Lead hepatotoxicity & potential health effects

Anuradha Mudipalli

National Center for Environmental Assessment-RTP Division, Office of Research & Development, U.S. EPA, North Carolina, USA

Received March 22, 2007

Occupational and environmental exposures to lead (Pb), one of the toxic metal pollutants, is of global concern. Health risks are increasingly associated with environmental exposures to Pb emissions from, for example, the widespread use of leaded gasoline in developing countries. Exposure occurs mainly through the respiratory and gastrointestinal systems, and the ingested and absorbed Pb is stored primarily in soft tissues and bone. Autopsy studies of Pb-exposed patients have shown a large amount (~33%) of the absorbed Pb in soft tissue stored in liver. In addition to neuronal encephalopathy observed in persons after exposure to very high concentrations of Pb, gastrointestinal colic (abdominal pain, constipation, intestinal paralysis) is a consistent early symptom of Pb poisoning in humans. Such severe gastrointestinal effects are consistently observed in patients with a blood Pb range of 30 to 80 µg/dl. Ingestion of Pb is one of the primary causes of its hepatotoxic effects. Hepatocarcinogenic effects of Pb reported in animal toxicology studies have led to new research into the biochemical and molecular aspects of Pb toxicology. Gains in the molecular understanding of Pb effects on hepatic drug metabolizing enzymes, cholesterol metabolism, oxidative stress, and hepatic hyperplasia suggest a potential role for Pb in damaging extrahepatic systems, including the cardiovascular system. This review also discusses the therapeutic potential of chelation therapy in treating Pb-induced hepatotoxicity in animals.

Key words Biochemical mechanisms - Chelation therapy - hepatotoxicity - lead poisoning

Lead (Pb) had modest early uses in ancient medicines and cosmetics. Today, it has industrial uses in, for example, building materials, paints, and gasoline. As a result, Pb has become ubiquitous in our environment. Lead exposure primarily occurs through manmade sources. Representing preindustrial exposure, blood Pb levels of native Americans 700 to 1000 years ago are estimated to have been around 0.016 µg/dl, a level that is 50 to 200 times lower than the current blood Pb level range (0.78 to 3.2 µg/dl) of Himalayan populations with no industrial Pb exposure. Lead exposure mainly occurs through the respiratory and gastrointestinal systems. Absorbed Pb (whether inhaled or ingested) is stored in soft tissues. Autopsy studies of Pb-exposed humans indicate that liver tissue is the largest repository (33%) of Pb from among the soft tissues followed by kidney cortex and medulla. As environmental exposures to Pb have increased, the toxic effects of Pb on various organ systems in the body have been recognized.
Multimedia exposure to Pb through occupational and environmental settings is of major concern in developing countries, such as India. Leaching of Pb from cooking and storage vessels containing Pb into water and from lead-containing cooking vessels with acidic foods conditions contribute to the potential Pb exposure through ingestion. In India, elevated Pb concentrations in beer (10 mg/l) and the use of Pb arsenate pesticides also contribute to increased Pb exposure through ingestion. Hence, an evaluation of Pb exposure through the ingestion route is critical in assessing its health effects including the morbidity associated with its extrapulmonary effects such as those that occur in the gastrointestinal and hepatic systems.

The liver, via the portal vein, is the first organ exposed to enterally absorbed nutrients and other xenobiotics. The liver is composed of highly active metabolic tissue containing a huge complement of detoxification machinery referred to as phase I and phase II enzyme systems that ideally serve to guard other physiological systems from the toxic effects of xenobiotic compounds. Earlier studies on potential hepatotoxicity of Pb in experimental animal systems used relatively high doses of inorganic Pb salts. These studies reported alterations in hepatic xenobiotic metabolism, cholesterol metabolism, liver cell proliferation, and DNA synthesis indicative of Pb-induced hepatic hyperplasia. The exhaustive literature base available on Pb hepatotoxicity that has accumulated over the past two decades reports examinations of physiological, pathological, histochemical, and cellular and molecular biological endpoints (including endpoints at the level of gene expression and regulation) and provides newer insights into Pb hepatotoxicity. Experimental evidence from animal toxicology studies are reviewed to integrate relevant information on occupational exposure and epidemiologic studies.

