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Lead Toxicity: From Overt to Subclinical to Subtle Health Effects

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Although the toxicity of lead was recognized centuries ago, concern was restricted to overt symptoms: colic, encephalopathy, anemia, or renal disease. Two major reasons for lack of progress in restricting toxicity were that interest was limited to occupational exposures and there was a lack of awareness of specific biochemical or metabolic effects. Identification of subclinical effects has been possible the last 15 or 20 years because of the development of sensitive measures to detect cognitive and behavioral changes that are not apparent clinically and because of methods to measure the reduced activity of heme enzymes. This progress was driven by basic and clinical research that resulted in a better understanding of cellular toxicology. The new awareness prompted the lowering of acceptable occupational exposures, as measured by blood lead from 80 to 40 to 60 μg/dL range, and the establishment of maximum recommended exposures in children to a blood lead level of 25 μg/dL. Lowering the lead content in gasoline has been accomplished by a nearly 50% decrease in average blood levels of persons in the United States (NHANES II data). Current research implicates lead as a contributing etiologic factor in a number of common diseases affecting large portions of the population such as subtle cognitive and neurological deficits, hypertension, congenital malformations, immunotoxicity, and deficits in growth and development. For each of these disorders there may be multiple etiologic factors; the scientific challenge is to develop sensitive methodology to detect the specific role of lead. Other potential subtle health effects include the influence of small amounts of lead on cell proliferation and lead as a cofactor in carcinogenesis. At the molecular level, lead may be competing with calcium to alter critical cell functions such as ion transport, energy production, and the function of heme-containing enzymes. The public health need for the next century will be to provide an environment that will reduce human exposure to lead to a level consistent with optimal human health. Although considerable success has been achieved through reduction of air emissions, decreases in lead levels found in food and water must occur in the face of increasing solubilization and mobilization of this metal by other environmental influences such as the acid rain phenomenon.

Introduction

The objective of this brief paper is to provide an overview of human health effects of lead, bringing particular attention to the gradual reduction of acceptable levels of lead exposure, since the clinical recognition of lead toxicity has progressed from overt to subclinical to subtle effects. The history of lead usage and health effects goes back more than 20 centuries, with a remarkably repetitive record of lead poisoning occurring well into this century. It is difficult to completely answer why so little progress has been made in controlling lead toxicity in previous years, which was not for lack of awareness because lead toxicity was frequently written about. One reason for the lack of change often cited was that the focus historically was on occupational toxicity—on the worker rather than the general population. Another reason may be the nature of clinical lead toxicity. It is an insidious problem; affected people had to develop overt illness, a severe anemia, or a neuropathy or encephalopathy before being recognized as being ill.

But all of this has changed in the past 15 to 20 years. We now realize that overt and subclinical toxicity form a continuum and the difference depends on the eyes one uses to see an effect, whether or not it is a child with an ataxic gait or a child whose nervous system function is measured by sensitive neurophysiologic probes. Both approaches measure toxicity; the difference in terms of health effect is only one of degree. However, to define and measure subclinical effects, we have had to understand pathogenetic mechanisms and develop methods to measure them; this has only been possible because of the continuing interests of a large number of scientists from a variety of clinical and basic disciplines. As a result there has been a progressive decrease in acceptable levels of exposure, both in the workplace and among the general public particularly young children. This work has not really been completed, so the present phase can best be described as the treatment or corrective phase.

The level of lead reduction that is actually achievable in the future is uncertain. Some have argued that the natural human exposure to lead is negligible (*1*). Certainly, at the present time, lead is ubiquitous in the

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environment. The metal is neither created or destroyed by man, but industrial activity results in redistributing and sometimes transforming the metal to chemical forms that are more available and more toxic to man.

Lead is and will continue to be an important metal with many industrial uses, the principle one being the electric battery industry. However, leaded gasoline combustion in vehicles has accounted for as much as 90% of the total anthropogenic sources of environmental lead. From 1975 to 1984 lead in gasoline decreased 73%, while ambient air lead decreased 71% (*2*). However, all human exposure to lead is not from the inhalation of airborne lead. For about four decades now more than 100 to 200,000 tons of lead per year have been emitted from automobile exhausts in the United States with some fixation to soil, uptake by plants, and flow into water sources. Presently the major sources of human exposure to lead by persons in the general population are food and beverage because of lead uptake by food sources and its presence in drinking water. Solubilization and mobilization of lead is enhanced by a lower pH of water, so there must be some concern for the effects of the acid rain phenomenon and its contribution to human lead exposure (*3*). This exposure may be an instance where it is reduced by one corrective measure, the phase-out of lead in gasoline, only to have human exposure further enhanced by a second environmental problem, the acid rain phenomenon.

