Reviews in Mineralogy & Geochemistry
Vol. 64, pp. 59-113, 2006

Metal Speciation and Its Role in Bioaccessibility and Bioavailability

Richard J. Reeder, Martin A. A. Schoonen

Department of Geosciences and Center for Environmental Molecular Science
Stony Brook University
Stony Brook, New York, 11794-2100, U.S.A.
e-mail: rjreeder@stonybrook.edu, martin.schoonen@stonybrook.edu

Antonio Lanzirotti

Consortium for Advanced Radiation Sources
University of Chicago
Chicago, Illinois, 60637, U.S.A.
e-mail: lanzirotti@bnl.gov

INTRODUCTION

Metals play important but varied roles in human health. Some metals are required for normal metabolic function, with optimal amounts for maximum benefit. Others are only known to cause toxic effects. Most of our knowledge of the function of metals in human health has been acquired in the last 100 years. However, evidence of adverse health effects attributed to metal exposures dates back to early civilizations. For example, it has been deduced that extensive mining and smelting of lead and its widespread use in the Roman Empire caused significant incidence of lead poisoning (Nriagu 1983; Hong et al. 1994).

The source of metals in the environment ultimately can be traced back to their occurrence primarily in rocks, with their release to soil, water, and air facilitated by weathering processes. Consequently, the natural occurrences of metals in soils and waters are strongly correlated to the varied distribution of rock types and the compositions of the constituent minerals. More than 2000 years ago the Greek physician Hippocrates recognized relationships between disease and location, illustrating that environmental factors influenced human health. Today there are many known geographic patterns of disease that have been correlated with properties of soils or waters, or even aerosol particles. It has been difficult to demonstrate cause-effect relationships for many correlations, and efforts to relate total concentration of a metal contaminant to toxic impact have proven difficult (Davies et al. 2005). This point illustrates the essential concept that the total amount of an element in an environmental setting is not necessarily a good measure of its potential health threat.

Within the last two centuries human activities have been highly effective in redistributing metals on local, regional, and even global scales. This has contributed to a greater exposure to humans. We have also changed the chemical forms of metals, sometimes with unfortunate consequences that include enhanced mobility in the environment as well as creation or enhancement of more toxic forms.

Nearly three-quarters of the elements in the periodic table are classified as metals. Inasmuch as all but a few of them occur in nature, it is probably correct to say that each one has (or will be found to have) an important role with regard to environmental health. In this paper, we focus primarily on a few of the so-called *heavy metals* that are known to be

1529-6466/06/0064-0003$10.00 DOI: 10.2138/rmg.2006.64.3

associated with adverse health effects. In a broad sense the heavy metals can be considered to include metals with atomic number greater than 20 (Ca), but many readers will know that a consistent definition has not found universal acceptance (Duffus 2002). Heavy metals should not necessarily be equated with toxicity, however, as a number of them are essential for human health in small amounts. Heavy metals have widespread occurrence as trace or minor constituents of soils, rocks, and waters, as well as living organisms (including humans). Commonly included among the heavy metals are arsenic, which is a metalloid, and selenium, a non-metal. Depending on factors such as oxidation state, electron configuration, ionic radius, and the presence of various ligands, metals exhibit a rich variety of coordination compounds in aqueous, solid, and even gaseous forms. The fact that the particular chemical form of a metal strongly influences its chemical behavior, mobility in the environment, uptake by organisms, and toxicity is certainly justification for characterizing and understanding metal speciation, not only in the environment but also in the body.

METAL SPECIATION CONCEPTS

Speciation refers to aspects of the chemical and physical form of an element. Oxidation state, stoichiometry, coordination (including the number and type of ligands), and physical state or association with other phases all contribute to define speciation. These properties govern the chemical behavior of elements, whether in environmental settings or in human organs, and play a crucial role in determining toxicity. The focus on metal speciation in this chapter reflects the varied roles it plays in human health. Metals such as iron and zinc are essential for metabolic function, but can be toxic in excess. Others, like cadmium and lead, have no known beneficial function and pose health risks even at low levels of exposure and uptake. The amount of exposure or uptake is obviously a key factor in assessing adverse health impacts, and defines the field of toxicology. However, the metal speciation is also a critical factor in determining toxicity. For example, inorganic dissolved mercury ($Hg^{2+}_{(aq)}$) and methyl mercury chloride ($CH_3HgCl_{(aq)}$) are both considered to be toxic, but the properties and behavior of the latter make it a significantly greater health threat (NRC 2000). Another example is illustrated by the two common oxidation states of chromium in soils and water. Hexavalent chromium in the form CrO_4^{2-} is soluble in water, making it mobile, and readily taken up by organisms. This form is also a known carcinogen (ATSDR 2000). Trivalent chromium tends to be insoluble, often forming hydroxide solids, and is considered an essential element in small amounts. We will see later that the pathways of uptake involve processes that may alter speciation, thereby changing a toxic form into a benign one, or the reverse.

Although the concept of speciation is now widely appreciated in many fields, there have been few efforts to provide a formal definition. Bernhard et al. (1986) pointed out that usage varies among different fields, ranging from evolutionary changes to distinctions based on chemical state. A Molecular Environmental Science Workshop convened by the U.S. Department of Energy in 1995 cited at least five aspects important for defining speciation: element identity, physical state, oxidation state, chemical formula, and detailed molecular structure (DOE 1995). Both reports emphasized the importance of techniques available for determining these properties and the limitations they place in our ability to identify and distinguish species.

Several Divisions within the International Union of Pure and Applied Chemistry (IUPAC) addressed speciation concepts in detail (Templeton et al. 2000), recommending that its usage in chemistry be restricted to distribution of chemically distinct species: “Chemical compounds that differ in isotopic composition, conformation, oxidation or electronic state, or in the nature of their complexed or covalently bonded substituents, can be regarded as distinct chemical species.” Although very similar to current usage in the Earth and environmental sciences, the IUPAC notion of speciation includes the isotopic identity of the element. Isotopic identify (i.e., mass differences within the nucleus) may have only a marginal effect on chemical

behavior, and then only when mass differences are large, such as with light isotopes. However, isotopic identity could also be significant in instances where particular isotopes have special characteristics, such as radioactivity.

The IUPAC concept of speciation does not draw attention to the significance of the physical state or association in distinguishing species. Earth and environmental scientists are particularly aware of the importance of physical associations. For example, occurrences of Cs^+ as an aquo ion in a soil water solution, sorbed at the surface of a mineral, or exchanged into an interlayer site in a smectite all represent distinct species, even though they may share the same oxidation state, coordination, ligands, and other local properties.

This last example also highlights the fact that multiple species of a metal commonly co-exist in real systems. Where different species of a given metal share some property, such as oxidation state, it can be difficult to distinguish among them. In fact our ability to characterize metal speciation is dependent on the techniques used and the information they provide. For example, the use of a technique that gives direct information regarding oxidation state may fail to distinguish metal species having the same oxidation state but are complexed by different ligands. Identification of species using a mass spectrometry method may fail to distinguish oxidation state, but also may require sample processing that could alter speciation. Such problems become amplified in complex solutions such as lung or gastric fluids, which contain numerous organic components, including peptides, amino acids, and phospholipids. Hence, species assessment is operationally defined, and no single technique provides information concerning all relevant aspects that define a species. Advantages can often be gained by use of several complementary techniques for species characterization. In view of the fact that many of the metals most relevant for human health occur at very low concentrations, adequate characterization may pose serious challenges. Later is this chapter, we consider some of the more commonly used techniques for assessment of metal speciation.

SIGNIFICANCE OF SPECIATION

The chemical and physical aspects that define speciation of a metal control its reactivity, including its solubility and uptake behavior, and, in many circumstances, toxicity. Solubility and uptake behavior, in turn, influence mobility of the metal in the environment, and therefore constrain pathways of exposure to organisms, including humans. During exposure the metal speciation directly influences absorption across a physiological membrane, which allows entry into systemic circulation. A transformation in speciation may occur in biological fluids (e.g., lung or gut fluids) prior to any absorption, however, which may affect absorption and subsequent toxicity. Within organ systems detoxification processes may further alter speciation and toxicity, and also influence transport, excretion, and storage. This oversimplified description illustrates the importance of metal speciation over the entire spectrum of process impacting the metal's fate from weathering to human impact. Readers are referred to Plumlee and Ziegler (2003) and Plumlee et al. (2006) for more a comprehensive discussion of these aspects.

The dependence of toxicity on speciation is now well known. The behavior of a metal may be completely changed by its oxidation state or its association with specific ligands, as exemplified by the contrasting toxicities of methylmercury and inorganic mercury species. The metalloid tin also shows markedly different health threats depending on its association with specific ligands. Neither metallic nor inorganic forms of tin present a health problem in small amounts; in fact, SnF_2 is a common additive of toothpaste. However, many organotin compounds, which are predominantly created by human industrial processes, are highly toxic (ATSDR 2005c). Tributylin tin, widely used as a biocide and antifouling agent for seagoing vessels since the 1970s (de Mora 1996), is a potent ecotoxicant (Alzieu 1996; Maguire 1996), persists in marine environments (Diez et al. 2002; Sudaryanto et al. 2004, 2005), accumulates

in tissue of fish and shellfish (Alzieu 1996; Sudaryanto et al. 2004), and may cause adverse health effects in humans (Kafer et al. 1992; Dopp et al. 2004).

One of the complicating aspects of speciation is that each species exhibits a distinct behavior, making generalizations about stability and reactivity difficult. In this chapter, we do not attempt to provide a comprehensive review of metal speciation. Instead, our approach is to illustrate important aspects of metal speciation on human health using selected examples.

As noted already, the total concentration of a particular element in any system, environmental or human, is not necessarily a good indicator of its potential health impact. Although this concept has been widely embraced by the research community and acknowledged by regulatory agencies, its impact on development of regulatory standards in the US has been limited. Even in the toxicology field, many bioassays do not consider speciation of metals. It is noteworthy, for example, that current methods for analysis of metals in soils (e.g., EPA 3050b) are designed to recover the more soluble metal fraction by use of acid digestion. As speciation techniques become more widely used and as understanding of the differences in behavior among species, including transformations, improves, it is likely that agencies will take greater account of these factors in formulating regulations. This is a critical area of research to which geochemists and mineralogist will be able to make important contributions.

ROLE OF METALS IN HUMAN HEALTH AND METAL TOXICITY CONCEPTS

The human body requires the uptake of several essential metals for its proper function. As briefly summarized in Appendix 1, a number of heavy metals are known to be essential. The roles of some of the essential heavy metals listed in Appendix 1, such as vanadium and tungsten, are not fully known. For others, including arsenic and tin, essential roles have been suggested, but not demonstrated. Some metals play a role in active centers of metalloenzymes. In fact, for metals such as cobalt this may represent the dominant "species" in the human body. Other metals, such as chromium(III) and vanadium, are metabolized within the body to form low molecular weight compounds that play a role in glucose metabolism.

The dose-response curve for an *essential* metal, schematically shown in Figure 1a, has a characteristic optimal range, flanked by suboptimal ranges. For some metals, a deficiency may be expressed as a specific disease. For example, chromium is important in the human metabolic system. A lack of chromium(III) disrupts glucose metabolism and can lead to obesity, diabetes, and cardiovascular disease, as well impairment of the reproductive system (Appendix 1).

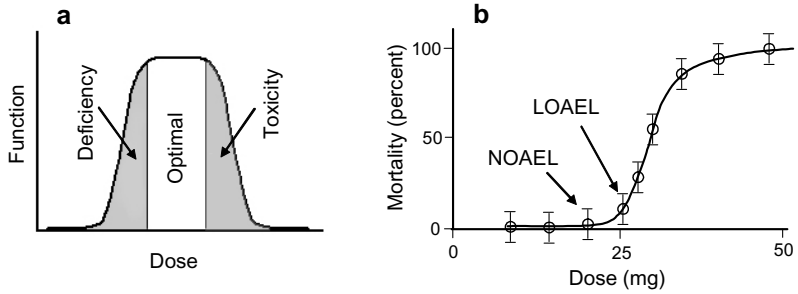

Figure 1. Schematic dose-response curves for (a) an essential metal and (b) a non-essential toxic metal. NOAEL and LOAEL are *no observed adverse effect level* and *lowest observed adverse effect level* (see Table 1).

It is important to note that deficiency in a metal may be caused by several factors. A diet lacking an essential metal is a common cause. For example, in China outbreaks of Keshan disease, a type of heart disease, are restricted to well defined geographic regions (Fordyce 2005). The occurrence of this disease is associated with a lack of dietary selenium. It is within this context that speciation is important. Crops grown on soil must be able to extract the selenium from the soils. Within Zhangjiakou District, Hebei Province, China, Keshan Disease has a high prevalence despite the fact that there is significant total selenium in the soil. In fact, a study in this region showed that the prevalence of the disease is not correlated with a lack of selenium in the soil as might be expected (Johnson et al. 2000). Rather, the cause for the selenium deficiency in the diet is a result of the fact that the soil-bound selenium is not in a form that is available to the plants. Soils in affected areas are rich in organic matter, and it is hypothesized that selenium is strongly bound with the organic fraction (Johnson et al. 2000). Alternatively, the organic matter may promote reduction of selenate to selenite, the latter commonly being more strongly sorbed to iron hydroxides in soils (Hartikainen 2005). This example illustrates the importance of speciation—in this instance in soils—rather than total concentration in understanding the incidence of a disease.

While geological factors have been shown to contribute to diseases, such as Keshan Disease, additional confounding factors can lead to complex patterns in the prevalence of a disease. The interaction between metals (or metalloids) is one of the confounding factors. Nutritional status is known to affect toxicity. For example, anemia—low iron status—promotes the uptake of nickel and manganese. Hence, individuals affected by anemia are at a higher risk for adverse effects of nickel and manganese uptake compared to healthy individuals exposed to the same nickel and manganese levels. Conversely, exposure to methylmercury has been shown to inhibit uptake of selenium, an antioxidant (Norling et al. 2004). A second major confounding factor is genetic predisposition. Genetic disorders that disrupt the uptake or transport of essential metals or formation of antioxidants are more difficult to diagnose and remedy. For example, Hallervorden-Spatz syndrome (HSS) is a neurodegenerative disease caused by a genetic defect that disrupts the function of ferritin, an iron storage protein. HSS leads to accumulation of iron in the brain and is thought to cause oxidative stress (Zhou et al. 2001).

Exposure and uptake of toxic metals, or even essential metals at levels in excess of the optimal range, can lead to adverse health effects. Exposure history is an important factor in evaluating the toxicity of a metal. Toxicologists distinguish between acute and chronic toxicity. Acute toxicity is that associated with short-term exposure to a toxicant, sometimes in lethal doses. Chronic toxicity is that associated with long-term exposure, and is usually the type most relevant to environmental toxicants. Studies based on rats or other laboratory animal models are commonly used to assess adverse health effects and to establish the *no observed adverse effect level* (NOAEL) and the *lowest observed adverse effect level* (LOAEL). A schematic dose-response curve for a toxic metal or substance is shown in Figure 1b. The outcomes of these studies form the basis for regulations. An overview of the methodology behind such studies is given by the U.S. EPA (*http://www.epa.gov*). Table 1 defines common terms used in such studies. Toxicity data for metals and other susbstances are available (on-line) from the Agency for Toxic Substance and Disease Registry (ATSDR) within the U.S. Department of Health and Human Services (*http://www.atsdr.cdc.gov*). For a web-based introduction to the basic concepts in toxicology and its terminology the reader is referred to the National Library of Medicine (National Institutes of Health) web page: *http://sis.nlm.nih.gov/enviro/toxtutor/Tox1/amenu.htm*.

BIOAVAILABILITY AND BIOACCESSIBILITY CONCEPTS

The term *bioavailability* is much used throughout the literature. There is a general understanding that the meaning addresses the potential for a substance to interact with an organism. With the use of this term now widespread among many disciplines, confusion

Table 1. Some abbreviations and terms relating to toxic effects of metals.

Abbreviation	Definition
NOAEL	No-observed-adverse-effect level. The actual doses (levels of exposure) used in studies that showed no observable adverse effects to the organism.
LOAEL	Lowest-observed-adverse-effect level; LOAELS have been classified into "less serious" or "serious" effects. "Serious" effects are those that evoke failure in a biological system and can lead to morbidity or mortality (e.g., acute respiratory distress or death). "Less serious" effects are those that are not expected to cause significant dysfunction or death, or those whose significance to the organism is not entirely clear.
MRL	Minimal Risk Level; an estimate of daily human exposure to a substance that is likely to be without an appreciable risk of adverse effects (noncarcinogenic) over a specified duration of exposure
CEL	Cancer effect level
LD_{50}	Lethal dose that leads to 50% mortality

Note: Other web resources that provide definitions of terms used in toxicology are *http://extoxnet.orst.edu/tibs/standard.htm* and *http://www.atsdr.cdc.gov/glossary.html*.

sometimes develops as specific meanings emerge from a particular context, discipline, or method of study. An example is the meaning of bioavailability shared by the toxicology and pharmacology fields, where it refers specifically to the fraction of an administered dose that is absorbed into the organism's circulatory system or into the organ where an effect occurs (Ruby et al. 1999; NRC 2003). The reason for such a restricted definition is clear upon consideration of the methodologies typically employed to evaluate efficiency of uptake of a drug or a toxicant. For example, a study might involve assays of blood levels of a given toxicant to identify peak plasma concentration and half-life resulting from specific oral dosages. Even this definition of bioavailability could find disfavor, since substances absorbed through the gastrointestinal tract of humans first circulate through the liver, where metabolism may limit the amount released to general systemic circulation.

This view of bioavailability is of little value to the soil geochemist examining the fraction of a metal in a soil that becomes soluble and mobile during a sequential extraction procedure. In fact, there may be no universally acceptable definition of bioavailability, a prospect that may be most evident where operational definitions are desired. The lack of a common view of bioavailability was addressed by a recent report from the National Research Council (2003) in the context of soil and sediment contaminants, and summarized by Ehlers and Luthy (2003). We refer the reader to this publication for a more detailed description of various definitions of bioavailability and the rationale for their use. Rather than propose a working definition, the NRC study recommended the adoption of a process-based view, with *bioavailability processes* being defined as "the individual physical, chemical, and biological interactions that determine the exposure of plants and animals to chemicals associated with soils and sediments," which is shown schematically in Figure 2.

While a process-oriented view will be intuitive for many geochemists, the NRC perspective is purposely limited on the organism side to exclude processes following transport of a substance across the biological membrane. Because of the context relating to soil and sediment contaminants, it places emphasis on those factors and processes that make the substance "available" to the organism, that is, in a form that can be transported across the organism's biological membrane. This is most commonly interpreted to be a soluble form. However, it is also possible that very small solids or colloidal particles could be transported across some

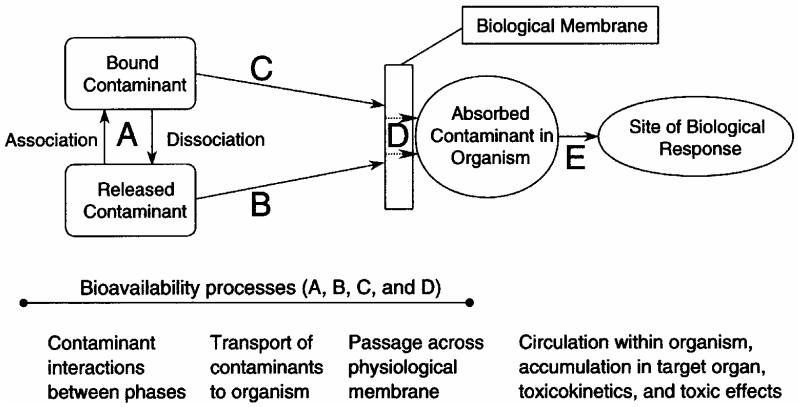

Figure 2. Schematic diagram illustrating the range of bioavailability processes as defined in the NRC study (2003). [Reprinted with permission from *Bioavailability of Contaminants in Soils and Sediments,* Copyright (2003) by The National Academies of Sciences, Courtesy of the National Academies Press, Washington, D.C.]

membranes, such as the linings of lungs. Ruby et al. (1996, 1999) used the term *bioaccessibility* to represent the fraction of a toxicant (or substance) that becomes soluble within the gut or lungs and therefore available for absorption through a membrane. The amount actually absorbed, which according to the view described above reflects bioavailability, may be less than the soluble fraction. This concept of bioaccessibility carries over to the release or solubilization of soil- and sediment-associated metals in environmental systems external to the organism. Selective sequential extraction procedures are familiar examples of protocols for solubilizing certain constituents in geomaterials (e.g., Tessier et al. 1979; Scheckel et al. 2003, 2005). This concept of bioaccessibility also applies to *in vitro* studies that assess the release (solubility) of solid-bound metals in simulated biological fluids, as models of human lung, gastric, or intestinal fluid. Such physiologically based extraction tests (PBET) are being evaluated as *in vitro* alternatives to more costly *in vivo* studies using animal models (Ruby et al. 1996, 1999; EPA 2005). Plumlee and Ziegler (2003) describe bioaccessibility tests and comparisons with bioavailability for a variety of earth materials. Similar *in vitro* approaches have been widely used in pharmacological studies to assess drug release (e.g., Lin and Lu 1997).

Agreement on the usage of bioavailability, bioaccessibility, and related terms seems unlikely in the near future, including within the environmental geochemistry and mineralogy fields. In response to the NRC report, Semple et al. (2004) suggested temporal and/or spatial distinctions between bioavailable and bioaccessible compounds. They proposed that the term bioavailable compound be used where the substance "is freely available to cross an organism's cellular membrane from the medium the organism inhabits at a given time." They proposed using bioaccessible to indicate a compound that "is available to cross an organism's cellular membrane from the environment, if the organism has access to the chemical," indicating that such a compound could subsequently become bioavailable once the proximity of the compound to the organism allows. Other authors have proposed the term *geoavailability* to represent the fraction of a toxicant or contaminant in a geologic material that becomes soluble or mobile as a result of various biogeochemical processes (Smith and Huyck 1999). This focus on terminology may appear to be only marginally relevant to the main topic. However, as Ehlers and Luthy (2003) note, bioavailability concepts may very well enter into risk assessment in the future and play an important regulatory role. Hence, we can expect further discussion of these concepts and related terminology, and it is incumbent on geoscientists to participate in this dialog.

In this chapter we refrain from recommending a preferred usage, and merely alert the reader to the potential for confusion. However, having noted that *bioaccessibility* can be generalized to include solubilization of metals in geomaterials, we tend to follow that usage here and retain the distinction with *bioavailability,* which we will use to refer to absorption across a physiological membrane. As emphasized in the following section, we believe there is merit in taking a broad view of the role of metal speciation, encompassing processes from weathering and transport in geomedia, through exposure and uptake by humans, to fate within organs.

PATHWAYS OF METAL UPTAKE— FROM SOIL, WATER, AND AIR TO HUMAN ORGANS

Historically, the gulf between the geosciences and the health sciences has tended to constrain the research activities in each community to address different parts of the broader subject of environmental health. Geoscientists have typically limited their attention to environmental processes that govern the bioaccessibility of metals up to and sometimes including exposure. Health scientists have naturally focused on the health impact, generally beginning with exposure. Clearly both communities recognize the continuum of processes that link the geologic and health aspects. In this chapter, the primary goal is to illustrate the role of metal speciation spanning both environmental and physiological processes.

General exposure pathways are described by Plumlee et al. (2006) elsewhere in this volume (also see Plumlee and Ziegler 2003). Exposure pathways for metals are the same as for other toxicants (*aka* xenobiotics): ingestion (gastrointestinal tract), inhalation (respiratory tract), and dermal contact. A particular metal may be present in a solid, a liquid, or a gas phase. The physical form commonly dictates the nature of the exposure. For example, arsenic that desorbs from iron oxide-coated sands in an aquifer will enter groundwater that may be a source of drinking water. Here, ingestion of dissolved arsenic is the main exposure pathway. Ingestion is also possible for metals in solid forms, including food and soil particles. Certain associations of a metal may also dictate exposure. For example, inhalation would be the primary exposure pathway for arsenic associated with airborne particles (e.g., in mineral dust). If a fraction of inhaled particles is transported by mucociliary action to the pharynx, then ingestion may become a secondary exposure pathway.

Environmental processes

Geochemists generally consider that metals are released to the human environment initially by weathering of rocks and are subsequently modified by various processes operating at or near Earth's surface, both natural and anthropogenic; these processes can enhance the environmental mobility of metals or lead to their sequestration. Human activities have been especially effective in redistributing metals and modifying their form, including speciation.

There are numerous physical, geochemical, and biologic processes that influence the behavior of metals in surficial settings. Much of the attention has focused on processes that mobilize or immobilize metals, since mobility generally facilitates exposure. A dissolved metal can be transported by fluid flow, eventually entering a water supply or being taken up within a food chain. In contrast, a metal that precipitates as a coating on mineral grains in a soil or aquifer is immobilized, and may be effectively eliminated from exposure unless remobilized by a subsequent process. In many circumstances, bioaccessibility in geomaterials is equated to the occurrence of a metal in a dissolved form. However, there are examples in which solid forms may have significant potential for exposure. Metals that are associated with small particles that are mobile may have greater potential for exposure. For example, colloids and airborne particles are mobile and both have been shown to have associated metals. Metals contained in the bottom sediments in streams, lakes, and marine settings may also enter the food chain if they are scavenged by bottom feeders or may even become remobilized upon interacting with gut juices.

The most important processes that control the mobility of metals in the environment include dissolution/precipitation, complexation with ligands, sorption/desorption by solids, biotransformation, uptake by soil and aquatic biota, and reduction/oxidation (redox) (Fig. 3). We provide only a brief overview of these processes here, as there are a number of existing sources that offer detailed reviews relating to metals (e.g., O'Day 1999; Traina and Laperche 1999; Brown and Parks 2001;Warren and Haack 2001; Sparks 2003).

Dissolution and precipitation involving metal species are subject to both thermodynamic and kinetic controls. Using the chemical analysis of a water as input, aqueous speciation programs, such as MINTEQA2 (U.S. EPA), PHREEQC (Parkhurst and Appelo 1999), and The Geochemists Workbench (*http://www.rockware.com*) (Bethke 2002), facilitate calculation of saturation states of the water with respect to a wide range of minerals and solids. However, there are many important environmental phases for which thermodynamic stability data are lacking, including many amorphous phases. Moreover, some of the phases controlling metal solubility in nature are solid solutions, and satisfactory solution models are available for relatively few environmentally relevant phases. Owing to a variety of possible kinetic factors, it is important to recognize that supersaturation does not guarantee that precipitation will occur. For example, inhibitors, often sorbed metals, may introduce kinetic constraints that limit precipitation (and dissolution). Plumlee et al. (2006) and Plumlee and Ziegler (2003) describe kinetic factors that influence the dissolution of asbestos and other mineral phases in human fluids.

Metal solubility is also strongly coupled to complexation in the aqueous phase. Metals that have a high affinity for either organic or inorganic ligands may exhibit significantly increased solubility through formation of complexes. The high affinity of UO_2^{2+} for dissolved CO_3^{2-} is an environmentally important example. In the absence of dissolved CO_3^{2-}, the total solubility of UO_2^{2+} controlled by equilibrium with the mineral schoepite ($UO_3 \cdot 2H_2O$) at neutral pH is 3.4 μM. With 1 mM total dissolved carbon dioxide in the solution the total UO_2^{2+} solubility is tenfold higher (56 μM) because of carbonate complexation.

Aqueous complexation may also influence the uptake of metals by aquatic and soil organisms. For example, dissolved Cu^{2+} and Ni^{2+} are readily taken up by some aquatic phytoplankton, whereas uptake is extremely limited when these same metals are strongly complexed with organic ligands or natural organic matter. Although many studies of aquatic organisms have shown correlations between metal uptake and the fraction of the metal occurring as a free ion

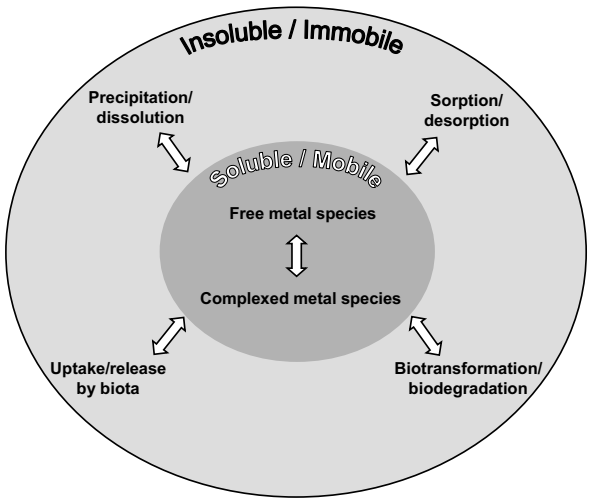

Figure 3. Dominant processes that control the mobility of metals in the environment. Oxidation/reduction may be associated with any of these processes, depending on the metal.

(i.e., the free-ion activity model)(Morel 1983; Campbell 1995), relatively little is known of the actual chemical form of the metal taken up (cf. Hudson 1998; Sunda and Huntsman 1998).

In systems that contain solid phases, sorption processes, involving transfer of dissolved metals to solid surfaces, represent some of the most important controls on dissolved metal concentrations (e.g., Brown et al. 1999; Sparks 2003). Sorption can be considered to include adsorption (i.e., accumulation of ions at the surface), surface precipitation (formation of a distinct phase at the surface), and co-precipitation (incorporation of ions into a phase, commonly as it precipitates). Sorption is generally most effective at removing a dissolved metal from solution when the metal is present at relatively low concentration and the available solid surface area is high. However, sorption also depends on several solution properties, including pH, ionic strength, and the presence of complexing ligands or competing species. Properties of the solid-liquid interface, including surface charge (Hochella and White 1990) and surface site coordination (Reeder 1996; Elzinga and Reeder 2002) are also important factors.

Generally, metal cations exhibit increasing adsorption as pH increases, as a response to decreasing proton charge on the surface. The change in adsorption efficiency typically occurs over a narrow pH range and is commonly referred to as the *adsorption edge.* The position of this edge with respect to pH may vary for different metals on the same sorbent, which generally reflects differing affinities of the metals for surface sites or different sorption mechanisms. For a given metal cation sorbing on different solid phases the pH range of the adsorption edge typically differs because of different surface charge properties and different surface sites. Adsorption of anions is usually greatest at low pH and decreases with increasing pH, also reflecting electrostatic properties of the surface. Anion species of acids may exhibit sorption maxima in their pH dependence, usually coinciding with their pK_a values and demonstrating differences in sorption behavior among the more and less protonated species. Metal cation sorption may also be influenced by complexation, which may vary with pH and ligand concentration. Uranium again provides a good example. U(VI), occurring as the UO_2^{2+} aqueous species, shows a rapid increase in adsorption on ferrihydrite with increasing pH (in the pH range 3.5-5.5), which is typical behavior for a cation (Fig. 4). However, at higher pH (7.5-9.5), U(VI) adsorption decreases abruptly as a result of the formation of uranyl carbonate anion complexes having low affinity for the surface (Waite et al. 1994). Increasing the dissolved carbonate concentration causes the edge at higher pH to shift to lower pH values due to increased carbonate complexation, thereby resulting in a narrower pH range of maximum adsorption.

Spectroscopic investigations have demonstrated that different types of surface complexes may form, including inner-sphere and outer-sphere types (Fig. 5). Although multiple factors may be important, inner-sphere surface complexes are generally more strongly bound to surfaces than outer-sphere complexes, and therefore may be less susceptible to desorption, or release to solution. For more information, readers are referred to the excellent reviews of sorption by Anderson and Rubin (1981), Davis and Kent (1990), Stumm (1992), Sparks (2003), and Sposito (1984, 2004).

Biotransformations commonly involve reduction/oxidation (redox) processes. Because large solubility differences sometimes exist between different oxidation states of a metal, bacterially mediated reduction or oxidation may be highly effective in controlling metal concentrations in environmental solutions. Uranium again provides a useful illustration. Over the environmentally relevant pH range, U(IV) is relatively insoluble, often forming oxide phases such as uraninite, UO_2. Uranium(VI) is relatively soluble and is generally considered the mobile form of uranium. Bacteria present in soil systems have been shown to reduce dissolved U(VI) to U(IV), resulting in the formation of uraninite (e.g., Lovley et al. 1991; Fredrickson et al. 2000). This process may effectively immobilize uranium in the subsurface. However, in oxidizing conditions, uraninite may become re-oxidized, resulting in its remobilization as U(VI) (e.g., Senko et al. 2002).

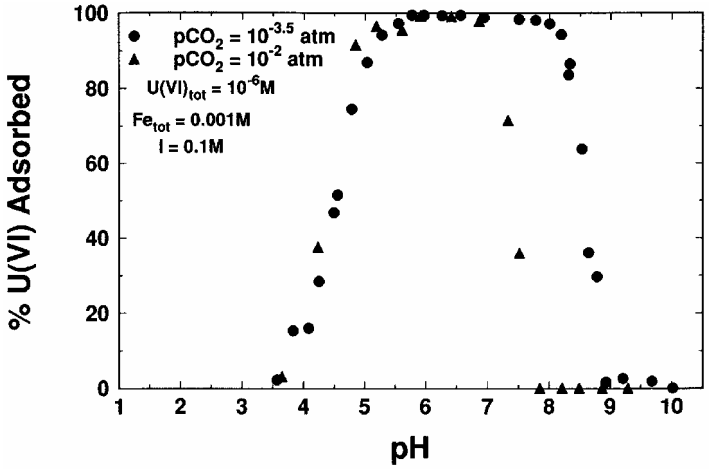

Figure 4. Uranium(VI) adsorption behavior on ferrihydrite as a function of pH and at different total CO_2 concentrations. The decrease in adsorption above pH 7.5-8 is attributed to formation of aqueous uranyl carbonate complexes that have low affinity for sorption. [Reprinted with permission from Waite et al., *Geochimica et Cosmochimica Acta*, Vol. 58, Fig. 6, p. 5470. Copyright (1994) Elsevier.]

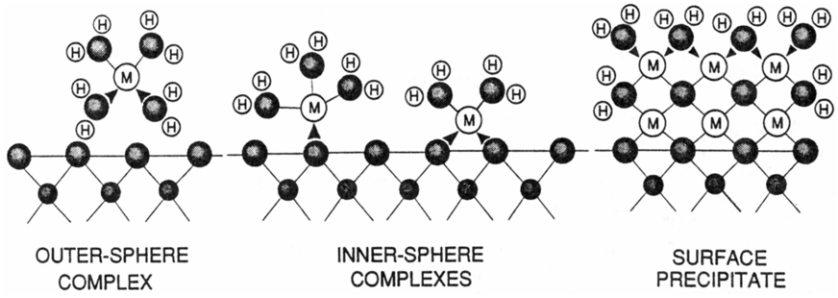

Figure 5. Diagrams showing outer- and inner-sphere adsorption complexes and a surface precipitate. [Reproduced from Brown (1990).]

Redox processes may also occur without biological mediation, however, kinetics are commonly sluggish. In surface environments important electron donors/acceptors include organic matter and compounds containing iron, manganese, and sulfur. Many redox processes occur at surfaces of solids and are associated with sorption processes. Adsorption of metals onto surfaces can change their electronic structure and promote redox reactions that are inhibited when the metal is dissolved. For example, it has been demonstrated that reduction of Cr(VI) co-adsorbed with Fe(II) onto an Fe(III)-oxide mineral substrate is significantly faster compared to the homogenous reaction between these two metal species (Buerge and Hug 1999). The same phenomenon has been observed for the reaction between Fe(II) and uranyl (Liger et al. 1999). While the effect of sorption on the kinetics of electron transfer reactions of transition metals in aquatic systems has received some attention, this phenomenon is likely to also play a role within the human body where transition metals may be ingested or inhaled as adsorbed species on mineral dust (Schoonen et al. 2006). Further information on electron transfer reactions in the environment is provided in reviews by Bartlett and James (1993), Sparks (2003), and Schoonen and Strongin (2005).

Because the processes described above are the dominant controls on metal mobility in the near-surface environment, they are also among the most important factors governing the environmental exposure of humans to metals. Next we consider the processes associated with metals following exposure.

Internal processes

After ingestion, inhalation, or dermal contact, the fate and impact of a metal are affected by absorption, distribution, metabolism (or biotransformation), and elimination. Collectively, these processes, commonly referred to as ADME, define the field of *toxicokinetics* (Guidotti 2005). The behavior of metals throughout these internal processes is strongly dependent on speciation. Moreover, metal speciation typically changes during these processes. The health effect of a metal toxicant, including the mechanism of toxicity, is also dependent on speciation and constitutes the field of *toxicodynamics*. We do not discuss toxicodynamics, except to note a few well known examples. Moreover, the level of material that we present here is very basic, and interested readers are encouraged to consult more comprehensive reviews. A useful starting point for geoscientists is the Environmental Health and Toxicology web page of the National Library of Medicine (National Institutes of Health) (*http://sis.nlm.nih.gov/enviro.html*).

Absorption involves transport of the metal across a physiological membrane (e.g., commonly a phospholipid bi-layer). This may occur via different mechanisms (e.g., passive, facilitated, or active transport) depending on the substance and its chemical form as well as the cell type (Dawson and Ballatori 1995; Foulkes 2000). Absorption of Cr(VI), for example, occurs via facilitated transport, following the sulfate and phosphate pathway, whereas Cr(III) transport is primarily by passive diffusion and much less efficient. Methylmercury has a significantly greater absorption efficiency than inorganic ionic mercury, which is mainly attributable to the higher lipid solubility of the methylated form. Some of the properties of metals and other substances that influence absorption are listed in Table 2. Metal speciation may be altered before absorption occurs, for example in the presence of gastric or lung fluid, or in mucus. Foulkes (2000) has emphasized that, because of their high affinity for complexing with proteins and other biological molecules in internal fluids, most non-essential heavy metals are

Table 2. Properties of a metal or substance that influence absorption at physiological membranes.

Property	Effect
Concentration/dose	Fractional absorption may vary with concentration, including inversely.
Molecule size and charge	Dependent on transport mechanism (passive, facilitated, or active). For passive diffusion, neutral charge and small size favor absorption.
Competing antagonists	Competition depending on transport mechanism.
pK_a of acids	Nonionized form of some acids more readily absorbed.
Lipid solubility	Lipid-soluble species more readily absorbed.
Particle size/surface area (solid)	Smaller particles solubilized more rapidly.
Phase identity (solid)	Relates to solubility and association of metal.
Solubility (solid)	More soluble solids generally allow greater absorption.
Sorption state (solid)	Metal sorbed on solid surface more readily released.
Matrix components (solid)	Other components in solid may enhance/reduce absorption.

transported across membranes as complexes, rather than as free metals. Transformations may also include changes in metal oxidation state. An example is the partial reduction of Cr(VI) to Cr(III), which begins in the saliva and gastric fluid and influences absorption. Absorption may be influenced by a number of factors, including the fed state. For example, in rats orally ingested Cr(VI) is absorbed more readily in the fasted than in the fed state (O'Flaherty 1996).

After absorption, a metal undergoes distribution and metabolism (also known as biotransformation). Some metals are distributed to all or most tissues and organs (e.g., arsenic), whereas others concentrate in specific organs, which may not be the target organs. Lead, for example concentrates in bone, yet its primary toxicity is in brain function. Both transport and storage depend on chemical form. Methylmercury, for example, readily crosses the blood-brain barrier, whereas inorganic mercury (Hg^{2+}) does not. Metal transport in the blood commonly involves an equilibrium partitioning between plasma and proteins, with some metal species preferentially entering red blood cells. More than 90% of the methylmercury in the blood enters red blood cells and binds with hemoglobin (NRC 2000). Metals absorbed from the gastrointestinal tract do not pass directly into general systemic circulation, but first enter portal circulation where some toxicants are metabolized by the liver.

Metabolism encompasses all of the biotransformations that modify the form (i.e., speciation) of a toxicant. These are typically enzymatic processes occurring with multiple steps and intermediate metabolites. Metabolites may be more or less biochemically active than the original substance. Generally metabolism serves to detoxify a toxicant, often by creating a chemical form that is more readily eliminated. For example, the methylation of arsenic in the liver facilitates its elimination and is considered a detoxification mechanism.

Elimination of metals occurs primarily through urine, feces, and by exhalation. Water soluble forms are excreted readily in urine. The liver secretes some metals (e.g., lead and mercury) into bile, which are then eliminated with feces or remain in enterohepatic circulation (Guidotti 2005). Some metals are stored in various tissues, and may provide evidence of exposure over time. For example, analysis of hair serves as a method for assessing chronic exposure to mercury. The primary storage site for lead is in bone and its analysis serves as a cumulative biomarker for exposure.

These ADME processes are usually unique to the particular species of metal, and clearly involve complex processes. Quantitative modeling of these processes is a major focus in the toxicokinetics field. Such *physiologically based pharmacokinetic* (PBPK) models have been developed for a number of metal toxicants, and are being improved as new data permit (O'Flaherty 1998). Inasmuch as quantitative modeling of environmental processes for metals is also well advanced, it may not be long before comprehensive models are developed that address both environmental and internal fate of metals.

The range of environmental and internal processes that we have described is shown conceptually in Figure 6. In the following section we illustrate the ways in which speciation influences bioaccessibility and bioavailability through four examples of metals/metalloids: arsenic, chromium, lead, and mercury.

ROLE OF METAL SPECIATION: ARSENIC

Arsenic has become the focus of worldwide concern following the recognition of its widespread and devastating health impact in many developing regions, most notably Bangladesh and West Bengal. The occurrence of arsenic in soils and aquatic systems, including the sediments and groundwater in the Bengal Basin region, and the geochemical processes that influence arsenic mobility have been reviewed recently by Plant et al. (2005) and Smedley and Kinniburgh (2002, 2005). Recent reviews of arsenic toxicity and human health effects

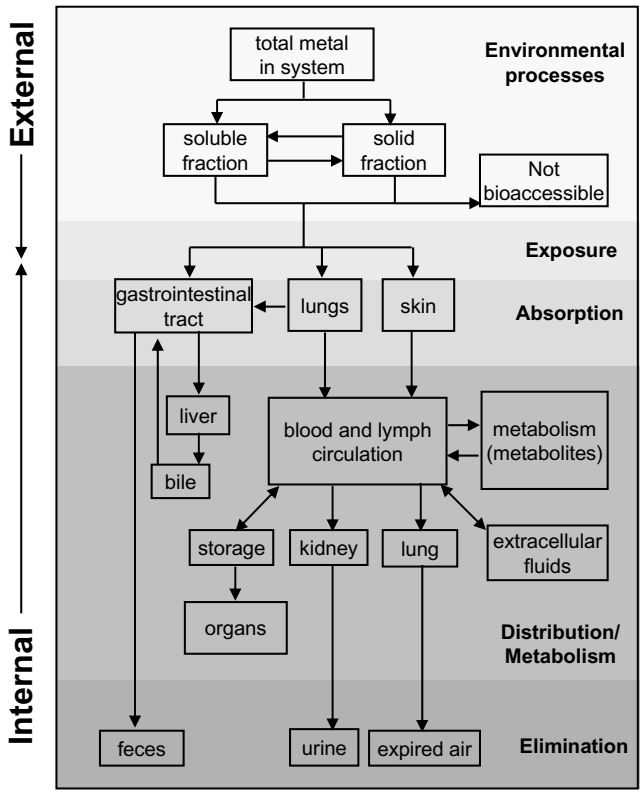

Figure 6. Idealized diagram illustrating the range of pathways and processes affecting the environmental and physiological behavior of metals. [Adapted from the National Library of Medicine (*http://www.sis.nlm.nih.gov/enviro/toxtutor/Tox2/a11.htm*).]

are given by NRC (1999, 2001), IPCS (2001), and ATSDR (2005a). Here, we provide a brief overview of the role of arsenic speciation as it relates to mobility in the environment, human exposure, bioavailability, and health effects.

Arsenic in the environment

Arsenic is not an abundant element in Earth's crust, with an average concentration in crustal rocks of ~2 ppm (Wedepohl 1995). However, it has a strong association with sulfide-bearing mineral deposits and especially with pyrite (FeS_2), which is widespread. Arsenic also exhibits an association with iron oxide and hydroxide minerals, which are abundant as weathering products. Common sources of arsenic in the environment are listed in Table 3. Arsenic occurs in the 3–, 1–, 0, 3+ and 5+ oxidation states, but in nature 1–, 3+, and 5+ oxidation states dominate. Arsenide minerals and sulfides in which As(I–) is a common substituent typically exhibit low solubilities in reducing natural waters. However, oxidative weathering results in the formation of arsenite [As(III)] and arsenate [As(V)] species; these occur as oxyanions or neutral species and may be quite soluble depending on pH and other solution properties. In solution, redox potential (Eh) and pH are the dominant controls on As speciation. An Eh-pH stability diagram is shown in Figure 7. Coexistence of As(III) and As(V) species in natural waters has been interpreted to indicate that electron transfer occurs, if not redox equilibrium. However, rates of electron transfer may be slow and strongly influenced

Table 3. Common sources of arsenic in the environment.

• Volcanic emissions and hot springs
• Dissolved in groundwater from interaction with rock (mobilization from igneous and sedimentary sources, oxidative dissolution of arsenic-bearing sulfide minerals)
• Mining waste, pH-mediated mine effluents, and tailings ponds
• Arsenic-containing pesticides (sodium arsenite or lead arsenate)
• Organic arsenic compounds as herbicides: monosodium methanoarsonate (MSMA), disodium methanoarsonate (DSMA), arsenic acid, and dimethylarsenic acid
• Waste from industrial metal smelting processes
• Leaching of wood preservatives: chromated copper arsenate (CCA) and ammoniacal copper arsenate
• Combustion of fossil fuels in electrical power plants
• Arsenic dusts and gases released during cement manufacture
• Animal waste management from feed additives in poultry (roxarsone to control coccidiosis and promote growth)
• Arsenic trioxide waste from glass manufacturing process (pre-1970)

by bacterial activity (e.g., Cullen and Reimer 1989; Smedley and Kinniburgh 2005). The acid dissociation constants (K_a), expressed here as pK_a values, for H_3AsO_4 are 2.22, 6.98, and 11.53. Below pH ~7 the dominant As(V) species is $H_2AsO_4^-$ and above pH ~7 $HAsO_4^{2-}$ is dominant, except at very low or high pH (Fig. 8). The pK_a values for H_3AsO_3 are 9.23, 12.13, and 13.4, so that $H_3AsO_3^0$ is the dominant dissolved inorganic As(III) species in most natural waters.

In view of its affinity for sulfur, As(III) may form thio-complexes (cf. Wilkin et al. 2003). Methylation may also occur in the environment as a result of microbial processes, resulting in formation of several As(III) and As(V) methyl species, including monomethylarsonous acid [MMA(III)], dimethylarsinous acid [DMA(III)], monomethylarsinous acid [MMA(V)], and dimethylarsinic acid [DMA(V)] (Fig. 9). Anderson and Bruland (1991) reported the formation of dimethylarsenate [DMA(V)], $(CH_3)_2AsO_2^-$, on a seasonal basis to become the dominant dissolved As species in a freshwater reservoir, followed by breakdown to arsenate. Bright et al. (1996) also reported methylated arsenic species in lake sediment pore waters, with formation presumed to be associated with bacterial activity. Other organic species include arsenobetaine and arsenocholine, which are forms commonly present in food. In the majority of natural waters, however, inorganic arsenic forms are most abundant (Smedley and Kinniburgh 2002).

Human exposure to arsenic is primarily through ingestion of water and food, although airborne particulates containing arsenic may be locally important. For this example, we focus on exposure through drinking water and to a lesser extent on ingestion of arsenic-containing solids. Consequently, solubility of arsenic is the main geochemical factor influencing exposure in this circumstance. Additionally, arsenic associated with particles that are mobile could also contribute to exposure. As noted earlier, numerous geochemical processes control metal solubility and mobility, including mineral precipitation/dissolution, sorption, and biotransformation. Metal speciation is a critical aspect of all these processes.

Arsenic sorption. Because dissolved arsenic concentrations are typically very low, even in contaminated systems, sorption processes can be highly effective in limiting As concentration and thereby controlling mobility and bioaccessibility. A number of studies have demonstrated that As(III) and As(V) species may sorb strongly with oxide and hydroxide minerals, especially iron, aluminum, and manganese oxyhydroxides. Sorption is strongly dependent on pH and

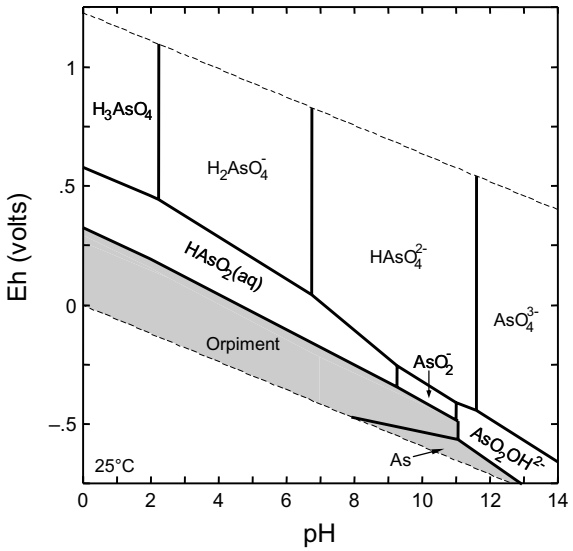

Figure 7. Eh-pH predominance diagram for arsenic in the presence of sulfur at 25 °C (As_{tot} = 10^{-6} M; S_{tot} = 10^{-4} M).

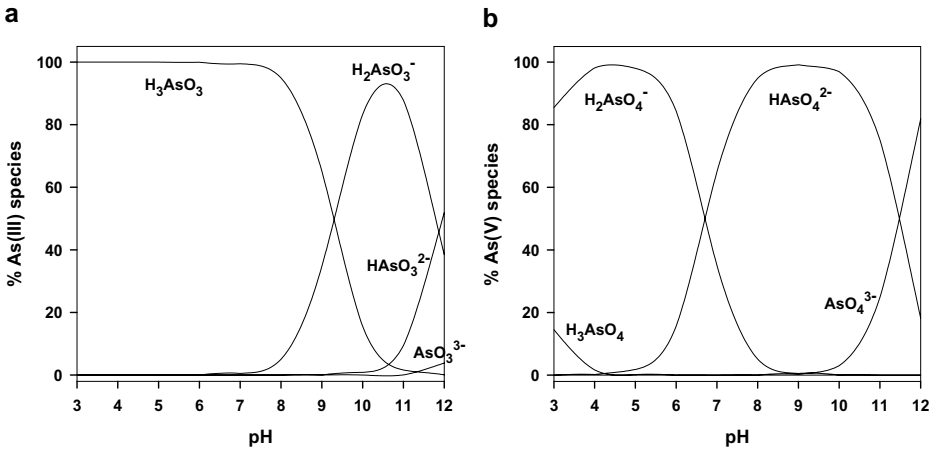

Figure 8. Aqueous speciation diagrams for (a) As(III) and (b) As(V) systems at 25 °C (As_{tot} = 10 ppb, and 1 mM NaCl).

other solution properties. Arsenite and arsenate exhibit very different sorption behaviors as a function of pH, and in some systems sorption is sensitive to ionic strength. As an example, Arai et al. (2001) compared As(III) and As(V) sorption on γ-alumina over the pH range 3-10 (Fig. 10). Arsenate sorption increases with decreasing pH, over the range from pH 9.5 (near the point of zero charge, PZC) to 4.5. This behavior is typical for sorption of anion species (e.g., Hingston 1981). Arsenate sorption is not observed to be sensitive to ionic strength; this has been interpreted to be suggestive of formation of inner-sphere surface complexes, which was confirmed using EXAFS spectroscopy (Arai et al. 2001).

Arsenite exhibits less overall sorption than arsenate on γ-alumina, with a broad maximum at pH ~8.5 (see also Goldberg 2002). A decrease in arsenite sorption is observed above pH 9 and likely reflects the change in dominant aqueous speciation to an anion, $H_2AsO_3^{1-}$. Over

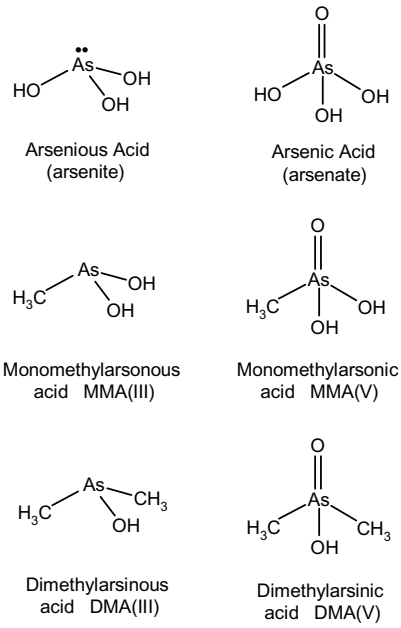

Figure 9. Common inorganic and methylated As(III) and As(V) species. [Adapted from O'Day (2006).]

the pH range 5-9, arsenite sorption is decreased at higher ionic strength, which is suggestive of outer-sphere surface complexation. *In situ* EXAFS spectroscopy showed that inner-sphere As(III) complexes dominate at pH < 5.5 and a mixture of inner- and outer-sphere complexes exists at pH > 5.5 (Arai et al. 2001). Several studies have shown that arsenate and phosphate compete for sorption sites, owing to similar chemical behaviors (e.g., Jain and Loeppert 2000). Consequently arsenate sorption may be significantly depressed where phosphate is elevated.

There are numerous studies of As(III) and As(V) sorption on other mineral and oxide surfaces that readers should consult for further insight (e.g., Fuller et al. 1993; Waychunas et al. 1993; Goldberg 2002; Smedley and Kinniburgh 2002; Stollenwerk 2003).

Arsenic redox behavior. Much of the arsenic present in surface environments has been derived from the oxidative weathering of reduced species in sulfide mineral deposits and primary rocks (Smedley and Kinniburgh 2002). Because of the relatively low solubility of reduced arsenic, much of the interest in redox behavior, from the perspective of health impact,

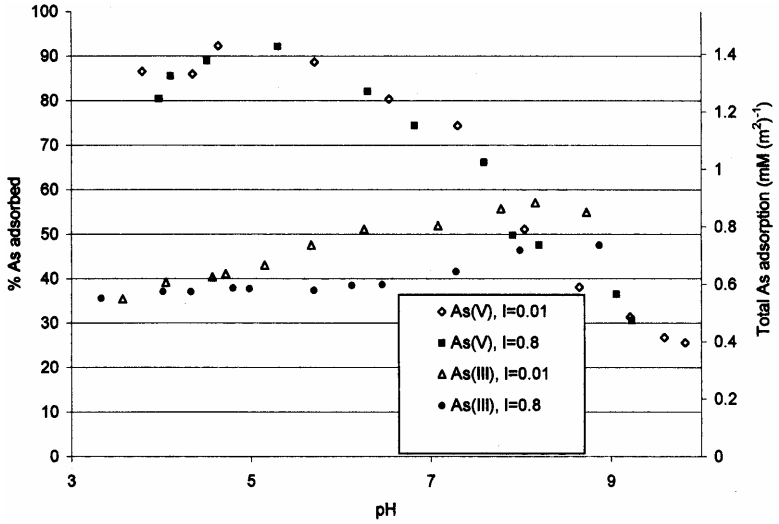

Figure 10. The pH and ionic strength dependence of As(III) and As(V) sorption on γ-alumina. [Reprinted with permission from Arai et al., *Journal of Colloid and Interface Science*, Vol. 253, Fig. 1, p. 83. Copyright (2001) Elsevier.]

has focused on transformations between As(III) and As(V) species. Both oxidation states are common in soils and near-surface waters, and it is not uncommon to have both As(III) and As(V) species co-existing in solutions and in solids (Hering and Kneebone 2002). The Eh-pH diagram shows that the predominance fields for species of both oxidation states coincide with Eh-pH conditions typical of many environmentally important settings (Fig. 7), so that redox transformations can be expected. However, in most cases As(III)/As(V) redox behavior is controlled by interaction with minerals, microbes, or organic matter that serve as electron donors or accepters. As is often the case in systems containing multiple redox couples, the As(III)/As(V) couple is commonly not in equilibrium with other redox couples (Spliethoff et al. 1995; Hering and Kneebone 2002). This reflects widely differing kinetics of electron transfer and strong dependence on mechanism. Hence As(III) species may persist in oxidizing systems and As(V) may persist in reducing conditions (cf. Inskeep et al. 2002).

Important oxidants for As(III) include Mn(IV) oxides (e.g., birnessite), titanium oxides, H_2O_2, and possibly dissolved ferric species (Foster et al. 1998; Manning et al. 2002; Voegelin and Hug 2003). Photochemical oxidation may also be important (Inskeep et al. 2002). Dissolved oxygen has been shown not to be an effective oxidant, except at high pH (e.g., Manning and Goldberg 1997). Dissolved sulfide has been shown to reduce As(V), with formation of intermediate sulfide complexes (Rochette et al. 2000). Many of the important redox processes occur at mineral surfaces and are associated with sorption. As noted above, arsenate is often associated with iron and aluminum hydroxides, and is typically more strongly sorbed by these phases than arsenite at circum-neutral and acidic pH conditions. An important mechanism of release of sorbed As(V) involves its reduction to As(III). This may occur either by arsenate reduction at the surface with subsequent As(III) release to solution, or by reductive dissolution of a ferric hydroxide sorbent followed by reduction of arsenate to arsenite as shown in Figure 11 (Inskeep et al. 2002). Reductive dissolution of iron (hydr)oxides (with sorbed arsenate) is thought to be a factor causing the elevated dissolved arsenic concentrations in groundwater in Bangladesh (e.g., Smedley and Kinniburgh 2002).

Microbial activity has been shown to cause both reduction and oxidation of arsenic species. We do not describe these reactions here, but refer readers to the overview of Inskeep et al. (2002).

Arsenic precipitation/dissolution. Precipitation and dissolution may also provide important constraints on As solubility. In many aquatic systems, however, arsenic concentrations

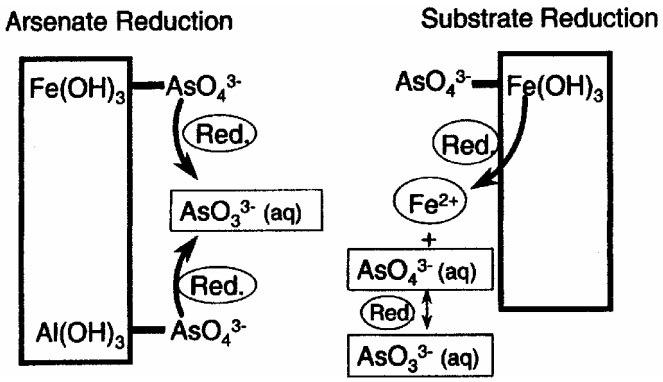

Figure 11. Schematic diagram showing reductive release mechanisms for As(III). [From Inskeep et al. (2002), *Environmental Chemistry of Arsenic*. p. 191, Fig. 4. Reproduced by permission of Routledge/ Taylor & Francis Group LLC. Copyright (2002).]

remain very low so that supersaturation with respect to arsenic oxides/hydroxides or to arsenite or arsenate salts is uncommon. However, high concentrations of cations may allow supersaturation and precipitation. Some remediation strategies rely on precipitation to remove arsenic from solution, such as wastewater. Additives such as lime, fly ash, Portland cement, and ferrihydrite have been used to induced formation of arsenic phases (e.g., Moon et al. 2004). Calcium arsenite and arsenate phases have received some attention as possible precipitates for immobilizing arsenic (e.g., Bothe and Brown 1999). However, the effectiveness of precipitation as a remediation strategy depends on the solubility of the solid. Overviews of important arsenic-containing phases and minerals are given by Cullen and Reimer (1989), Nordstrom and Archer (2003), and O'Day (2006). Foster (2003) describes arsenic speciation in a number of solid phases, including that for arsenic bound with oxides and hydroxides. Arsenic mineral solubilities vary widely according to arsenic oxidation state, composition, crystal structure. The total dissolved arsenic concentration and the concentrations of individual arsenic species in equilibrium with different arsenic-containing phases depend on the overall solution composition, including pH, ionic strength, T, and P. Aqueous speciation programs facilitate such calculations and provide an equilibrium distribution of aqueous species. Nordstrom and Archer (2003) have critically evaluated the thermochemical data available for selected arsenic phases and aqueous species. In general, As(III) and As(V) oxides are moderately soluble in most aqueous solutions. As_2O_3 occurs as arsenolite and claudetite, whereas As_2O_5 is not known as a mineral. Arsenic sulfides, such as realgar and orpiment, tend to be relatively insoluble in most reduced solutions, as are metal arsenides and arsenic sulfosalts. Metal arsenates and arsenites exhibit a range of solubilities depending on the metal involved and other factors (cf. Sadiq 1997). As described below, solubility is important in controlling the bioavailability of arsenic-containing solids following ingestion or inhalation.

Arsenic coprecipitation. Arsenic may also coprecipitate with mineral phases that are forming, thereby providing an effective mechanism for removing arsenic from solution. For example, coprecipitation with ferrihydrite has been shown to significantly reduce dissolved arsenate (e.g., Fuller et al. 1993; Richmond et al. 2004). The similarity of AsO_4^{3-} to other tetrahedral oxoanions, such as PO_4^{3-} and SO_4^{2-}, should allow As(V) substitution (e.g., Myneni et al. 1997; Foster 2003). In addition As commonly substitutes for S in pyrite and other sulfides.

Arsenic in the body

Arsenic absorption. Once ingested, dissolved As(III) and As(V) are both readily absorbed in the human gastrointestinal tract. Studies that systematically compare the relative absorption efficiencies of dissolved As(III) and As(V) in humans from oral exposure are lacking. However, an NRC review cited studies of humans and animals indicating 80-90% absorption for dissolved As(III) and As(V) doses (NRC 1999). MMA(V) and DMA(V) are also readily absorbed in the gastrointestinal tract of humans (ATSDR 2005a), as is arsenobetaine from ingestion of fish (IPCS 2001).

Arsenic present in or associated with solid forms generally shows lower absorption efficiency than dissolved arsenite or arsenate, especially for phases exhibiting low solubility. There are few systematic studies of gastrointestinal absorption (oral bioavailability) in humans for different As compounds. Studies in animal models have been summarized by ATSDR (2005a) and IPCS (2001). Although significant differences exist between different animals, solubility of the arsenic source is the most significant factor influencing absorption. For example, gastrointestinal absorption in rodents is low for relatively insoluble GaAs (<22%) compared to more soluble sodium arsenite and sodium arsenate (80-90%) (Yamauchi et al. 1986; IPCS 2001).

Gastrointestinal absorption from arsenic-containing soils and sediments has also been studied using animal models. The summary given by IPCS (2001) shows great variation depending on the animal model and the characteristics of the soil (not surprisingly). A series of

recent studies sponsored by the U.S. EPA (Region 8) have examined the relative bioavailability of arsenic in various soils, sediments, and mine waste material using a juvenile swine model (*http://www.epa.gov/region8/r8risk/hh_rba.html*). These *in vivo* studies compared absorption of arsenic in different solid forms relative to that of sodium arsenate, which is expected to be completely soluble in gastric fluid. A summary of this work (EPA 2005) reports that relative bioavailabilities range from 10 to 60% for the substances examined. Lowest bioavailabilities were observed when arsenic was present as As_2O_3 or in reduced forms, such as arsenides or As-containing sulfides. Higher bioavailability was observed for As-sorbed onto metal oxyhydroxides, such as FeOOH. Owing to the complex and heterogeneous nature of the test materials used in these studies the speciation of the arsenic was not fully characterized. Nevertheless, the wide range of relative bioavailabilities observed underscores the importance of speciation. Furthermore, the task of characterizing the speciation in such materials represents a challenge for geochemists and mineralogists.

Arsenic metabolism. The most important metabolic pathway of inorganic arsenic in humans involves methylation. The liver is the most important site of methylation. The likely *in vivo* mechanisms of methylation involve reduction of As(V) to As(III) and oxidative methyl transfer to produce monomethylarsonic acid [MMA(V)] and dimethylarsinic acid [DMA(V)] as illustrated in Figure 12. Methylation is enzymatically controlled, and S-adenosylmethionine is the primary methyl donor (Zakharyan et al. 1995). Glutathione is a likely electron donor for the reduction steps (Scott et al. 1993). The process does not go to completion as indicated by the presence of As(III), MMA(V), and DMA(V) in human urine (e.g., Le 2002), which serve as important biomarkers for arsenic exposure. Methylation has been found to be variable among individuals, by gender, and according to diet (Vahter 2000). Methylation is highly variable among other mammals (Vahter 2002).

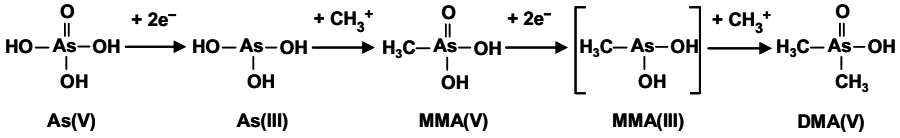

Figure 12. Summary of the human arsenic methylation process involving reduction and oxidative addition of methyl groups. [Reproduced with permission from *Environmental Health Perspectives*, Le XC et al. (2000).]

Methylation has long been considered a detoxification mechanism for arsenic, as MMA(V) and DMA(V) are less reactive and less toxic than inorganic arsenic (Styblo et al. 2000; Vahter 2002), and methylation facilitates elimination through urine and decreases retention (NRC 1999). However, several recent studies have noted the release and persistence of the intermediate metabolite MMA(III), which is highly reactive and possibly more toxic than inorganic As(III) (Styblo et al. 2000; NRC 2001). Therefore, methylation should not be considered solely as a detoxification process (NRC 2001).

Arsenic elimination. The principal pathway by which arsenic is eliminated is through the urine. Studies have shown that approximately 33-38% of an ingested arsenic dose is eliminated in the urine within 48 h and 45-58% within 4-5 days (Tam et al. 1979; Buchet et al. 1981). Elimination of ingested As(V) is slightly more rapid than for As(III) (Pomroy et al. 1980). MMA(V) and DMA(V) are eliminated more rapidly: 75-78% within 4 days (Buchet et al. 1981).

Arsenic toxicity. Mechanisms of arsenic toxicity are still not well understood, and a description of proposed models is beyond the scope of this chapter. Excellent summaries of

the vast spectrum of adverse health effects associated with arsenic exposure are provided by the NRC reviews (1999; 2001), the Agency for Toxic Substances and Disease Registry (2005a) within the U.S. Center for Disease Control (*http://www.atsdr.cdc.gov*), and the IPCS (2001), sponsored by the World Health Organization (*http://www.inchem.org/documents/ehc/ehc/ehc224.htm*). At least one contributing reason for the variety of health effects associated with arsenic is the fact that arsenic is transported to and retained to some degree in all major organs, as indicated by postmortem findings.

It is commonly stated that the toxicity from arsenic exposure varies in the order As(III) > As(V) >> MMA(V) ≅ DMA(V) (and other organic forms). Ingested MMA(V) and DMA(V) are metabolized less and eliminated more rapidly and to a greater degree than inorganic arsenic (IPCS 2001; ATSDR 2005a). Arsenic present in fish, primarily arsenobetaine, apparently undergoes little metabolism and is readily eliminated (Le 2002). At high doses, studies have shown that more arsenic is retained after ingestion of As(III) than As(V) (NRC 1999). A significant factor contributing to the greater toxicity of As(III) is its greater solubility in lipids and its ability to cross cell membranes more readily than As(V) species (Schoolmeester and White 1980), in part due to its neutral charge at physiological pH. Studies using laboratory animals have generally shown lower LD_{50} concentrations for As(III) ingestion compared to As(V) ingestion (IPCS 2001), which supports the greater toxicity of the As(III) form. As noted above, there is recent evidence suggesting that the toxicity of the metabolite MMA(III) may be more toxic that inorganic As(III). However, the ATSDR (2005a) has emphasized that studies based on laboratory animals do not provide good quantitative models for human toxicity.

Finally, many related factors are known or thought to modify arsenic toxicity. For example, diet has been shown to influence arsenic toxicity (Peraza et al. 1998) and more specifically the degree of methylation (e.g., Steinmaus et al. 2005), which may be one factor that explains variability of health effects among individuals or groups. Another aspect of interest is the observation that co-exposure to selenium may reduce the toxicity of arsenic (e.g., Levander 1977). The mechanism for this effect is under investigation, but may involve the formation of an arsenic-selenium complex either with reduced toxicity or more rapid elimination. Recent studies by Gailer and co-workers (2000, 2002) have identified the formation of a seleno-bis (S-glutathionyl) arsinium complex, $[(GS)_2AsSe]^-$, in rabbits injected with arsenite and selenate, with rapid excretion to bile.

ROLE OF METAL SPECIATION: CHROMIUM

Chromium is one of the more abundant heavy metals, with an average crustal concentration of 126 ppm (Wedepohl 1995). There is a long history of mining and processing of chromium, fueled by numerous industrial applications. Chromium is an important component in steel and other alloys, paints, magnetic recording tape, electroplating, wood preservative, and leather tanning, and serves as an anticorrosive agent in water-cooling systems. Other sources of chromium in the environment are listed in Table 4. Its widespread use has been accompanied by releases to the environment that pose persistent health hazards. Chromium is among the metals included in the ATSDR/EPA National Priority List of hazardous substances, and is present at a majority of sites on the CERCLA National Priority List (*http://www.atsdr.cdc.gov/cercla*). The environmental behavior and toxicology of chromium have been studied extensively. Readers are directed to the reviews of the environmental behavior of chromium by Rai et al. (1989), Fendorf (1995), and Kimbrough et al. (1999). Summaries of the environmental toxicology of chromium are given in ATSDR (2000), IPCS (1988), EPA (1998a,b), and O'Flaherty et al. (2001).

Chromium in the environment

Chromium may exist in oxidation states ranging from –II to VI. In the near-surface environment, the III and VI oxidation states are most important. It is well known that Cr(III)

Table 4. Some common sources of chromium in the environment.

- Occupational exposure from production of chromate, stainless-steel, chrome plating
- Air emissions and water effluents from ferrochrome production, ore refining, tanning industries, chemical manufacturing industries (e.g., dyes for paints, rubber and plastic products), metal-finishing industries (e.g., chrome plating), manufacturing of pharmaceuticals, wood, stone, clay and glass products, electrical and aircraft manufacturing, steam and air conditioning supply services, cement production
- Air emissions from incineration of refuse and sewage sludge
- Combustion of oil and coal
- Oxidation and leaching from stainless steel into a water-soluble form
- Motor vehicle exhaust and emissions from automobile brake linings and catalytic converters
- Tobacco smoke

is an essential micronutrient, playing a role in glucose metabolism. In contrast, Cr(VI) is considered a known carcinogen (by inhalation route) and is associated with both acute and chronic health effects (by inhalation and ingestion), as well as being a contact allergen (EPA 1998a; ATSDR 2000). Cr(IV) and Cr(V) are thought to play a role in the mechanism of toxicity and have been associated with production of reactive oxygen species and carbon-based radicals that may interact with DNA (cf. EPA 1998a; Gaggelli et al. 2002). In minerals, water, and most soils, chromium is dominantly coordinated with oxygen. Cr(III) shows a strong preference for octahedral coordination. The Cr^{3+} aqua ion undergoes hydrolysis, yielding $Cr(OH)^{2+}$, $Cr(OH)_2^{+}$, $Cr(OH)_3^{0}$, and $Cr(OH)_4^{-}$ species depending on pH (Fig. 13). Cr(III) may also form aqueous complexes with organic and inorganic ligands. Cr(VI) occurs almost exclusively in tetrahedral coordination with oxygen, as the chromate anion (CrO_4^{2-}). The pK_{a2} value for chromic acid, H_2CrO_4, is ~6.5, so that $HCrO_4^{-}$ dominates in solutions below pH 6.5, and CrO_4^{2-} dominates above (Fig. 13). Cr(VI) also occurs as dichromate, $Cr_2O_7^{2-}$, but this species only become important at millimolar Cr concentrations and greater.

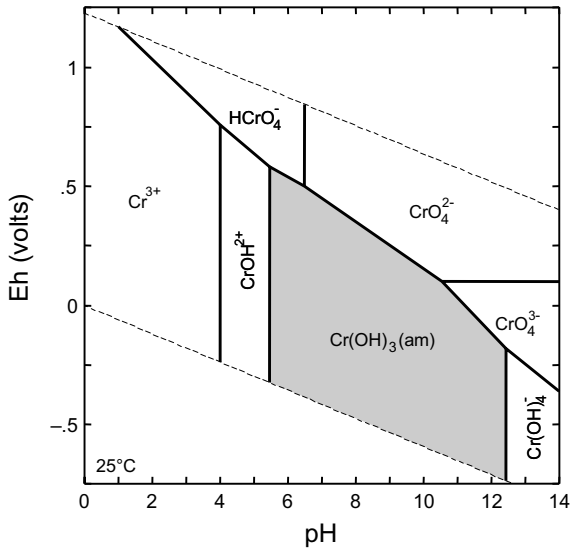

Figure 13. Eh-pH predominance diagram for aqueous chromium at 25 °C (Cr_{tot} = 1 μM).

Interconversion between Cr(III) and Cr(VI) depends on oxidation potential and pH (Fig. 13). Inasmuch as chromium is usually present at very low concentration in soils, surface sediments, and aquatic systems, other redox couples, commonly involving iron or organic matter, are usually important in controlling chromium oxidation state. Chromium occurring in natural minerals is dominantly as Cr(III), as in the important ore mineral chromite ($FeCr_2O_4$). The occurrence of Cr(VI) species and compounds is usually a result of anthropogenic activities or oxidative weathering.

Solubility and dissolution/precipitation. There are significant differences in the behavior of Cr(III) and Cr(VI) species under most surface conditions. Cr(III) compounds and minerals are largely insoluble except at low and very high pH. Over the pH range 6-10.5 precipitation of crystalline or amorphous $Cr(OH)_3$ or $(Fe,Cr)(OH)_3$ effectively limits dissolved Cr(III) concentrations to values below the current EPA drinking water MCL of 0.1 mg/L (Rai et al. 1987; Sass and Rai 1987), thereby limiting the oral exposure of this Cr species through water.

In contrast, Cr(VI) is highly soluble over the pH range of most natural waters. Chromate salts of sodium, potassium, magnesium, and calcium are highly soluble and rarely limit environmental Cr(VI) concentrations. Only $PbCrO_4$ (crocoite) is relatively insoluble. This striking difference in solubility between Cr(III) and Cr(VI) species has the unfortunate consequence that the essential micronutrient, Cr(III), is largely absent as a dissolved species in water while the toxic form, Cr(VI), is soluble and therefore potentially mobile. This solubility difference, however, also presents a potential remediation strategy based on conversion of soluble Cr(VI) to insoluble Cr(III).

Chromium reduction/oxidation. Owing to the significant difference in Cr(III) and Cr(VI) mobilities in the near-surface environment, redox processes are important for influencing chromium bioaccessibility and exposure to organisms. Although pH dependent, the Cr(III/VI) redox potential is relatively high, and CrO_4^{2-} is a strong oxidant. Ferrous iron, organic matter, and sulfides have been shown to reduce Cr(VI) to Cr(III) readily even in the presence of dissolved oxygen (James and Bartlett 1983a,b; Rai et al. 1989; Fendorf 1995; Patterson et al. 1997). Reduction by Fe(II) has been shown to result in the formation of $(Fe^{III},Cr^{III})(OH)_3$, which is insoluble and stable (Eary and Rai 1988). Cr(VI) reduction may be facilitated at surfaces of minerals containing Fe(II). For example, magnetite and ferrous biotite surfaces have been shown to reduce Cr(VI) to Cr(III) (Ilton and Veblen 1994; Peterson et al. 1996; Peterson et al. 1997). Fendorf et al. (2000) also demonstrated that Cr(VI) may be reduced by bacterial processes even in aerobic conditions.

Rai et al. (1989) and Fendorf (1995) report several studies in which Cr(III) was rapidly oxidized to Cr(VI) by manganese oxides, which are common constituents in soils. The effectiveness of this process, however, may be limited by formation of a hydrous $Cr(OH)_3$ or CrOOH precipitate at the MnO_2 surface (Fendorf et al. 1992; Charlet and Manceau 1993).

Chromium sorption. Owing to the high mobility of Cr(VI) in environmental systems much attention has been focused on the sorption behavior of this Cr species and its effectiveness in reducing mobility. As anion species (CrO_4^{2-} and $HCrO_4^-$), Cr(VI) exhibits greatest sorption at low pH (Fig. 14). Above pH 7 sorption may not be effective in removing Cr(VI) from solution (Zachara et al. 1987, 1989, 2004). In view of their nearly ubiquitous occurrence, iron oxides and hydroxides are likely to be some of the most effective sorbents. Chromate sorption may be strongly diminished as a result of competition for available surface sites by other anion species, such as carbonate, phosphate, and sulfate (e.g., Zachara et al. 1987).

Chromium in the body

Chromium exposure. The previous discussion focused primarily on environmental processes that control the behavior of different chromium species in soils, sediments, and aquatic systems. These influence human exposure mainly through ingestion of water, but may

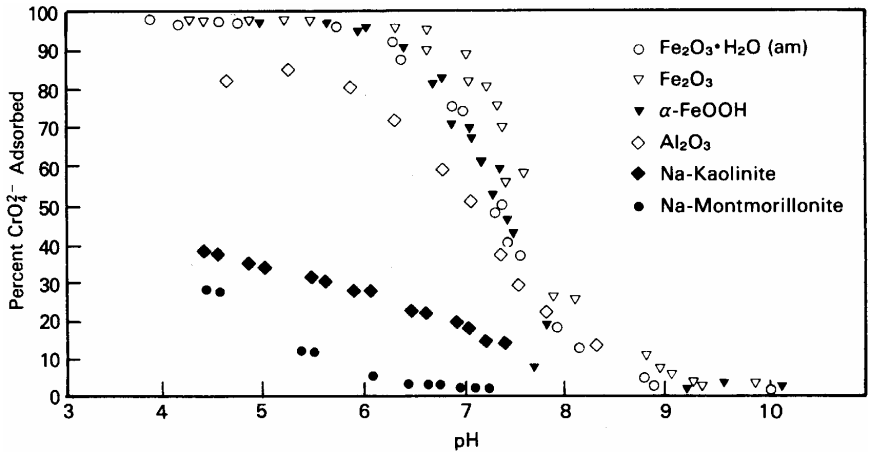

Figure 14. The pH dependence of chromate adsorption on different mineral and solid sorbents. [Reprinted with permission from Rai D et al., *The Science of the Total Environment*, Vol. 86, Fig. 4, p. 21. Copyright (1989) Elsevier.]

also relate to accidental ingestion of soil or inhalation of mineral dusts that contain chromium. The largest source of chromium intake for the general population is from food, where it occurs in the Cr(III) state (WHO 2003). Inhalation of Cr-containing airborne particles from industrial emissions, both Cr(III) and Cr(VI), may be locally significant, but most instances are associated with occupational exposures. Exposure may also occur through dermal contact. Here, we restrict the following discussion to illustrate examples where chromium speciation is relevant to health effects. Much of this information has been taken from the comprehensive reviews provided by IPCS (1988), EPA (1998a,b), ATSDR (2000), and WHO (2003). Readers are also referred to O'Flaherty (1996) and O'Flaherty et al. (2001).

Absorption and distribution. The efficiency of chromium absorption in the gastrointestinal tract is relatively low, and contrasts with the much greater absorption of arsenic noted in the previous section. Cr(VI) exhibits greater absorption than Cr(III), but both depend on the chemical form and other factors, including food and nutritional status. After oral exposure, most studies have shown absorption of soluble Cr(III) of ~0.5%, whereas absorption of soluble Cr(VI) compounds was 2-7% (ATSDR 2000; EPA 1998a,b), with a similar absorption efficiency for dissolved Cr(VI). However, Cr(VI) is partially reduced to Cr(III) in gastric fluid (and elsewhere in the body) (De Flora et al. 1987), which decreases the total absorption owing to the lower absorption efficiency of Cr(III). Essentially no absorption was observed following oral exposure of Cr(III) as insoluble Cr_2O_3 (Finley et al. 1996). Absorption of dietary Cr(III) is approximately 0.5-2% (EPA 1998b), and absorption of Cr(III) in the form Cr(III) picolinate (the form commonly used in vitamins) is as much as 3% (Gargas et al. 1994). Studies using lab animals generally support the greater absorption efficiency of Cr(VI) over Cr(III). Cr(III) absorption, however, may be enhanced when Cr(III) is complexed by organic ligands (e.g., oxalate) (ATSDR 2000) or when present in biologically active complexes (Mertz and Roginski 1971; O'Flaherty 1996).

Studies of inhalation exposure also show greater absorption of Cr(VI) than Cr(III) in the lungs. This has been confirmed by studies using lab animals, which have also indicated that the efficiency of chromium absorption in the lungs may be significantly greater than in the GI tract. Studies using a rat model (summarized in ATSDR 2000) showed 53-85% absorption from Cr(VI) particles (including clearance to the pharynx and into the GI tract) and 5-30%

absorption from Cr(III) particles. Lab animal studies also show that absorption depends strongly on the solubility of the inhaled compound, with greater absorption for more soluble phases (Bragt and van Dura 1983). This supports the view that dissolution of a solid to a soluble form is normally required for absorption across a physiological membrane.

The greater absorption of Cr(VI) compared to Cr(III) is largely due to the facilitated transport of chromate across cell membranes, following the same anion channel pathway as sulfate and phosphate (Wiegand et al. 1985). Absorption of Cr(III) is limited, occurring only by passive diffusion and/or phagocytotsis, which are much less effective. In the blood, Cr(VI) is able to cross the membrane of red blood cells, where it is rapidly reduced to Cr(III) and interacts with proteins. In contrast, Cr(III) is largely restricted to the plasma (WHO 2003). The greater tendency of Cr(VI) to cross cell membranes is also reflected in chromium distribution following exposure. One study using a mouse model showed that Cr was detected only in the liver following one year of exposure to Cr(III) chloride (a soluble form). In contrast, Cr was detected in all organs for mice exposed to soluble Cr(VI) for the same period (ATSDR 2000).

O'Flaherty (1996) has suggested that because of its low absorption efficiency the factors that influence bioaccessibility of any particular environmental chromium source are likely to be the single most important determinant of toxicity.

Metabolism and elimination. The essential role of Cr(III) is associated with a biologically active Cr(III) complex that is involved in glucose metabolism (Anderson 1986). Cr(VI) is not stable in the body, and a variety of electron donors, such as ascorbate and glutathione, cause reduction to Cr(III) species (De Flora et al. 1987). This proceeds throughout the body, including in saliva, gastric and lung fluids, blood, and in major organs. This reduction of Cr(VI) can be considered a defense or detoxification mechanism. However, Cr(V) and Cr(IV) intermediates are formed during reduction; these have been associated with formation of reactive oxygen species, and may be involved in the mechanism of toxicity (Gaggelli et al. 2002; Levina et al. 2003).

Absorbed chromium is mostly excreted as Cr(III) complexes through the urine within a period of several hours to several days (ATSDR 2000). The absence of Cr(VI) in urine, even after exposure to Cr(VI), indicates that reduction is complete within this time frame. Some chromium is retained in tissue and bone for periods on the order of months or longer. The large chromium fraction that is not absorbed following oral exposure is eliminated in the feces.

ROLE OF METAL SPECIATION: LEAD

Lead in the environment

Lead poisoning is one of the most common and serious environmental issues in industrialized nations, particularly with regard to its effect on the cognitive development of young children. Although lead occurs in the environment naturally, the vast majority of the instances of elevated lead levels in the environment that are of concern for human health are the result of human activity. Anthropogenic sources of lead include the mining, smelting, and refining of lead ore, emissions from coal and oil combustion, emissions from combustion of leaded gasoline, lead-based paints and solders, lead arsenate pesticides, and waste incineration. Much of the review of lead in the environment provided here is taken from the U. S. Department of Health and Human Service's Agency for Toxic Substances and Disease Registry on the effects of lead in the environment and its toxicology (ATSDR 2005b). In addition to the ATSDR overview, excellent reviews are also provided by Baxter and Frech (1995) and Ryan et al. (2004). Here, we provide a brief overview of the role of lead speciation as it relates to mobility in the environment and human exposure, bioavailability, and health effects.

Lead as a mineral is extremely rare (Wedepohl 1995). The most common form of lead in the Earth's crust is galena (PbS), but other important ore minerals include cerussite ($PbCO_3$), anglesite ($PbSO_4$), and minium (Pb_3O_4). Lead minerals are commonly found in association with zinc, copper, and iron sulfides and are commonly associated with gold and silver ores. Lead can also occur as a trace element in coal, oil, and wood. The majority of the lead used in industry today comes from recycling. Lead occurs in both the 2+ and 4+ oxidation states, but in nature the 2+ oxidation state dominates.

Lead species in natural waters. In most ground and surface waters, the solubility of lead compounds is generally low. The solubilities are dependent on water pH, the presence of coexisting ionic species and ligands, water salinity, and the organic matter content. Solubility tends to be highest in soft, acidic water. Eh-pH diagrams for the system Pb-CO_2-H_2O are shown in Figures 15a and 15b for 5 ppb and 207 ppb Pb_{tot}, respectively (the EPA action level for Pb in drinking water is 15 ppb; 207 ppb = 1 μM). Lead can exist as the Pb^{2+} ionic species at pH < 7.5 in fresh water, but readily complexes with dissolved carbonate at pH > 5.4 and forms lead carbonates, $PbCO_3$ and $Pb_2(OH)_2CO_3$, limiting its solubility (Long and Angino 1977). In natural waters lead will readily precipitate in the form of various lead hydroxides, carbonates, sulfates and phosphates, so that the amount of lead remaining dissolved in solution is typically low (Mundell et al. 1989). In highly oxidizing waters, such as may be found in municipal water systems, lead may precipitate as plattnerite (PbO_2). In surface waters most of the lead that is in suspension occurs as colloidal and undissolved particles. In seawater lead carbonate complexes are also prevalent, but lead chloride complexes and surface complexes with iron and manganese oxides can also occur (Long and Angino 1977; Elbaz-Poulichet et al. 1984).

Lead species in the atmosphere. The largest influx of lead to the environment has historically come from emissions to the atmosphere due to automotive and industrial sources. Lead particles are emitted from smelters primarily in the form of elemental lead and lead-sulfur compounds, $PbSO_4$,

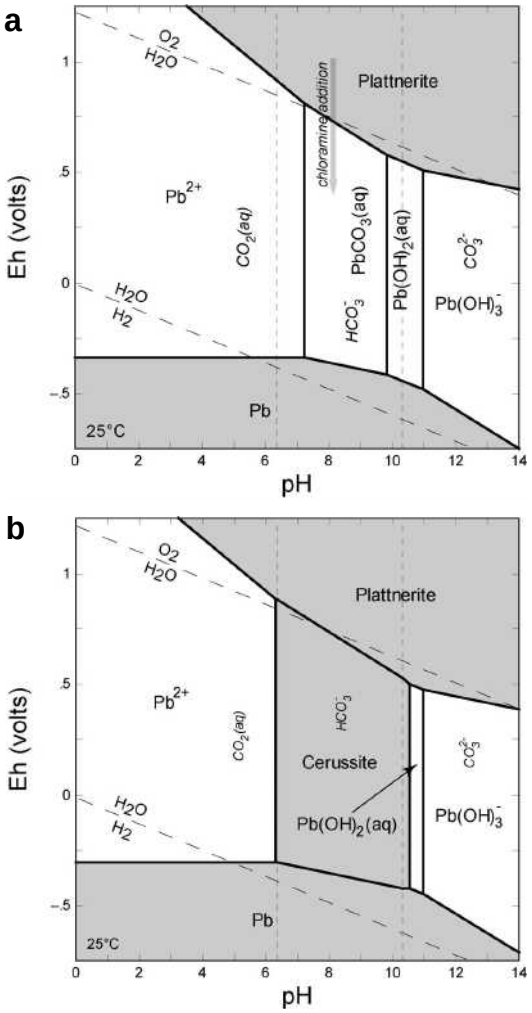

Figure 15. Eh-pH predominance diagrams for lead in the system Pb-CO_2-H_2O, 25 °C. (a) Pb_{tot} = 5 ppb (0.024 μM), C_{tot} = 18 ppm (1.5 mM); (b) