with this curvilinear relationship in exposure assessment are typified by the recent reports of Saenger and coworkers (1982) concerning children and Hansen et al. (1981) reporting on adult lead workers.

In the former study, it was noted that significant percentages of children having mild to moderate exposure as discernible by blood lead and EP measurement were found to have urinary outputs of lead upon challenge with CaNa_2EDTA that qualified them for therapeutic chelation therapy under CDC guidelines. In adult workers, Hansen et al. (1981) observed that a sizeable fraction of subjects with only modest elevations in blood lead excreted lead upon CaNa_2EDTA challenge which significantly exceeded the upper end of normal. It was noted that this occurred at a blood lead of 35 $\mu\text{g}/\text{dl}$ and above.

The biological basis for the non-linearity of the relationship between blood lead and chelatable lead, in major part, appears to be the existence of a sizeable pool of lead in bone which is labile to chelation. Evidence pointing to this was summarized in Section 10.3.3.

PRELIMINARY DRAFT

The question of how long any lead in this compartment of bone remains labile to chelation has been addressed by several investigators in studies of both children and adults. The question is relevant to the issue of the utility of EDTA challenge in assessing evidence for past lead exposure.

Chisolm and coworkers (1976) found that a group of adolescent subjects (N=55; 12-22 y. old) who had a clinical history of lead poisoning as young children and whose mean blood lead was 22.1 µg/dl at the time of study yielded chelatable lead values which placed them on the same regression curve as a second group of young children with current elevations of blood lead. The results with the adolescent subjects did not provide evidence that they might have had a past history of lead poisoning. According to the authors, this suggests that chelatable lead at the time of excessive exposure was not retained in a pool that remained labile to chelation years later, but underwent subsequent excretion or transfer to the inert compartment of bone. One problem with drawing conclusions from this study is that all of the adolescents apparently had one or more courses of chelation therapy and were removed to housing where re-exposure would be minimal as part of their clinical management at the time that lead poisoning was diagnosed. One must assume that chelation therapy then removed a significant portion of the mobile lead burden while placement in lead-free housing would have reduced the extent of any further exposure. The question next raised is how the group of adolescents studied would compare to subjects who had excessive chronic lead exposure as young children but who did not require or receive chelation therapy.

Former lead workers challenged with CaNa_2EDTA show chelatable lead values which are significantly above normal years after workplace exposure ceases (e.g., Alessio et al., 1976; Prêrovská and Teisinger, 1970). In the case of former lead workers, blood lead also remains elevated, suggesting that the mobile lead pool in bone remains in equilibrium with blood.

The closer correspondence of chelatable lead with actual tissue lead burdens, compared to blood lead, is also reflected in a better correlation of this parameter with such biological indicators of impairment as EP. Saenger et al. (1982), in the study noted above, found that the only significant correlation with erythrocyte protoporphyrin was obtained with the µmPb/mmol EDTA ratio. In adults, similarly, Alessio et al. (1976) found that EP in

PRELIMINARY DRAFT

former lead workers was more significantly correlated with chelatable lead than with blood lead.

Consideration of both the intake vs. blood lead and the blood lead vs. chelatable lead curves would lead to the prediction that the level of lead exposure, per se, is more closely related to tissue lead burden than is blood lead, and this appears to be the case in experimental animals. In the reports of Azar et al. (1973) and Grant et al. (1980), it may be seen that levels of lead in brain, kidney, and femur follow more of a direct proportionality with the level of dosing than with blood lead.

Finally, there is the question of how adequately an internal indicator such as blood lead reflects changes in tissue burden with abrupt changes in exposure.

In the study of Björklund and coworkers (1981), lead levels in both blood and brain were monitored over a 6-week period in rats exposed to lead in drinking water. Blood lead rose rapidly by day 1, during which time brain lead content was only slightly elevated. After day 1, the rate of increase in blood lead began to taper off while brain lead began to rise in a near-linear fashion up to the end of the experiment. From day 7 to 21, blood lead increased from ca. 45 to 55 $\mu\text{g}/\text{dl}$, while brain lead increased ca. 2-fold.

Abrupt reduction in exposure, similarly, appears to be associated with a more rapid response in blood than in soft tissues, particularly brain. In the report of Goldstein and Diamond (1974), termination of intravenous administration of lead to 30-day-old rats resulted in a 7-fold drop of lead in blood by day 7. At the same time, there was no significant decrease in brain lead. A similar difference in brain vs. blood response was reported by Momcilovic and Kostial (1974).

In all of the above studies, it may be seen that blood lead was of limited value in reflecting changes in what is, for children, the significant target organ for lead exposure, the brain. With an abrupt increase in exposure, the problem relates to a much more rapid approach to steady-state in blood than in brain. Conversely, the biological half-time for lead clearance from blood in the young rats of both the Goldstein and Diamond and Momcilovic and Kostial studies was much less than it appeared to be for lead movement from brain.

10.7 METABOLISM OF LEAD ALKYL

The lower alkyl lead compounds used as gasoline additives, tetraethyl (TEL) and tetramethyl (TML) lead, are much more toxic, i.e., neurotoxic, on an equivalent dose basis, than inorganic lead. These agents are emitted in auto exhaust and their rate of environmental degradation is dependent on such factors as sunlight, temperature, and ozone levels. There is also some concern that organolead compounds may result from biomethylation in the environment (see Chapter 6). Finally, there appears to be a problem with the practice among children of sniffing leaded gasoline.

The available information dealing with metabolism of lead alkyls is mainly derived from experimental animal studies, workers exposed to the agents, and cases of lead alkyl poisoning.

10.7.1 Absorption of Lead Alkyls in Humans and Animals

The respiratory intake and absorption of TEL and TML in the vapor state was investigated by Heard and coworkers (1979) who used human volunteers inhaling ^{203}Pb -labeled TEL and TML. Initial lung deposition rates were 37 and 51 percent for TEL and TML, respectively. Of these amounts, 40 percent of TEL was lost by exhalation by 48 h., while the corresponding figure for TML was 20 percent. The remaining fraction was absorbed. In this study, the effect of gasoline vapor on these parameters was not investigated.

Mortensen (1942) reported that adult rats inhaling TEL labeled with ^{203}Pb (0.07-7.00 mg TEL/l) absorbed 16-23 percent of the fraction reaching the alveoli. It was found that gasoline vapor had no effect on the absorption rates.

Respiratory absorption of organolead bound to particulate matter has not been specifically studied as such. According to Harrison and Laxen (1978), TEL or TML is not adhered to particulate matter to any significant extent, but the toxicologically equivalent trialkyl derivatives, formed from photolytic dissociation or ozonolysis in the atmosphere, may well be.

Gastrointestinal Absorption

Information on the rate of absorption of lead alkyls through the gastrointestinal tract has not been forthcoming in the literature. Given the level of gastric acidity, pH 1.0, in humans, one would expect TML and TEL to be rapidly converted to the corresponding trialkyl forms, which are comparatively more stable (Bade and Huber, 1970). Given the similarity of

PRELIMINARY DRAFT

chemical and biochemical behavior of trialkyl leads to their Group IV analogs, the trialkyltins, the report of Barries and Stoner (1958) that triethyltin is quantitatively absorbed from the GI tract indicates that triethyl- and trimethyllead would be extensively absorbed via this route.

Percutaneous Absorption of Lead Alkyls

In contrast to inorganic lead salts, both TEL and TML are rapidly and extensively absorbed through the skin in rabbits and rats (Kehoe and Thamann, 1931; Laug and Kunze, 1948), and lethal effects can be rapidly induced in these animals by just skin exposure. Laug and Kunze (1948) observed that systemic uptake of TEL was still 6.5 percent even though most of the TEL was seen to have evaporated from the skin surface. The rate of passage of TML was somewhat slower than that of TEL in the study of Davis et al. (1963), while uptake of either agent was retarded somewhat when applied in gasoline.