Discussion

So what is the challenge for the next century? The major scientific question will be to determine the lowest level of lead exposure that is toxic. There have been real advances in understanding the toxicology of lead over the past 15 to 20 years. Table 1 summarizes the levels of blood lead thought to be associated with different kinds of effects. The first three effects are signs of overt toxicity, which are easily recognized but not thought to occur until blood lead levels reached 80 μg/dL or more [the occupational health standard prior to 1978 (*4*)]. The recognition that subclinical but nonetheless harmful effects occur at lower blood lead levels began in the 1970s. Because the scientific understanding of lead effects and increasingly sensitive techniques for measuring them have evolved, the recognizable toxicological effect of lead has been found to occur at much lower levels. These findings have driven the need to restrict exposure, both in the workplace and ambient environment, to lower levels. The 1985 report from Centers for Disease Control (CDC) recommended that 25 μg/dL of blood lead be considered as the action level in children. However, there is a growing body of information that this level may be too high, particularly for the fetus (*5*). Although this figure illustrates the progress that has taken place, there is now a need to address the problem of subtle effects. These are effects that differ from clinical effects and can only be recognized by very sensitive epidemiological studies or by laboratory techniques. In general these are not lead-specific effects; they are common in the general population and they are multifactorial, that is, lead is only one of a number of possible etiological factors.

Cognitive and behavioral effects are perhaps the single most important subtle ones. These are discussed in detail later in this conference (*6*). An effect of lead on blood pressure was suggested more than 100 years ago (*7*), but until recently both epidemiological and experimental results were inconsistent, probably because of methodological differences and problems. A number of studies involving lead workers and people in the general population have shown a more consistent relationship between lead exposure and the increase in blood pressure or hypertension (*8,9*). Analyses of NHANES II data (*10*) from white males, ages 40 to 59 years, showed a significant association between blood lead and blood pressure, even after including—in multiple regression analyses—all known factors previously established as being correlated with blood pressure. No threshold was found where the blood lead level was not significantly related to blood pressure across a range of 7 to 34 μg/dL of blood lead. An interesting aspect of these studies is that large initial increments in blood pressure occurred at relatively low blood lead levels, followed by blood pressure increments leveling off at higher blood lead levels. A growing amount of experimental work provides mechanistic explanations for this effect of lead, and both the epidemiological and experimental studies to

Table 1. Blood lead levels (μg/dL) associated with different toxicological effects of lead and no-effect levels recognized by different government agencies.

Effect	Adults	Children	No effect
Gastrointestinal colic	> 80	> 80	60–70 Adults (WHO, 1977)[a]
Acute encephalopathy	> 80	> 80	50–60 Children (WHO, 1977)
Anemia	> 80	> 70	40–60 (OSHA, 1978)[a]
Chronic renal disease	> 60	—	30 (CDC, 1978)[a]
Cognitive and behavioral	> 30	> 15	
Peripheral neuropathy	> 30	> 20	
Reproductive testicular	> 40	—	
Increase EP	> 20–25	> 15	
ALA-D	> 20	> 10	
Pyridine-5-nucleosidase		> 10	
Subtle effects, tolerable level?			25 (CDC, 1985)

[a]WHO, World Health Organization; OSHA, Occupational Safety and Health Administration; CDC, Centers for Disease Control.

date were reviewed in a 1988 symposium (*11*).

An epidemiologic study reported by Needleman and co-workers (*12*) suggests a relationship between cord blood lead levels from 8.7 to >35 g/dL and a number of minor congenital malformations such as hemangiomas, minor skin anomalies, and undescended testes. However, these findings were not confirmed in two other studies (*13,14*).

In the late 1970s at least three epidemiologic studies relating blood lead levels to skeletal growth and stature suggested an adverse effect (*15,16*), but it has not been possible to separate lead effects from the contribution of dietary, racial, and other factors.

Schwartz et al. (*17*) analyzed anthropometric measurements with age, sex, and other variables and found that height, weight, and chest circumference were explained by five variables: sex, age, race, diet (that is calories or protein), and blood lead levels.

This association between lead exposure and growth is supported by animal studies and seems to have further support from ongoing prospective studies. However, at present there is no dose-response information and the mechanistic basis is largely speculative.

An important point to realize in relating these effects to lead is that in all three of these end points, the lead effect is only one of a large number of possible contributing factors. Probably the lead effect is never the only factor, so it may be recognized in population studies, but it is unlikely that the lead effect is accurately weighted in a particular individual.

In discussing subtle effects it is also appropriate to mention cellular or molecular effects that are measured only in cells of experimental animals or *in vitro* models. Figure 1 is a photomicrograph showing the ultrastructure of a kidney cell from a lead-poisoned rat. The same changes are seen in kidney biopsies of lead-exposed workmen. This is a sick cell; it is active and functioning but at a reduced level. The cell shows two major cellular effects of lead, the lead-induced nuclear inclusion body and ultrastructural changes in the mitochondria.

The lead-induced inclusion body is a lead-protein complex (*18*). It probably forms initially in the cytoplasm and migrates into the nucleus (*19*). The protein has been partially characterized (*20,21*), but its origin has not been determined. This protein migration may indeed be a very subtle effect of lead. Lead is bound to soluble and insoluble nuclear protein fractions with exposure from lead in the ambient environment (*20*). Lead does induce protein synthesis (*22,23*), and inhibitors of protein synthesis have impaired the formation of lead-protein complexes in cells in tissue culture (*19*), but the origin of the lead that forms the complex is not known. A positive view of this phenomenon is that it sequesters cellular lead from other organelles into a relatively harmless form (*24*), but there may be subtle effects of nuclear lead

FIGURE 1. Electron photomicrograph of proximal renal tubule of rat with lead toxicity. Nucleus contains dense, intranuclear inclusion body adjacent to nucleolus. Mitochondria are swollen with distorted cristae.

on the regulation of cell growth.

Whether or not lead is a carcinogen is not completely resolved at present, according to the International Agency for Research on Cancer (IARC) (*25*). A single injection of lead stimulates [^{3}H]-thymidine in renal tubular cells, which is not believed to be due to repair synthesis of DNA, because lead-stimulated DNA synthesis is followed by a wave of mitoses in tubular epithelium that does not occur after repair synthesis of DNA. Lead induces liver cell proliferation (*26*), but lead-induced liver cell hyperplasia does not support initiation by liver carcinogens (*27*). Lead does induce *in vitro* transformation of Syrian hamster embryo cell cultures (*28*),which implies that lead does in some way affect gene expression. Sirover and Loeb (*29*) found that lead salts decrease fidelity of DNA transcription, but lead is not mutagenic in the Salmonella (Ames test) assay for point mutations (*30*) or in the host-mediated assay in mice (*31*).

Lead does induce renal tumors in male rats and other rodents but not in all species of animals. Epidemiologic studies of lead smelter workers and battery workers have demonstrated a significant excess of malignancies at all sites and the lungs but not specifically the kidneys (*32*).

The lead effect on functions of the mitochondrion site may be the major site for the subtle biochemical effects of lead. Lead was shown to reduce oxidative phosphorylation in kidney mitochondria about 20 years ago and it brought attention to the mitochondrion as perhaps the most sensitive organelle or intracellular focus for lead toxicity (*33*). Impaired energy metabolism results in reduction of the normal function of the cell regardless of the organ system. Lead has been shown to affect the function of mitochondria in every organ that reflects lead toxicity clinically: the brain, the kidney, and the hematopoietic system in particular. There is also impairment of other functions of the mitochondrion including sodium and potassium ATPases, and Ca ATPases that lead to impaired cation regulation, particularly with regard to calcium (*34*). Calcium serves as an important intracellular messenger, and small changes in the calcium concentration bring about large changes in cell response so that regulation of calcium fluxes across plasma membrane are critical to proper cell function (*35*). For instance, calcium serves as a messenger in both neural and endocrine cells, so small amounts of lead in neural cells may bring about changes in neuronal function (*36*). Also, calcium serves as an important intracellular messenger in the regulation of cardiac and smooth muscle function and may relate to the pathogenesis of hypertension (*37*). Effects on calcium homeostasis in smooth muscle cells in arterioles may be the basis for the role of lead in hypertension.

Lead has been found to have multiple effects on heme metabolism, producing several different clinical effects (*38*). The major effect is on erythropoiesis, but there are also subtle effects on heme-containing enzyme systems, such as cytochrome P-450 systems, and decreased vitamin D metabolism. This metabolism requires a heme-containing hydroxylase in the kidney for the hydroxylation of 25-hydroxyvitamin D to 1,25-dehydroxyvitamin D, which is important in stimulating gastrointestinal absorption of calcium (*39*).

Finally, there are polymorphisms of the heme-synthesizing enzyme, ALA-D, which may have different degrees of sensitivity to lead and may be related to differences in susceptibility to a particular level of lead exposure. Whether or not there are different groups of people in the general population that have different susceptibilities to lead has been a perplexing problem. It has long been recognized that in the workplace several men may be doing the same job, working in the same environment, and only a small number actually develop symptoms of toxicity. The same is true in epidemiology studies of children: siblings in one family are affected whereas those next door are not affected. Various explanations have been offered for these differences such as diet, particularly the content of calcium and iron, and personal hygiene; for children these differences have been termed the mother effect that includes many things. Recently there has been some evidence suggesting that genetic polymorphisms of aminolevulinic acid dehydratase might account for some of the difference in the susceptibility between two people. Rogan and coworkers (*40*) noted that children with a more-than-average increase in erythrocyte protoporphyrin for a particular level of blood lead have a more-than-average decrease in *d*-aminolevulinic acid dehydratase (ALA-D), suggesting differences in responsiveness of this enzyme in different people. The difference in these enzymes has not yet been demonstrated in people having lead toxicity, but we now know that at least two different genetic forms of this enzyme are present in the general population (*41*).

As the reader can see, the mitochondrion is the site for several subtle and not-so-subtle effects of lead. A more complete understanding of the relationship of these effects to measurable health effects should provide a better basis for deciding what is the minimal toxic effect of lead.

Summary

Research over the past 15 to 20 years has resulted in methods of detection of subclinical effects of lead resulting in some lowering of human exposure. Many effects at the molecular level are also recognized but without knowledge of human dose-response. Challenges for next century are for toxicologist and epidemiologists to determine the minimum lead health effect and for environmentalists to provide an environment assuring minimal human exposure.

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