**Hepatic drug metabolism**

**Phase I enzymes**: In the 1980s, acute exposure to Pb acetate was shown to decrease rat hepatic CYP450 content while increasing levels of urinary δ-aminolevulinic acid (ALA). A reversal of the decreased CYP450 levels was observed when rats were co-treated with CYP450 inducers such as phenobarbital, suggesting that the CYP450 decrease may be due to Pb-induced inhibition of heme synthetic enzymes. Decreased activities of the estradiol-17β-enzyme observed in rat liver treated with triethyl lead chloride indicated that not only inorganic but also organic Pb compounds inhibit CYP450 activities. A single dose of Pb nitrate (5 to 10 mmol/kg body wt) was found to decrease hepatic microsomal CYP450 enzyme levels as well as to decrease aminopyrene-N-demethylase activity. These decreases were followed by increases in levels of phase II components such as glutathione (GSH), glutathione-S-transferase (GST), and NAD(P)H:quinone oxidoreductase (DT diaphorase). This response mimics the biochemical phenotype of hepatocyte nodules, suggesting a carcinogenic potential for Pb nitrate and other Pb compounds.

Degawa et al investigated whether the Pb-induced decrease in CYP450 levels was due to inhibition of specific isoforms of CYP450. Their results indicated that Pb nitrate preferentially inhibits cytochrome P4501A2 activity. Inducers such as 3 methylcholanthrene and 2-methoxy-4-aminoazobenzene also preferentially inhibit cytochrome P4501A2 at both the protein and mRNA levels. Comparative study with other metals (i.e., nickel, cobalt, cadmium) indicated that the specific inhibition of P4501A2 was unique to lead nitrate among studied metals. This specific inhibition of P4501A2 by Pb nitrate was found to be due to the inhibition of heme synthesis. Degawa et al also reported that Pb nitrate inhibited the induction of CYP1A mRNA by aromatic amines but not by aryl hydrocarbons, suggesting a role for other cellular factors in the transcriptional activation of CYP1A genes. Lead nitrate-induced inhibition of CYP450 activities conferred protection from carbon tetrachloride (0.3 ml/kg)-induced hepatotoxicity. Subchronic (2- to 3-month) exposure to Pb-acetate (5 to 50 mg/kg body wt) has been found to inhibit CYP450s and cytochrome b5 in rat liver and kidney. The Pb-induced inhibition in hepatic CYP450s appears to be mediated by two different mechanisms: (i) the inhibitory effects of Pb at the level of transcription, or (ii) the decreased synthesis of heme and the subsequent decrease in heme saturation of the P450/apo P450 ratio. For example, Pb nitrate-induced production of tumour necrosis factor-alpha (TNF-α) in rat liver has been implicated in the suppression of constitutive expression of CYP1A2 mRNA in rat hepatocytes. The inhibition of constitutive and aromatic amine-induced expression of CYP1A2 in rat liver may be in part mediated by TNF-α-associated mechanisms, including transcriptional regulation.

**Phase II enzymes**: As mentioned above, a single injection of lead nitrate can induce GST activity.
Further, immunohistochemical analyses have indicated that a single injection of Pb nitrate can also induce hepatocyte glutathione-S-transferase-placental (GST-P), an enzyme that is otherwise undetectable in normal rat liver\textsuperscript{11,12}. Both lead acetate and lead nitrate were found to induce GST-P in rat liver. Detailed investigations into the molecular mechanisms underlying the induction of GST-P revealed that regulation occurred at various levels viz., transcriptional, post-transcriptional, and the post-translational. Suzuki et al\textsuperscript{13} used a transgenic approach to investigate the transcriptional regulation of Pb-induced GST-P and identified GST-P enhancer I (GPEI), an enhancer whose core consists of two AP-1 (c-Jun/c-Fos heterodimer) site-like sequences located at the 5' flanking region of this gene. These authors demonstrated that GPEI is an essential element in the Pb-associated activation of GST-P and that the trans-activating factor AP-1 is likely to be involved, at least in part, in the transcriptional activation of the GST-P gene by Pb via the GPEI sequence. On the other hand, a few other investigators\textsuperscript{14-16} have reported suppression of GST activity with a concomitant decrease in GSH levels on exposure to both inorganic and organic Pb compounds. Other hepatic GSH-dependent enzymes such as GSH peroxidase and GSH reductase were suppressed on acute exposure to Pb\textsuperscript{17}.

**Human hepatic injury and cytochrome P450 function**

Occupational exposure to Pb at levels resulting in high blood Pb concentrations (>70 µg/dl) in factory workers\textsuperscript{18-20} has been found to increase phase II hepatic enzyme activity with simultaneous suppression in cytochrome P450 activity. Panel studies performed to evaluate possible associations between Pb exposure and liver injury by measuring serum enzymes (i.e., alkaline phosphatase and lactate dehydrogenase) provided mixed results depending on blood Pb levels. A study on 100 male workers (gas station attendants and garage, printing, and construction workers) indicated significantly higher serum levels of alkaline phosphatase and lactate dehydrogenase when their mean blood Pb levels were 78 µg/dl\textsuperscript{21}. Such alterations were not found in a longitudinal study of battery workers whose mean blood Pb levels ranged from 60 µg/dl in 1989 to 30 µg/dl in 1999\textsuperscript{22}.

The effects of Pb exposure on hepatic CYP450 activity were indirectly assessed in children and adults by measuring urinary levels of P450 metabolites in a few clinical epidemiologic studies. Saenger et al\textsuperscript{23} measured urinary excretion of 6-β-hydroxycortisol, a metabolite derived primarily from the oxidation of cortisol and mediated by hepatic CYP3A4 activity, in children. Children with high blood Pb levels that were qualified for chelation therapy (EDTA-provoked urinary Pb levels of >500 µg/24 h) exhibited significantly lower excretion of 6-β-hydroxycortisol, suggesting decreased CYP3A4 activity. An epidemiological study (n = 118) performed in the general population of Bangkok, Thailand, evaluated the effects of Pb exposure on CYP2A6 activity by measuring urinary excretion of 7-hydroxy coumarin after a single oral dose of coumarin. The study reported a significant association between increased urinary Pb and decreased 7-hydroxy coumarin excretion in male, but not in female, adults\textsuperscript{24}.

**Cholesterol metabolism**

Lead-induced hyperplasia involves alterations in hepatic cholesterol metabolism that results in simultaneous increase in both liver and serum total cholesterol levels. Contrary to the general trend of suppression of CYP-450s as discussed above, lanosterol 14α-demethylase (CYP51), an essential enzyme for cholesterol biosynthesis, was found induced in Pb nitrate-mediated liver hyperplasia\textsuperscript{25,26}.

Lead nitrate-mediated development of hepatic hypercholesterolaemia involves the activation of cholesterol biosynthetic enzymes (i.e., 3-hydroxy-3-methylglutaryl-CoA reductase, farnesyl diphosphate synthase, squalene synthase, CYP51) and the simultaneous suppression of cholesterol-catabolic enzymes such as 7α-hydroxylase\textsuperscript{25}. The biochemical pathway for cholesterol synthesis showing the specific site(s) for potential Pb interactions is depicted in Fig.1.

On the other hand, Pillai and Gupta\textsuperscript{26} reported suppression in the activities of the hepatic steroid metabolizing enzyme 17-β-hydroxy steroid reductase in rat pups following gestational and lactational Pb exposures. Recent studies designed to evaluate the molecular events associated with this induction process investigated the role of transcriptional regulation. The 17-β-hydroxy steroid reductase gene has various regulatory elements and its constitutive expression in liver is mediated by sterol regulatory element (SRE) and by the SRE binding proteins-1a, 2, and 1c. Kojima et al\textsuperscript{27} reported that Pb nitrate induced expression of CYP51 in the livers of both immature (4 wk old) and mature (7 wk old) rats and that this induction appeared to be mediated by the upregulation of SRE binding protein-2. The induction of the cytokines interleukin-
to increase cellular uptake of Pb and demonstrated inhibition of ALAD activity in primary rat hepatocyte cultures.

The competition for the iron transporter DMT1 [divalent metal (ion) transporter 1] by Pb has been implicated in the incorporation of zinc into protoporphyrin, resulting in elevated levels of zinc protoporphyrin. Iron deficiency concomitant with Pb intoxication can further complicate the Pb effects on heme synthetic metabolism. Coproporphyrinuria, a well-characterized condition of Pb intoxication results from the effects of Pb on porphyrins, which are intermediary metabolites in heme synthesis.^23,33

Quintanilla-Vega et al.^34 reported that mouse 3T3 hepatocyte cultures, when incubated with a micromolar concentration of Pb acetate, increased cellular porphyrin content and excretion. This increased porphyrin production may have occurred following an accumulation of protoporphyrin and coproporphyrin, with resultant coproporphyrinuria.^32,33 Dietary supplementation of selenium and monensin were found to increase the Pb-induced porphyrin accumulation in chicken liver.^35 Investigations into the effect of Pb on zinc protoporphyrin synthesis in cultured chick and rat hepatocytes found decreased levels of protoporphyrin only in rat but not chick hepatocytes, suggesting species-specific differences in the effects of Pb on porphyrin metabolism.^16

Transferrin (TF) is the major iron-transport protein in serum and other biological fluids capable of transporting various metals. Lead was found to inhibit TF endocytosis and iron transport across the cell membrane of rabbit reticulocytes.^37 Studies by Adrian et al.^38 using transgenic mice with the human TF gene demonstrated Pb-mediated suppression of TF transgene expression in mouse liver at the transcriptional level. This suppression occurred without affecting mouse endogenous hepatic TF gene expression. Further studies by the same group using HepG2 cells in an in vitro culture system indicated Pb-mediated suppression of recombinant, as well as endogenous TF.^39 These observations suggest potential effects of Pb in the human hepatic system and a possible interference in iron metabolism. These studies also suggest that potential complications associated with Pb effects on iron metabolism occur in populations with iron deficiency conditions such as those prevalent in developing countries, including India.

**Fig. 1.** Schematic presentation of lead (Pb) mediated events in cholesterol synthesis pathway.

1α and TNF-α in rat liver prior to the induction of the genes for the synthesis of enzymes suggested that Pb nitrate-induced cholesterol synthesis is independent of sterol homeostasis regulation.

**Heme metabolism**

Heme metabolism in the liver is an essential component of various cytochrome P-450s that participate in cellular redox reactions and xenobiotic detoxification pathways in the liver tissue, hence heme plays a vital role in liver function. Lead-induced anaemia, the most commonly observed condition associated with severe Pb intoxication, results from a shortening of erythrocyte life span and an inhibition of haemoglobin synthesis. Lead effects on heme synthesis have been documented to occur in both animals and humans via its inhibitory effects on ALA dehydrogenase (ALAD), the enzyme involved in the final step of heme synthetic pathway. Fifteen per cent of heme is produced in the liver.

Initial studies by Lake and Gerchenson were the first to demonstrate Pb nitrate effects on hepatic heme biosynthesis in a rat liver cell line (RLC-GAI). Bondy investigated the effects of various organic metal compounds on ALAD activity and reported that triethyl lead chloride has the same potency as lead nitrate in inhibiting ALAD both in vitro and in vivo. These studies also demonstrated that both liver and blood ALAD exhibit similar sensitivity to Pb compounds. Another group used the Pb acetate + dithiocarbamate complex
Mediators and molecular mechanisms implicated in Pb-induced hepatic hyperplasia

Lead nitrate, a known mitogen, is considered a carcinogen that induces liver cell proliferation in rats without any accompanying liver cell necrosis. It has been recognized that this proliferation is a transient process and that apoptosis plays a major role in the regression of lead nitrate-induced hepatic hyperplasia. Initial studies that used DNA replication as an indicator of the cell proliferation by monitoring titrated thymidine incorporation into DNA reported that a single injection of Pb nitrate initiated proliferation by 3 days, with complete regression of the induced proliferation by 15 days. Using liver-derived REL cells in vitro cell culture Apostoli et al demonstrated dose- and time-dependent induction of cell proliferation by other Pb salts (Pb acetate, Pb chloride, Pb monoxide, Pb sulphate). Unlike other tumour promoters, the Pb compounds did not exhibit effects on cell junctional coupling. Lead nitrate-induced liver hyperplasia was also shown to result in sexual dimorphism in all phases of the proliferation as well as in apoptosis.

Earlier studies to investigate potential molecular mechanisms involved in Pb-induced hepatic hyperplasia indicated that peak DNA synthesis occurs at 36 h after a single injection of Pb nitrate (10 µM/100 g body wt) along with induced expression of c jun, c myc, and c Ha-ras oncoproteins. Significant change in liver DNA hypomethylation with age as well as liver cell specificity with Pb treatment was also reported. The DNA synthesis associated with Pb treatment appeared to be due to increased activity and expression of DNA polymerase-β. Differential activation of various protein kinase C (PKC) isoforms (downregulation of PKC-α, and marked activation of PKC-ε) suggest their involvement in DNA synthesis and related signal transduction pathways.

Proliferation of normal and pre-neoplastic hepatic cells treated with plasma from male Wistar rats given a single injection of Pb nitrate is indicative of a potential involvement of secreted growth factors in Pb-induced hepatic hyperplasia. Efforts to identify involvement of specific growth factors in liver cell proliferation indicated no role for hepatocyte growth factor or for the tumour growth factors, TGF-α and TGF-β. However, the enhanced expression of TNF-α preceding the onset of hepatocyte DNA synthesis suggests a role for TNF-α in Pb nitrate-induced hepatocyte proliferation. Additional studies carried out by the same group using a series of inhibitors to suppress Pb nitrate-induced expression of liver TNF-α demonstrated ablation of hepatocyte proliferation. Further, these studies also demonstrated that Pb nitrate-induced proliferation is not restricted to hepatocytes as it also occurs in other cell types (e.g., Kupffer cells, endothelial cells, perportal nondescript cells).

Lead nitrate-induced secretion of TNF-α is also known to play a role in conferring sensitivity to lipopolysaccharide (LPS) treatment. Lead nitrate-induced proliferation involved induction of inducible nitric oxide synthase (iNOS) along with TNF-α and that expression was mediated by nuclear factor kappa B (NF-kB). Lead nitrate-induced expression of neurotrophins such as nerve growth factor, brain-derived neurotrophic factor neurotrophin-3 and their receptors tyrosine kinase receptor (Trk) and neurotrophin receptor (p75NTR) suggests their potential role in mediating the mitogenic signals related to hepatic hyperplasia.

Oxidative stress

The accumulation of significant amounts of Pb in liver tissue was implicated in the induction of an oxidative stress response in the liver. Although Pb is considered to be a poor inducer of oxidative stress, lipid peroxidation with concomitant inhibition of several antioxidant enzymes such as superoxide dismutase (SOD), catalase, GSH peroxidase, GSH reductase was reported. This was accompanied by a simultaneous increase in glutathione disulphide (GSSG) and a reduction in the GSH/GSSG ratio. The mechanisms of Pb-induced oxidative stress are depicted in Fig. 2. Along with its role in Pb-induced hepatotoxicity, oxidative stress was also noted to play a significant role in the regression phase of hepatic hyperplasia with the generation of lipoperoxide (LPO) and other oxidants and the induced expression of cytokine mediators, including TNF-α. These mediators are also associated with the significant decline in intracellular ATP concentration observed in mouse hepatocyte culture and in oxidative DNA damage and the apoptosis of hepatocytes.

Using freshly isolated cultures of hepatocytes and Kupffer cells in a co-culture system exposed to Pb acetate (2 to 50 µM) and LPS (0.1 to 1000 ng/ml), Pagliara et al reported a role for proteolysis in Pb-induced hepatocyte cell death. In vitro Pb nitrate treatment did not induce apoptosis in cultured hepatocytes. However, hepatocyte apoptosis was evident when the hepatocytes were incubated with culture medium derived from Kupffer cells that had been
exposed to Pb nitrate. These experiments demonstrated that proteolysis can be initiated by Kupffer cell-derived signals. In *in vivo* experiments using gadolinium chloride, a Kupffer cell toxicant, to specifically destroy Kupffer cells, it was demonstrated that Pb nitrate treatment did not induce hepatic apoptosis or an oxidative response, confirming the role for Kupffer cell-derived factors in hepatocyte proteolysis and ultimate cell death. Pre-treatment of rats with gadolinium chloride was also found to abolish the altered expression of galactose receptors. The role of activated Kupffer cells, macrophages, and TNF-α in chemical-induced hepatotoxicity is presented schematically in Fig. 3.

The role of glucocorticoid-mediated signal transduction in the hepatotoxicity of Pb was evaluated by Heiman and Tonner using H4-II-E-C3 hepatoma cells (HTC). Acute exposure of cells to Pb (10 µM) was found to inhibit processes involved in glucocorticoid-mediated enzyme induction (*e.g.*, tyrosine aminotransferase activity) in a dose-dependent manner at both the transcriptional and translational level without altering glucocorticoid receptor binding characteristics. Tonner and Heiman also reported Pb induced hepatotoxicity by glucocorticoid-mediated signaling and its involvement in the interference with calcium-mediated events as well as the differential modulation and translocation of protein kinase isoforms α and β into the nucleus.

**Therapeutic intervention of Pb-induced hepatic toxicity and oxidative stress**

The current approved clinical intervention to mitigate Pb toxicity is to give chelating agents that form an insoluble complex with Pb to remove it from Pb-burdened tissues. Supplementation with antioxidant agents was explored in experimental laboratory animals to evaluate the protective effects of these agents in reducing overall toxicity, particularly hepatotoxicity. Initial chelation intervention studies aimed at reducing Pb toxicity in infants were explored in rat pups. Single agent chelation therapy with EDTA, or combination therapy with the mono-3-methylbutan-1-yl (monoisomyl) ester of meso-2,3-dimercaptosuccinic acid (Mi-DMSA) and meso-2, 3-dimercaptosuccinic acid (meso-DMSA) did not confer protection to suckling rat pups. The treatment however decreased the trace metal contents in liver tissue. On the other hand, sodium molybdate supplementation has been found to provide significant protection from Pb uptake by blood, liver, and kidney and also to reduce the formation of lipid peroxidation radicals.
Experimental antioxidant therapy in laboratory animals was explored to understand the protection conferred by various antioxidants against Pb-induced hepatic oxidative stress. Ascorbic acid\textsuperscript{72}, vitamin E\textsuperscript{73}, N-acetylcysteine, lipoic acid\textsuperscript{74-76} and L-methionine\textsuperscript{77} either alone or in combinations were found to confer protection against Pb-induced oxidative stress and reduce liver Pb burden. This was associated with declines in liver drug metabolic enzymes and heme synthesis. Other studies\textsuperscript{77-79} have demonstrated that treatment with a combination of chelation and antioxidants resulted in a significant decline in tissue Pb burden and provided some protection against Pb-induced oxidative stress. One of the combination therapies using ascorbic acid and meso-DMSA did not confer protection compared to chelation therapy alone in suckling rats\textsuperscript{80}.

**Summary/conclusion**

Extensive \textit{in vivo} and \textit{in vitro} laboratory animal experimental evidence clearly points to potential hepatotoxicity resulting from exposure to Pb. These studies, ranging from simple biochemical and structural alterations to molecular characterization of hepatic hyperplasia or liver cell proliferation, have demonstrated pathologic changes indicative of liver toxicity. The complexity associated with the perturbations in various metabolic pathways (e.g., heme synthesis, cholesterol, drug metabolism), iron transport, and induction of mediators (e.g., cytokines, lipid perox radicals, Kupffer-cell derived mediators) all suggest an interplay of events in mediating the hepatic toxicity associated with Pb. The limited epidemiological data preclude extrapolating these animal toxicology observations to hepatotoxic effects in humans, and the potential contribution of such toxic effects to human morbidity is not clear. However, in developing countries such as India that have increased ambient levels of Pb, the addition of occupational exposures, nutritional (e.g., iron, protein) deficiencies, and infections may confound the overall impact of Pb on human health. Further, the mild to moderate dysfunction in hepatic drug metabolism associated with Pb toxicity (and its interactions with drug efficacy) may also have greater impact on general public health.

**Acknowledgment**

Author acknowledges the encouragement of Drs Lester Grant, and Mary Ross in the preparation of this manuscript and thank Drs Lori White and George Woodall for the critical review of the manuscript.

**References**


6. Degawa M, Arai H, Kubota M, Hashimoto Y. Ionic lead, but not other ionic metals (Ni\textsuperscript{2+}, Co\textsuperscript{2+} and Cd\textsuperscript{2+}), suppresses 2-methoxy-4-aminoazobenzene-mediated cytochrome P450IA2 (CYP1A2) induction in rat liver. \textit{Bioll Pharm Bull} 1995; 18 : 1215-8.


Heiman AS, Tonner LE. The acute effect of lead acetate on glucocorticoid regulation of tyrosine aminotransferase in hepatoma cells. Toxicology 1995; 100: 57-68.


Ercal N, Treeratpan P, Hammond TC, Matthews RH, Granennam NH, Spitz DR. \( \text{In vivo} \) indices of oxidative stress in lead-exposed C57BL/6 mice are reduced by treatment with \( \text{meso}-2,3\)-dimercaptosuccinic acid or \( \text{N-acetyl-L-cysteine} \). Free Radical Biol Med 1996; 21: 157-61.


Reprint requests: Dr Anuradha Mudipalli, National Center for Environmental Assessment-RTP Division, Mail Drop B243-02 United States Environmental Protection Agency, Research Triangle Park, NC 27709, USA e-mail: Mudipalli.anu@epa.gov