10.7.2 Biotransformation and Tissue Distribution of Lead Alkyls

In order to have an understanding of the in vivo fate of lead alkyls, it is useful to first discuss the biotransformation processes of lead alkyls known to occur in mammalian systems.

Tetraethyl and tetramethyl lead both undergo oxidative dealkylation in mammals to the triethyl or trimethyl metabolites, which are now accepted as being the actual toxic forms of these alkyls.

Studies of the biochemical mechanisms for these transformations, as noted by Kimmel et al. (1977) indicate a dealkylation mediated by a P-450 dependent monooxygenase system in liver microsomes, with intermediate hydroxylation. In addition to the rat (Cremer, 1959; Stevens et al., 1960; Bolanowska, 1968), mice (Hayakawa, 1972) and rabbits (Bolanowska and Garczynski, 1968) this transformation also occurs in humans accidentally poisoned with TEL (Bolanowska et al., 1967) or workers cronically exposed to TEL (Adamiak-Ziemba and Bolanowska, 1970).

The rate of hepatic oxidative deethylation of TEL in mammals appears to be rather rapid, Cremer (1959) reporting a maximum conversion rate of ca. 200 µg TEL/g rat liver/h. In comparison with TEL, TML may undergo transformation at either a slower rate (in rats) or more rapidly (in mice), according to Cremer and Calloway (1961) and Hayakawa (1972).

Other transformation steps involve conversion of triethyl lead to diethyl form, the process appearing to be species-dependent. Bolanowska (1968) did

PRELIMINARY DRAFT

not report the formation of diethyl lead in rats, while significant amounts of this form are present in the urine of rabbits (Arai et al., 1981) and humans (Chiesura, 1970). Inorganic lead is formed in various species treated with tetraethyl lead, which may arise from degradation of the diethyl lead metabolite or some direct process (Bolanowska, 1968), the latter suggested as occurring in rats, since little or no diethyllead is found, but significant amounts of inorganic lead.

Formation of inorganic lead with lead alkyl exposure may account for the hematological effects seen in subjects chronically exposed to the lead alkyls (see Hematological Effects section), including children who inhale leaded gasoline vapor.

Distribution of Lead Alkyls

Partitioning of triethyl- or trimethyl lead, the corresponding active metabolites of TEL and TML, between the erythrocyte and plasma appears to be species-dependent. Byington and coworkers (1980) studied the partitioning of triethyl lead between cells and plasma in vitro using washed human and rat erythrocytes and found that human cells had a very low affinity for the alkyl lead while rat cells bound the alkyl lead in the globin moiety at a ratio of 3 molecules per Hb tetramer. Similarly, it was found that injected triethyl lead was associated with whole blood lead values ca. 10-fold that of plasma in the rat. The available literature on TEL poisoning in man is consistent with this, significant plasma values of lead having been routinely reported (Boeckx et al., 1977; Golding and Stewart, 1982). These data indicate that the rat is a poor model for study of the adverse effects of lead alkyls in human subjects.

The biological half-time in blood for the lead alkyls depends on whether clearance of the tetraalkyl or trialkyl forms is being observed. Heard and coworkers (1979) found the ²⁰³Pb-labeled TML and TEL inhaled by human volunteers to be rapidly cleared from blood, by 10 h., followed by a reappearance of lead. The fraction of lead in plasma initially was quite high, ca. 0.7, suggesting tetra/trialkyl lead, while the subsequent rise in blood lead showed all of it essentially in the cell, which would indicate inorganic or possibly diethyl lead. Triethyl lead in rabbits was more rapidly cleared from blood of rabbits (3-5 d.) than was the trimethyl form (15 d.) when administered as such (Hyakawa, 1972).

PRELIMINARY DRAFT

Tissue distribution of lead in both humans and animals exposed to TEL and TML primarily involves the trialkyl metabolites, levels being highest in liver, followed by kidney, then brain (Bolanowska et al., 1967; Grandjean and Nielsen, 1979). Of interest in this regard is the observation of Nielsen et al. (1978) that measureable amounts of trialkyl lead were measured in samples of brain tissue from subjects with no known occupational exposure.

The available information on tissue retention of triethyl- or trimethyl lead provides variable data. Bolanowska (1968) noted that tissue levels of triethyl lead in rats was almost constant for 16 days after a single injection of TEL. Hayakawa (1972) found that the half-time of triethyl lead in brain was 7-8 d. for rats, the half-time for trimethyl lead being much longer. In humans, there was reported two tissue compartments for triethyl lead, having half-times of 35 and 100 d. (Yamamura et al., 1975).

10.7.3 Excretion of Lead Alkyls

Excretion of lead through the renal tract is the main route of elimination in various species exposed to lead alkyls (Grandjean and Nielsen, 1979). The chemical forms of lead in the urine suggest that the relative amounts of the various forms is species-dependent. Arai et al. (1981) found that rabbits given TEL parenterally excreted lead mainly in the form of diethyl lead (69 percent) and inorganic lead (27 percent), triethyl lead accounting only for 4 percent. In rats, Bolanowska and Garczynski (1968) found that the relative amount of triethyl lead was somewhat higher in urine than was the case for rabbits. In humans, Chiesura (1970) found that trialkyl lead never was greater than 9 percent of total lead content in workers having had heavy TEL exposure. Adamiak-Ziemba and Bolanowska (1970) reported similar data, the fraction of triethyl lead in the urine was ca. 10 percent of total lead.

The urinary rates of lead excretion in human subjects with known level of TEL exposure were reported by Adamiak-Ziemba and Bolanowska (1970). In workers involved with blending and testing of leaded gasoline, workplace air levels of TEL (as Pb) ranged from 0.037-0.289 mg Pb/m³ and the corresponding urine levels ranged from 14-49 µg Pb/l, of which ca. 10 percent was triethyl lead.

PRELIMINARY DRAFT

REFERENCES

- Adamiak-Ziemka, J.; Bolanowska, W. (1970) Occupational exposure to tetraethyl lead as estimated by tetraethyl lead concentration in air and triethyl lead concentration in urine. *Med. Proc.* 21: 172-179.
- Alessio, L.; Bartazzi, A.; Morelli, O.; Toffoletto, F. (1976) Free erythrocyte protoporphyrin as an indicator of the biological effect of lead in adult males. III. Behavior of free erythrocyte protoporphyrin in workers with past lead exposure. *Int. Arch. Occup. Environ. Health* 38: 77-86.
- Alexander, F. W.; Delves, H. T. (1981) Blood-lead levels during pregnancy. *Int. Arch. Occup. Environ. Health* 48: 35-39.
- Alexander, F. W.; Delves, H. T.; Clayton, B. E. (1973) The uptake and excretion by children of lead and other contaminants. In: *Environmental Health Aspects of Lead*. Luxembourg, Commission of the European Communities. Pp. 319-330.
- Allcroft, R. (1950) Lead as a nutritional hazard to farm livestock. Iv. Distribution of lead in the tissues of bovines after ingestion of various lead compounds. *J. Comp. Path. Ther.* 60: 190-208.
- Anders, E.; Bagnell, C. R. Jr.; Krigman, M. R.; Mushak, P. (1982) Influence of dietary protein composition on lead absorption in rats. *Bull. Environ. Contam. Toxicol.* 28: 61-67.
- Air Quality Criteria for Lead. (1977) U.S. Environmental Protection Agency, Office of Research and Development, Washington, D.C. Special Series: EPA-600/8-77-017, December.
- Arai, F.; Yamamura, Y.; Yoshida, M. (1981) Excretion of triethyl lead, diethyl lead and inorganic lead after injection of tetraethyl lead in rabbits. *Sangyo Igaku* 23: 496-504.
- Araki, S. (1973) On the behavior of "active deposit of lead (Teisinger)" in the Japanese free from occupational exposure to lead. *Ind. Health (Kawasaki)* 11: 203-224.
- Araki, S.; Ushio, K. (1982) Assessment of the body burden of chelatable lead: a model and its application to lead workers. *Brit. J. Ind. Med.* 39: 157-160.
- Aungst, B. J.; Dolce, J. A.; Fung, H. L. (1981) The effect of dose on the disposition of lead in rats after intravenous and oral administration. *Toxicol. Appl. Pharmacol.* 61: 48-57.
- Awad, L.; Huel, G.; Lazar, P.; Boudene, C. (1981) Facteurs de variation inter-individuelle de la plombémie. *Rev. Epidemiol. Sante Publ.* 29: 113-124.

PRELIMINARY DRAFT

- Azar, A.; Trockimowicz, H. J.; Maxfield, M. E. (1973) Review of lead studies in animals carried out at Haskell Laboratory--two-year feeding study and response to hemorrhage study. In: Environmental Health Aspects of Lead. Proceedings of an International Symposium, Amsterdam, 1972. C.E.E., Luxembourg, pp. 199-210.
- Bade, V.; Huber, F. (1970) Reactionen von Organometallverbindungen. IV. Über den Einfluss der organischen Liganden und das Zentral atoms auf die Geschwindigkeit der Acidolyse von Tetraorganoplumbanen und Tetraorganostannanen. J. Organometal. Chem. 24: 387-397.
- Baloh, R. W. (1974) Laboratory diagnosis of increased lead absorption. Arch. Environ. Health 28: 198-206.
- Barltrop, D. (1969) Transfer of lead to the human foetus. In: Mineral Metabolism in Pediatrics, D. Barltrop and W. L. Burland, eds., Davis Co., Philadelphia. Pp. 135-151.
- Barltrop, D. (1972) Children and environmental lead. In: Lead in the Environment, P. Hepple, ed., Institute of Petroleum.
- Barltrop, D. (1975) Assessment of the Health Hazard of Various Lead Compounds. Center for Disease Control, U.S. Dept. of Health, Education and Welfare. Atlanta, Ga.
- Barltrop, D. (1982) Nutritional and maturational factors modifying the absorption of inorganic lead from the gastrointestinal tract. In: Health Evaluation of Heavy Metals in Infant Formula and Junior Food (Hildebrandt and E.H.F. Schmidt, eds.) Springer-Verlag, New York, in press.
- Barltrop, D.; Khoo H. E. (1975) The influence of nutritional factors on lead absorption. Postgrad. Med. J. 51: 795-800.
- Barltrop, D.; Meek, F. (1975) Absorption of different lead compounds. Postgrad. Med. J. 51: 805-809.
- Barltrop, D.; Strehlow, C. D. (1977) The absorption of lead by children. In: Trace Element Metabolism in Man and Animals-3, M. Kirchgessner, ed., Friesing, W. German. Pp. 332-334.
- Barltrop, D.; Meek, F. (1979) Effect of particle size on lead absorption from the gut. Arch. Environ. Health 34: 280-285.
- Barltrop, D.; Barrett, A. J.; Dingle, J. T. (1974) Subcellular distribution of lead in the rat. J. Lab. Clin. Med. 77: 705-712.
- Barnes, J. M.; Stoner, H. B. (1958) Toxic properties of some dialkyl and trialkyl tin salts. Brit. J. Ind. Med. 15: 15-22.
- Barry, P. S. I. (1975) A comparison of concentrations of lead in human tissues. Brit. J. Ind. Med. 32: 119-139.

PRELIMINARY DRAFT

- Barry, P. S. I. (1978) Distribution and storage of lead in human tissues. In: The Biogeochemistry of Lead in the Environment. (J. O. Nriagu, ed.) Elsevier, Amsterdam, pp. 97-150.
- Barry, P. S. I. (1981) Concentrations of lead in the tissues of children. Brit. J. Ind. Med. 38: 61-71.
- Barry, P. S. I.; Mossman, D. B. (1970) Lead concentrations in human tissues. Brit. J. Ind. Med. 27: 339-351.
- Barton, J. C.; Conrad, M. E. (1981) Effect of phosphate on the absorption and retention of lead in the rat. Amer. J. Clin. Nutr. 34: 2192-2198.
- Barton, J. C.; Conrad, M. E.; Harrison, L.; Nuby, S. (1978) Effects of calcium on the absorption and retention of lead. J. Lab. Clin. Med. 91: 366-376.
- Barton, J. C.; Conrad, M. E.; Nuby, S.; Harrison, L. (1978) Effects of iron on the absorption and retention of lead. J. Lab. Clin. Med. 92: 536-547.
- Barton, J. C.; Conrad, M. E.; Harrison, L.; Nuby, S. (1980) Effects of vitamin D on the absorption and retention of lead. Amer. J. Physiol. 238: G124-130.
- Batschelet, E.; Brand, L.; Steiner, J. (1979) On the kinetics of lead in the human body. J. Math. Biol. 8: 15-23.
- Beattie, A. D.; Moore, M. R.; Goldberg, A. (1972) Tetraethyl lead poisoning. Lancet (2): 12-15.
- Bell, R. R.; Spickett, J. T. (1981) The influence of milk in the diet on the toxicity of orally ingested lead in rats. Rd. Cosmet. Toxicol. 19: 429-436.
- Berg, L. R.; Nordstrom J. O.; Ousterhout (1980) The prevention of chick growth depression due to dietary lead by increased dietary calcium and phosphorus levels. Poult. Sci. 59: 1860-1863.
- Bianco, A.; Gibb, F. R.; Morrow, P. E. (1974) Inhalation study of a submicron size lead aerosol. In: Proceedings of the Third International Congress of the International Radiation Protection Association. Pp. 1214-1219, Report CONF. No. 730907-P-2.
- Björklund, H.; Lind, B.; Piscator, M.; Hoffer, B.; Olson, L. (1981) Lead, zinc and copper levels in intraocular brain tissue grafts, brain and blood of lead-exposed rats. Toxicol. Appl. Pharmacol. 60: 424-430.
- Blake, K.C.H. (1976) Absorption of ²⁰³Pb from the gastrointestinal tract of man. Environ. Res. 11:1-4.
- Boeckx, R. L.; Postl, B.; Codin, F. J. (1977) Gasoline sniffing and tetraethyl lead poisoning in children. Pediatr. 60: 140-145.

PRELIMINARY DRAFT

- Bolanowska, W. (1968) Distribution and excretion of triethyl lead in rats. Brit. J. Ind. Med. 25: 203-208.
- Bolanowska, W.; Garczynski, H. (1968) Metabolism of tetraethyl lead in rabbits. Med. Prac. 19: 235-243, 1968.
- Bolanowska, W.; Piotrowski, J.; Carzynski, H. (1967) Triethyl lead in the biological material in cases of acute tetraethyl lead poisoning. Arch. Toxicol. 278-282.
- Booker, D. V.; Chamberlain, A. C.; Newton, D.; Stott, A. N. B. (1969) Uptake of radioactive lead following inhalation and injection. Brit. J. Radiol. 42: 457-466.
- Boudene, C.; Malet, D.; Masse, R. (1977) Fate of ^{210}Pb inhaled by rats. Toxicol. Appl. Pharmacol. 41: 271-276.
- Bruenger, F. W.; Stevens, W.; Stower, B. J. (1973) The association of ^{210}Pb with constituents of erythrocytes. Health Phys. 25: 37-42.
- Buchet, J. P.; Roels, H.; Hubeimont, G.; Lauwerys, R. (1978) Placental transfer of lead, mercury, cadmium and carbon monoxide in women. II. Environ. Res. 15: 494-503.
- Bushnell, P. J.; DeLuca, H. F. (1981) Lactose facilitates the intestinal absorption of lead in weanling rats. Science 211: 61-63.
- Butt, E. M.; Musdaum, R. E.; Gilmore, T. C.; Dideo, S. L. (1974) Trace metal levels in human serum and blood. Arch. Environ. Health 8: 52-57.
- Byington, K. H.; Yates, D. A.; Mullins, W. A. (1980) Binding of triethyl lead chloride by hemoglobin. Toxicol. Appl. Pharmacol. 52: 379-385.
- Castellino, N.; Aloj, S. (1964) Kinetics of the distribution and excretion of lead in the rat. Brit. J. Ind. Med. 21: 308-314.
- Castellino, N.; Lamanna, P.; Grieco, B. (1966) Biliary excretion of lead in the rat. Brit. J. Ind. Med. 23: 237-239.
- Cavalleri, A.; Minoia, C.; Pazzoli, L.; Baruffini, A. (1978) Determination of plasma lead levels in normal subjects and in lead-exposed workers. Brit. J. Ind. Med. 35: 21-26.
- Centers for Disease Control. (1978) Statement: Preventing Lead Poisoning in Young Children. United States Public Health Service, Atlanta, Ga.
- Cerklewski, F. L.; Forbes, R. M. (1976) Influence of dietary zinc on lead toxicity in the rat. J. Nutr. 106: 689-696.
- Cerklewski, F. L. (1979) Influence of dietary zinc on lead toxicity during gestation and lactation in the female rat. J. Nutr. 109: 1703-1709.

PRELIMINARY DRAFT

- Chamberlain, A. C.; Heard, M. J.; Little, P.; Newton, D.; Wells, A. C.; Wiffin, R. D. (1978) Investigations into lead from motor vehicles. AERE, Harwell, England. Report No. AERE-R9198, November.
- Chamberlain, A. C.; Heard, M. J. (1981) Lead tracer and lead balances. In: Environmental Lead, D. R. Lynum, L. G. Piantanida and J. F. Cole, eds., Academic Press, New York, Pp. 175-198.
- Chesney, R. W.; Rosen, J. F.; Hamstra, A. J.; Smith, C.; Mahaffey, K. R.; DeLuca, H. F. (1981) Absence of season variation in serum concentrations of 1,25-dihydroxy-vitamin D despite a rise in 25-hydroxy-vitamin D levels in summer. J. Clin. Endocrinol. Metab. 53: 139-142.
- Chiesura, P. (1970) Urinary excretion of tetraethyl lead catabolites in man. Med. Lav. 61: 437-441.
- Chisolm, J. J. Jr. (1981) Dose-effect relationships for lead in young children: evidence in children for interactions among lead, zinc and iron. In: Environmental Lead, D. R. Lynum, L. G. Piantanida and J. R. Cole, eds., Academic Press, New York. Pp. 1-7.
- Chisolm, J. J., Jr.; Harrison, H. E. (1956) The exposure of children to lead. Pediatr. 18: 943-957.
- Chisolm, J. J. Jr.; Barltrop, D. (1979) Recognition and management of children with increased lead absorption. Arch. Dis. Childhood 54: 249-262.
- Chisolm, J. J. Jr.; Mellits, E. D.; Barrett, M. B. (1976) Inter-relationships among blood lead concentration, quantitative daily ALAU and urinary lead output following calcium EDTA. In: Effects and Dose-Response Relationships of Toxic Metals, G. F. Nordberg, ed., Elsevier, Amsterdam. Pp. 416-433.
- Clarkson, T. W.; Kench, J. R. (1958) Uptake of lead by human erythrocytes in vitro. Biochem. J. 69: 432-439.
- Cohen, N. (1970) The retention and distribution of lead-210 in the adult baboon. Ph.D. Thesis, New York University.
- Conrad, M. E.; Barton, J. C. (1978) Factors affecting the absorption and excretion of lead in the rat. Gastroenterology 74: 731-740.
- Cramer, K.; Goyer, R. A.; Jagenburg, R. A.; Wilson, M. H. (1974) Renal ultra structure, renal function and parameters of lead toxicity in workers with different periods of lead exposure. Brit. J. Ind. Med. 31: 113-127.
- Cremer, J. E. (1959) Biochemical studies on the toxicity of tetraethyl lead and other organo-lead compounds. Brit. J. Ind. Med. 16: 191-199.
- Cremer, J. E.; Calaway, S. (1961) Further studies on the toxicity of some tetra- and trialkyl lead compounds. Brit. J. Ind. Med. 18: 277-282.

PRELIMINARY DRAFT

- Dacre, J. C.; TerHaar, G. L. (1977) Lead levels in tissues from rats fed soils containing lead. Arch. Environ. Contam. Toxicol. 6: 111-119.
- Danscher, G.; Hall, E.; Fredens, K.; Fjerdingstad, E.; Fjerdingstad, E. J. (1975) Heavy metals in the amygdala of the rat: zinc, lead and copper. Brain Res. 94: 167-172.
- Davis, R. K.; Horton, A. W.; Larson, E. E.; Stenmer, K. L. (1963) Inhalation of tetramethyl lead and tetraethyl lead. Arch. Environ. Health 6: 473-479.
- Day, J. D.; Ferguson, J. E.; Chee, T. M. (1979) Solubility and potential toxicity of lead in urban street dusts. Bull. Environ. Contam. Toxicol. 23: 497-502.
- Day, J. P.; Hart, M.; Robinson, M. S. (1975) Lead in urban street dusts. Nature 253: 343-245.
- Delves, H. T.; Clayton, B. E.; Carmichael, A.; Bauer, M.; Smith, M. (1982) An appraisal of the analytical significance of tooth-lead measurements as possible indices of environmental exposure of children to lead. Ann. Clin. Biochem., in press.
- Department of the Environment. (1982) The Glasgow Duplicate Diet Study (1979/1980). Central Directorate on Environmental Pollution, Pollution Report No. 11, HMSO.
- Der, R.; Fahim, Z.; Hilderbrand, D.; Fahim, M. (1974) Combined effect of lead and low protein diet on growth, sexual development and metabolism in female rats. Res. Commun. Chem. Pathol. Pharmacol. 9: 723-738.
- DeSilva, P. E. (1981) Determination of lead in plasma and studies on its relationship to lead in erythrocytes. J. Ind. Med. 38: 209-217.
- Dobbins, A.; Johnson, D. R.; Nathan, P. (1978) Effect of exposure to lead on maturation of intestinal iron absorption of rats. J. Toxicol. Environ. Health 4: 541-550.
- Duggan, M. J.; Williams, S. (1977) Lead-in-dust in city streets. Sci. Total Environ. 7: 91-97.
- El-Gazzar, R. M.; Finneli, V. N.; Boiano, J.; Petering, H. G. (1978) Influence of dietary zinc on lead toxicity in rats. Toxicol. Lett. 1: 227-234.
- Everson, J.; Patterson, C. C. (1980) "Ultra-clean" isotope dilution/mass spectrometric analyses for lead in human blood plasma indicate that most reported values are artificially high. Clin. Chem. 26: 1603-1607.
- Fjerdingstad, E. J.; Danscher, E.; Fjerdingstad, E. (1974) Hippocampus: selective concentration of lead in the normal rat brain. Brain Res. 80: 350-354.
- Forbes, G. B.; Reina, J. C. (1972) Effect of age on gastrointestinal absorption (Fe, Sr, Pb) in the rat. J. Nutr. 102: 647-652.

PRELIMINARY DRAFT

- Fremlin, J. M.; Edmonds, M. I. (1980) The determination of lead in human teeth. *Nucl. Inst. Meth.* 173: 211-215.
- Gage, J. C.; Litchfield, M. H. (1968) The migration of lead from polymers in the gastrointestinal tract. *Fd. Cosmet. Toxicol.* 6: 329-338.
- Gage, J. C.; Litchfield, M. H. (1969) The migration of lead from paint films in the rat gastrointestinal tract. *J. Oil. Col. Chem. Assoc.* 52: 236-243.
- Gerber, B. T.; Wei, E. (1974) Influence of dietary factors on the gastrointestinal absorption of lead. *Toxicol. Appl. Pharmacol.* 27: 685-691.
- Gershanik, J. J.; Brooks, G. G.; Little, J. A. (1974) Blood lead values in pregnant women and their offspring. *Amer. J. Obstet. Gynecol.* 119: 508-511.
- Golding, A. S.; Stewart, R. M. (1982) Organic lead encephalopathy: behavioral change and movement disorder following gasoline inhalation. *J. Clin. Psychiatry* 43: 70-72.
- Goldstein, G. W.; Diamond, I. (1974) Metabolic basis of lead encephalopathy. Brain dysfunction in metabolic disorders. *Res. Publ. Assoc. Nerv. Ment. Dis.* 53: 293-304.
- Goldstein, G. W.; Asburg, A. K.; Diamond, I. (1974) Pathogenesis of lead encephalopathy, uptake of lead and reaction of brain capillaries. *Arch. Neurol.* 31: 382-389.
- Goyer, R. A. (1978) Calcium and lead interactions: Some new insights. *J. Lab. Clin. Med.* 91: 363-366.
- Goyer, R. A.; May, P.; Cates, M. M.; Krigman, M. R. (1970) Lead and protein content of isolated intranuclear inclusion bodies from kidneys of lead-poisoned rats. *Lab. Invest.* 22: 245-251.
- Grandjean, P. (1978) Regional distribution of lead in human brains. *Toxicol. Lett.* 2: 65-69.
- Grandjean, P.; Nielsen, T. (1979) Organolead compounds: Environmental health aspects. *Residue Rev.* 72: 97-148.
- Grant, R. L.; Calvery, H. O.; Lang, E. P.; Morris, H. J. (1938) The influence of calcium and phosphorus on the storage and toxicity of lead and arsenic. *J. Pharmacol. Exp. Ther.* 64: 446-457.
- Grant, L. D.; Kimmel, C. A.; West, G. L.; Martinez-Vargas, C. M.; Howard, J. L. (1980) Chronic low-level lead toxicity in the rat. I. Effects on postnatal physical and behavioral development. *Toxicol. Appl. Pharmacol.* 56: 42-58.
- Greenhalgh, J. R.; James, A. C.; Smith, H.; Hodgson, A. (1979) Absorption of lead ions from the lung. In: National Radiological Protection Board Annual Research and Development Report No. NRPB/R&D3. Pp. 38-42.

PRELIMINARY DRAFT

- Griffin, T. B.; Coulston, F.; Wills, H.; Russell, J. C.; Knelson, J. H. (1975) Clinical studies on men continuously exposed to airborne particulate lead. *Environ. Qual. Safety (Suppl. 2)*: 221-240.
- Gross, S. B. (1981) Human oral and inhalation exposures to lead: summary of Kehoe balance experiments. *J. Toxicol. Environ. Health* 8: 333-377.
- Gross, S. B.; Pfitzer, E. A. (1974) Influence of pathological change on lead in human tissues. *Trace Subst. Environ. Hlth.* 8: 335-340.
- Gross, S. B.; Pfitzer, E. A.; Yeager, D. W.; Kehoe, R. A. (1975) Lead in human tissues. *Toxicol. Appl. Pharmacol.* 32: 638-651.
- Gutniak, O.; Koziolowa, H.; Kowalskii. (1964) Free protoporphyrin content of erythrocytes in chronic tetraethyl lead poisoning. *Lancet* (1): 1137-1138.
- Hambidge, K. M. (1977) The role of zinc and other trace metals in pediatric nutrition and health. *Pediatr. Clin. North Am.* 24: 95-105.
- Hamilton, D. L. (1978) Interrelationships of lead and iron retention in iron-deficient mice. *Toxicol. Appl. Pharmacol.* 46: 651-661.
- Hammond, P. B. (1971) The effects of chelating agents on the tissue distribution and excretion of lead. *Toxicol. Appl. Pharmacol.* 18: 296-310.
- Hammond, P. B. (1973) The effects of D-penicillamine on the tissue distribution and excretion of lead. *Toxicol. Appl. Pharmacol.* 26: 241-246.
- Hammond, P. B.; O'Flaherty, E. J.; Gartside, P. D. (1981) The impact of air lead on blood-lead in man--a critique of the recent literature. *Rd. Cosmet. Toxicol.* 19: 631-638.
- Hansen, J. P. B.; Dossing, M.; Paulex, P. E. (1981) Chelatable lead body burden (by calcium-disodium EDTA) and blood lead concentration in man. *J. Occup. Med.* 23: 39-43.
- Harris, P.; Halley, M. R. (1972) Lead levels in cord blood. *Pediatr.* 49: 606-608.
- Harrison, R. M. (1979) Toxic metals in street and household dusts. *Sci. Total Environ.* 11: 89-97.
- Harrison, R. M.; Laxen, D. P. H. (1978) Sink processes for tetraalkyl lead compounds in the atmosphere. *Environ. Sci. Technol.* 12: 1384-1392.
- Harrison, G. E.; Carr, T. E. F.; Sutton, A.; Humphreys, E. R.; Rundo, J. (1969) Effect of alginate on the absorption of lead in man. *Nature* 224: 1115-1116.
- Hart, M. H.; Smith, J. L. (1981) Effect of vitamin D and low dietary calcium on lead uptake and retention in rats. *J. Nutr.* 111: 694-698.

PRELIMINARY DRAFT

- Hayakawa, K. (1972) Micro-determination and dynamic aspects of alkyl lead compounds in vivo. II. Studies on the dynamic aspects of alkyl lead compounds in vivo. Jap. J. Hyg. 26: 526-535.
- Heard, M. J.; Chamberlain, A. C. (1982) Effects of minerals and food on uptake of lead from the gastrointestinal tract in humans. Human Toxicol., in press.
- Heard, M. J.; Wells, A. C.; Newton, D.; Chamberlain, A. C. (1979) Human uptake and metabolism of tetraethyl- and tetramethyllead vapor labeled with lead-203. In: Manage. Control Heavy Met. Environ. Int. Conf. CEP Consultants, Ltd., Edinburg, pp. 103-108.
- Holtzman, R. B. (1978) Application of radio lead to metabolic studies. In: The Biogeochemistry of Lead in the Environment. Part B, J. O. Nriagu, ed., Elsevier, Amsterdam/New York. Pp. 37-96.
- Horiguchi, S.; Utsunomiya, T. (1973) An estimate of the body burden of lead in the healthy Japanese population. An attempt to assume absorption and excretion of lead in the healthy Japanese population. Part 2. Osaka City Med. J. 19: 1-5.
- Horiuchi, K.; Horiguchi, S.; Suekane, M. (1959) Studies on the industrial lead poisoning. Absorption, transportation, deposition and excretion of lead. 6. The lead contents in organ tissues of the normal Japanese. Osaka City Med. J. 5: 41-70.
- Hunter, J. M. (1977) The summer disease: an integrative model of the seasonality aspects of childhood lead poisoning. Soc. Sci. Med. 11: 691-698, 1977.
- Hunter, J. M. (1978) The summer disease: some field evidence on seasonality in childhood lead poisoning. Soc. Sci. Med. 12: 85-94.
- Hsu, F. S.; Kroak, L.; Pond, W. G.; Duncan, J. R. (1975) Interaction of dietary calcium with toxic levels of lead and zinc in pigs.
- Hursh, J. B.; Mercer, T. T. (1970) Measurement of ^{212}Pb loss rate from human lung. J. Appl. Physiol. 28: 268-274.
- Hursh, J. B.; Suomela, J. (1968) Absorption of Pb from the gastrointestinal tract of man. Acta Radiol. 7: 108-120.
- Indraprasit, S.; Alexander, G. V.; Gonick, H. C. (1974) Tissue composition of major and trace elements in uremia and hypertension. J. Chron. Dis. 27: 135-161.
- James, A. C. (1978) Lung deposition of submicron aerosols calculated as a function of age and breathing rate. Annual Research and Development Report, National Radiological Protection Board, Harwell, England. Pp. 71-75.

PRELIMINARY DRAFT

- Johnson, N. E.; Tenuta, K. (1979) Diet and lead blood levels of children who practice pica. *Environ. Res.* 18: 369-376.
- Jugo, S. (1980) Chelatable fraction of ^{203}Pb in blood of young and adult rats. *Environ. Res.* 21: 336-342.
- Jugo, S.; Maljkovic, T.; Kostial, K. (1975a) Influence of chelating agents on the gastrointestinal absorption of lead. *Toxicol. Appl. Pharmacol.* 34: 259-263.
- Jugo, S.; Maljkovic, T.; Kostial, K. (1975b) The effect of chelating agents on lead excretion in rats in relation to age. *Environ. Res.* 10: 271-279.
- Kaplan, M. L.; Peresie, H. J.; Jeffcoat, M. K. (1980) The lead content of blood and deciduous teeth in lead-exposed beagle pups. In: *Low Level Lead Exposure: Clin. Impl. Curr. Res.*, H. L. Needleman, ed., Raven Press, New York. Pp. 221-230.
- Kehoe, R. A. (1961a) The metabolism of lead in man in health and disease. I. The normal distribution of lead. *R. Inst. Public Hlth. Hyg. J.* 24: 81-97.
- Kehoe, R. A. (1961b) The metabolism of lead in man in health and disease. II. The metabolism of lead under abnormal conditions. *R. Inst. Public Hlth. Hyg. J.* 24: 129-143.
- Kehoe, R. A. (1961c) The metabolism of lead in man in health and disease. III. Present hygienic problems relating to the absorption of lead. *R. Inst. Public Hlth. Hyg. J.* 24: 177-203.
- Kehoe, R. A.; Thamann, F. (1931) The behavior of lead in the animal organism. II. Tetra ethyl lead. *Amer. J. Hyg.* 13: 478-493.
- Keller, C. A.; Doherty, R. A. (1980a) Distribution and excretion of lead in young and adult female mice. *Environ. Res.* 21: 217-228.
- Keller, C. A.; Doherty, R. A. (1980b) Bone lead mobilization in lactating mice and lead transfer to suckling offspring. *Toxicol. Appl. Pharmacol.* 55: 220-228.
- Keller, C. A.; Doherty, R. A. (1980c) Lead and calcium distribution in blood, plasma and milk for the lactating mouse. *J. Lab. Clin. Med.* 95: 81-99.
- Kello, D.; Kostial, K. (1973) The effect of milk diet on lead metabolism in rats. *Environ. Res.* 6: 355-360.
- Kimmel, E. C.; Fish, R. H.; Casido, J. E. (1977) Bioorganotin chemistry: Metabolism of organotin compounds in microsomal mono-oxygenase system and in mammals. *J. Agr. Food Chem.* 25: 1-9.
- Kimmel, C. A.; Grant, L. D.; Sloan, C. S.; Gloden, B. C. (1980) Chronic low-level toxicity in the rat. I. Maternal toxicity and perinatal effects. *Toxicol. Appl. Pharmacol.* 56: 28-41.

PRELIMINARY DRAFT

- Klaassen, C. D.; Shoeman, D. W. (1974) Biliary excretion of lead in rats, rabbits and dogs. *Toxicol. Appl. Pharmacol.* 29: 434-446.
- Klauder, D. S.; Murthy, L.; Petering, H. G. (1973) Effects of dietary intake of lead acetate on copper metabolism in male rats. *Trace Subst. Environ. Health* 1: 131-136.
- Klauder, D. S.; Petering, H. G. (1975) The protective value of dietary copper and iron against some toxic effects of lead in rats. *Environ. Health Perspect.* 12: 77-80.
- Klauder, D. S.; Petering, H. G. (1977) Anemia of lead intoxication: a role of copper. *J. Nutr.* 107: 1779-1785.
- Klein, A. W.; Koch, T. R. (1981) Lead accumulations in brain, blood and liver after low dosing of neonatal rats. *Arch. Toxicol.* 47: 257-262.
- Klein, M.; Namer, R.; Harper, E. P.; Corbin, R. (1970) Earthenware containers as a source of fatal lead poisoning. *New Engl. J. Med.* 283: 669-672.
- Kostial, K.; Kello, D. (1979) Bioavailability of lead in rats fed "human" diets. *Bull. Environ. Contam. Toxicol.* 21: 312-314.
- Kostial, K.; Momcilovic, B. (1974) Transport of lead 203 and calcium 47 from mother to offspring. *Arch. Environ. Health* 29: 28-30.
- Kostial, K.; Simonovic, I.; Pisonic, M. (1971) Lead absorption from the intestine in newborn rats. *Nature* 233: 564.
- Kostial, K.; Kello, D.; Jugo, S.; Rabar, I.; Maljkovic, T. (1978) Influence of age on metal metabolism and toxicity. *Environ. Health Perspect.* 25: 81-86.
- Laug, E. P.; Kunze, F. M. (1948) The penetration of lead through the skin. *J. Ind. Hyg. Toxicol.* 30: 256-259.
- Lloyd, R. D.; Mays, C. W.; Atherton, D. R.; Bruenger, F. W. (1975) ²¹⁰Pb studies in beagles. *Health Phys.* 28: 575-583.
- Lepow, M. L.; Bruckman, L.; Rubino, R. S.; Markowitz, S.; Gillette, M.; Kapish, J. (1974) Role of airborne lead in increased body burden of lead in Hartford children. *Environ. Health Perspect.* 7: 99-102.
- Lorenzo, A. V.; Gewurtz, M.; Maher, C.; Davidowski, L. I. (1977) The equilibration of lead between blood and milk of lactating rabbits. *Life Sci.* 21: 1679-1684.
- McLachlin, J. R.; Goyer, R. A.; Cherian, M. G. (1980) Formation of lead-induced inclusion bodies in primary rat epithelial cell culture: Effects of actinomycin D and cycloheximide. *Toxicol. Appl. Pharmacol.* 56: 418-431.

PRELIMINARY DRAFT

- Mackie, A. C.; Stephens, R.; Townsend, A.; Waldron, H. A. (1977) Tooth lead levels in Birmingham children. Arch. Environ. Health 32: 179-185.
- Mahaffey, K. R.; Michaelson, I. A. (1980) The interaction between lead and nutrition. In: Low Level Lead Exposure: The Clinical Implications of Current Research, H. L. Needleman, ed., Raven Press, New York. Pp. 159-200.
- Mahaffey, K. R.; Goyer, R. A.; Haseman, J. (1973) Dose response to lead ingestion in rats on low dietary calcium. J. Lab. Clin. Med. 82: 92-100.
- Mahaffey, K. R.; Treloar, S.; Banes, T. A.; Peacock, B. J.; Parekh, L. E. (1976) Difference in dietary intake of calcium and phosphorus in children having normal and elevated blood lead concentrations. J. Nutr. 106 xxx (Abs.).
- Mahaffey, K. R.; Annett, J. L.; Barbano, H. E.; Murphy, R. S. (1979) Preliminary analysis of blood lead concentrations for children and adults. HANES II, 1976-1978. Trace Subst. Environ. Health 13: 37-51.
- Mahaffey-Six, K.; Goyer, R. A. (1970) Experimental enhancement of lead toxicity by low dietary calcium. J. Lab. Clin. Med. 76: 933-942.
- Mahaffey-Six, K.; Goyer, R. A. (1972) The influence of iron deficiency on tissue content and toxicity of ingested lead in the rat. J. Lab. Clin. Med. 79: 129-136.
- Manton, W. I.; Cook, J. D. (1979) Lead content of cerebrospinal fluid and other tissues in amyotrophic lateral sclerosis (ALS). Neurology 29: 611.
- Marcus, A. H. (1979) The body burden of lead: Comparison of mathematical models of accumulation. Environ. Res. 19: 79-90.
- Marcus, S. M. (1982) Experience with D-penicillamine in treating lead poisoning. Vet. Hum. Toxicol. 24: 18-20.
- Markowitz, M. E.; Rosen, J. F. (1981) Zinc and copper metabolism in CaNa_2 EDTA-treated children with plumbism. Pediatr. Res. 15: 635.
- Mehani, S. (1966) Lead retention by the lungs of lead exposed workers. Ann. Occup. Hyg. 9: 165-171.
- Momcilovic, B. (1978) The effect of maternal dose on lead retention in suckling rats. Arch. Environ. Health 33: 115-117.
- Momcilovic, B.; Kostial, K. (1974) Kinetic of lead retention and distribution in suckling and adult rats. Environ. Res. 8: 214-220.
- Moore, M. R.; Meredith, P. A.; Watson, W. S.; Sumner, D. J.; Taylor, M. K.; Goldgerg, A. (1980) The percutaneous absorption of lead-203 in humans from cosmetic preparations containing lead acetate as assessed by whole body counting and other techniques. Fd. Cosmet. Toxicol. 18: 399-405.

PRELIMINARY DRAFT

- Morgan, A.; Holmes, A. (1978) The fate of lead in petrol-engine exhaust particulates inhaled by the rat. *Environ. Res.* 15: 44-56.
- Morgan, A.; Holmes, A.; Evans, J. C. (1977) Retention, distribution and excretion of lead by the rat after intravenous injection. *Brit. J. Ind. Med.* 34: 37-42.
- Morrison, J. N.; Quarterman, J.; Humphries, W. R. (1977) The effect of dietary calcium and phosphate on lead poisoning in lambs. *J. Comp. Pathol.* 87: 417-429.
- Morrow, P. E.; Beiter, H.; Amato, F.; Gibb, F. R. (1980) Pulmonary retention of lead: an experimental study in man. *Environ. Res.* 21: 373-384.
- Mortensen, R. A. (1942) The absorption of lead tetraethyl with radioactive lead as indicator. *J. Hyg. Toxicol.* 24: 285.
- Munro, I. C.; Willes, R. F.; Truelove, J. F. (1975) Absorption and tissue distribution of inorganic lead in the developing infant monkey. *Toxicol. Appl. Pharmacol.* 33: 128 (Abs. No. 17).
- Mykkönen, H. M.; Wasserman, R. H. (1981) Gastrointestinal absorption of lead (²⁰³Pb) in chicks: Influence of lead, calcium and age. *J. Nutr.* 111: 1757-1765.
- Mykkönen, H. M.; Dickerson, J. W. T.; Lancaster, M. C. (1979) Effect of age on the tissue distribution of lead in the rat. *Toxicol. Appl. Pharmacol.* 51: 447-454.
- National Academy of Science: National Research Council. (1976) Recommendations for the Prevention of Lead Poisoning in Children. Washington, D.C.
- Needleman, H. L.; Shapiro, J. M. (1974) Dentine lead levels in asymptomatic Philadelphia school children: Subclinical exposure in high and low risk groups. *Environ. Health Perspect.* 7: 27-33.
- Nielsen, T.; Jensen, K. A.; Grandjean, P. (1978) Organic lead in normal human brains. *Nature* 274: 602.
- Niklowitz, W. J.; Mandybur, T. I. (1975) Neurofibrillary changes following childhood lead poisoning. *J. Neuropathol. Exp. Neurol.* 34: 445-455.
- Nozaki, K. (1966) Method for studies on inhaled particles in human respiratory system and retention of lead fume. *Ind. Hlth. (Japan)* 4: 118-128.
- Nygaard, S. P.; Ottosen, J.; Hansen, J. C. (1977) Whole-blood lead concentration in Danes: relation to age and environment. *Dan. Med. Bull.* 24: 49-51.
- O'Flaherty, E. J.; Hammond, P. B.; Lerner, S. I. (1982) Dependence of apparent blood lead half-life on the length of previous lead exposure in humans. *Fundam. Appl. Toxicol.*, in press.

PRELIMINARY DRAFT

- Okazaki, H.; Aronson, S. M.; DiMaio, D. J.; Olvera, J. E. (1963) Acute lead encephalopathy of childhood. *Trans. Am. Neurol. Assoc.* 88: 248-250.
- Ong, C. N.; Lee, W. R. (1980a) Distribution of lead-203 in human peripheral blood in vitro. *Brit. J. Ind. Med.* 37: 78-84.
- Ong, C. N.; Lee, W. R. (1980b) High affinity of lead for fetal hemoglobin. *Brit. J. Ind. Med.* 37: 292-298.
- Ong, C. N.; Lee, W. R. (1980c) Interaction of calcium and lead in human erythrocytes. *Brit. J. Ind. Med.* 37: 70-77.
- Owen, G.; Lippmann, G. (1977) Nutritional status of infants and young children. *Pediatr. Clin. North Am.* 24: 211-227.
- Pott, F.; Brockhaus, A. (1971) Vergleich der enteralen and pulmonalen resorptions quote von bleibindungen. *Zentralb. Bakt. Hyg. Orig. B.* 155: 1-17.
- Pounds, J. G.; Wright, W. R. (1982) Cellular metabolism of lead: a kinetic analysis in the rat hepatocyte. *Toxicologist* 2: 55.
- Pounds, J. G.; Marlar, R. J.; Allen, J. R. (1978) Metabolism of lead-210 in juvenile and adult Rhesus monkeys (*Macaca mulatta*). *Bull. Environ. Contam. Toxicol.* 19: 684-691.
- Prêrovská, I.; Teisinger, J. (1970) Excretion of lead and its biological activity several years after termination of exposure. *Brit. J. Ind. Med.* 27: 352-355.
- Prpic-Majic, D.; Mueller, P. K.; Beritic, T.; Stanley, S. A.; Turiss, S. (1973) Delta-amino-levulinic acid dehydratase activity, blood lead levels and the reticulocyte count. In: *Environmental Health Aspects of Lead. Proceedings of an International Symposium. Amsterdam, 1972. C.E.C., Luxembourg, pp. 211-220.*
- Quarterman, J.; Morrison, J. N. (1975) The effect of dietary calcium and phosphorus on the retention and excretion of lead in rats. *Brit. J. Nutr.* 34: 351-362.
- Quarterman, J.; Morrison, E. (1978) The effect of age on the absorption and excretion of lead. *Environ. Res.* 17: 78-83.
- Quarterman, J.; Morrison, J. N.; Humphries, W. R. (1978) The influence of high dietary calcium and phosphate on lead uptake and release. *Environ. Res.* 17: 60-67.
- Quarterman, J.; Morrison, E.; Morrison, J. N.; Humphries, W. R. (1978) Dietary protein and lead retention. *Environ. Res.* 17: 68-77.
- Rabinowitz, M. B.; Wetherill, G. W.; Kopple, J. D. (1973) Lead metabolism in the normal human: stable isotope studies. *Science* 182: 725-727.

PRELIMINARY DRAFT

- Rabinowitz, M. B.; Wetherhill, G. W.; Kopple, J. D. (1974) Studies of human lead metabolism by use of stable isotope tracers. *Environ. Health Perspect.* 7: 145-153.
- Rabinowitz, M. B.; Wetherhill, G. W.; Kopple, J. D. (1976) Kinetic analysis of lead metabolism in healthy humans. *J. Clin. Invest.* 58: 260-270.
- Rabinowitz, M. B.; Wetherhill, G. W.; Kopple, J. D. (1977) Magnitude of lead intake from respiration by normal man. *J. Lab. Clin. Med.* 90: 238-248.
- Rabinowitz, M. B.; Kopple, J. D.; Wetherill, G. W. (1980) Effect of food intake and fasting on gastrointestinal lead absorption in humans. *Am. J. Clin. Nutr.* 33: 1784-1788.
- Rader, J. I.; Peeler, J. T.; Mahaffey, K. R. (1981) Comparative toxicity and tissue distribution of lead acetate in weanling and adult rats. *Environ. Health Perspect.* 42: 185-195.
- Rader, J. I.; Celesk, E. M.; Peeler, J. T.; Mahaffey, K. R. (1982) Tissue distribution and retention of equivalent doses of lead acetate in young and adult rats. *Trace Subst. Environ. Hlth.* 15. In press.
- Raghavan, S. R. V.; Gonick, H. C. (1977) Isolation of low-molecular-weight protein from human erythrocytes. *Proc. Soc. Exp. Biol. Med.* 155: 164-167.
- Raghavan, S. R. V.; Culver, B. D.; Gonick, H. C. (1980) Erythrocyte lead-binding protein after occupational exposure. I. Relationship to lead toxicity. *Environ. Res.* 22: 264-270.
- Randall, R. E. G.; Baily, P.; Soskolne, C. L. (1975) The effect of particle size in absorption of inhaled lead. *J. Am. Ind. Hyg. Assoc.* 36: 207-213.
- Rastogi, S. C.; Clausen, J. (1976) Absorption of lead through the skin. *Toxicol.* 6: 371-376.
- Robinson, T. R. (1974) Delta-amino levulinic acid and lead in urine of lead antiknock workers. *Arch. Environ. Health* 28: 133-139.
- Rosen, J. F.; Markowitz, M. E. (1980) D-penicillamine: its actions on lead transport in bone organ culture. *Pediatr. Res.* 14: 330-335.
- Rosen, J. F.; Zarata-Salvador, C.; Trinidad, E. E. Plasma lead levels in normal and lead-intoxicated children. *J. Ped.* 84: 45-48.
- Rosen, J. F.; Chesney, R. W.; Hamstra, A. J.; DeLuca, H. F.; Mahaffey, K. R. (1981) Reduction in 1,25-dihydroxy vitamin D in children with increased lead absorption. In: *Organ-Directed Toxicity. Chemical Indices and Mechanisms*, S. S. Brown and D. S. Davies, eds., Pergamon Press, New York. Pp. 91-95.
- Ryu, J. E.; Ziegler, E. E.; Forman, S. J. (1978) Maternal lead exposure and blood lead concentration in infancy. *J. Pediatr.* 93: 476-478.

PRELIMINARY DRAFT

- Saegner, P.; Rosen, J. F.; Markowitz, M. E. (1982) The diagnostic significance of CaNa_2EDTA testing in children with increased lead absorption. *Amer. J. Dis. Child.* 136: 312-315.
- Sartor, F. A.; Rondia, D. (1981) Setting legislative norms for environmental lead exposure; results of an epidemiological survey in the east of Belgium. *Toxicol. Lett.* 7: 251-257.
- Scalon, J. (1971) Umbilical cord lead concentration. *Amer. J. Dis. Child.* 121: 325.
- Shelling, D. H. (1932) Effect of dietary calcium and phosphorus on toxicity of lead in the rat: rationale of phosphate therapy. *Proc. Soc. Exp. Biol. Med.* 30: 248-254.
- Sherlock, J.; Smart, G.; Forbes, G. I.; Moore, M. R.; Patterson, W. J.; Richards, W. N.; Wilson, T. S. (1982) Assessment of lead intakes and dose-response for a population in Ayr exposed to a plumbosolvent water supply. *Human Toxicol.* 1: 115-122.
- Smith, C. M.; DeLuca, H. F.; Tanaka, Y.; Mahaffey, K. R. (1978) Stimulation of lead absorption by vitamin D administration. *J. Nutr.* 108: 843-847.
- Sobel, A. E.; Gawron, D.; Kramer, B. (1938) Influence of vitamin D in experimental lead poisoning. *Proc. Soc. Exp. Biol. Med.* 38: 433-435.
- Sobel, A. E.; Yuska, H.; Kramer, B. (1940) The biochemical behavior of lead. I. Influence of calcium, phosphorus and vitamin D on lead in blood and bone. *J. Biol. Chem.* 132: 239.
- Sorrell, M.; Rosen, J.; Roginsky, M. (1977) Interactions of lead, calcium, vitamin D and nutrition in lead-burdened children. *Arch. Environ. Health* 32: 160-164.
- Steenhout, A.; Pourtois, M. (1981) Lead accumulation in teeth as a function of age with different exposures. *Brit. J. Ind. Med.* 38: 297-303.
- Stephens, R.; Waldron, H. A. (1975) The influence of milk and related dietary constituents on lead metabolism. *Fd. Cosmet. Toxicol.* 13: 555-563.
- Stevens, C. D.; Feldhake, C. J.; Kehoe, R. A. (1960) Isolation of triethyl lead ion from liver after inhalation of tetraethyl lead. *J. Pharmacol. Exp. Ther.* 128: 90-94.
- Stowe, H. D.; Goyer, R. A.; Krigman, M. R.; Wilson, M.; Cates, M. (1973) Experimental oral lead toxicity in young dogs. *Arch. Pathol.* 95: 106-116.
- Stuik, E. J. (1974) Biological response of male and female volunteers to inorganic lead. *Int. Arch. Arbeitsmed.* 33: 83-97.
- Task Group on Lung Dynamics. (1966) Deposition and retention models for internal dosimetry of the human respiratory tract. *Health Phys.* 12: 173-307.

PRELIMINARY DRAFT

- Thompson, J. A. (1971) Balance between intake and output of lead in normal individuals. *Brit. J. Ind. Med.* 28: 189-194.
- Tola, S.; Hernberg, S.; Asp, S.; Nikkanen, J. (1973) Parameters indicative of absorption and biological effect in new lead exposure: A prospective study. *Brit. J. Ind. Med.* 30: 131-141.
- Trotter, M.; Hixon, B. B. (1974) Sequential changes in weight, density and percentage ash weight of human skeletons from an early fetal period through old age. *Anat. Rec.* 179: 1-18.
- U.S. Environmental Protection Agency, Criteria and Standards Division. (1979) The Environmental Lead Problem. An Assessment of Drinking Water from a Multi-Media Perspective. NTIS No. BB-296 556.
- Willoughby, R. A.; Thirapatsakun, T.; McSherry, B. J. (1972) Influence of rations low in calcium and phosphorus on blood and tissue lead concentrations in the horse. *Amer. J. Vet. Res.* 33: 1165-1173.
- Winnecke, G.; Brockhaus, A.; Kramer, Y.; Ewers, M.; Kiyamek, G.; Lechner, H.; Janke, W. (1981) Neuropsychological comparison of children with different tooth levels. International Conference: Heavy Metals in the Environment, Amsterdam. CEP Consultants, Edinburgh. Pp. 553-556.
- World Health Organization. Environmental Health Criteria. 3. Lead. United States Environmental Programme. Geneva, 1977.
- Yamamura, Y.; Takakura, J.; Hirayama, F.; Yamauchi, H.; Yoshida, M. (1975) Tetraethyl lead poisoning caused by cleaning work in the aviation fuel tank. *Jap. J. Ind. Health* 17: 223.
- Yip, R.; Norris, T. N.; Anderson, A. S. (1981) Iron status of children with elevated blood lead concentrations. *J. Pediatr.* 98: 922-955.
- Yu, Y.; Alvarez, G. H.; Mahaffey, K. R. (1978) Comparative effects of feeding lead acetate and phospholipid-bound lead on blood and tissue lead concentrations in young and adult rats. *Bull. Environ. Contam. Toxicol.* 20: 561-567.
- Ziegler, E. E.; Edwards, B. B.; Jensen, R. L.; Mahaffey, K. R.; Forman, S. J. (1978) Absorption and retention of lead by infants. *Pediatr. Res.* 12: 29-34.
- Zielhuis, R. L.; Del Castilho, P.; Herber, R. F. M.; Wibowo, A. A. E. (1978) Levels of lead and other metals in human blood: suggestive relationships, determining factors. *Environ. Health Perspect.* 25: 103-110.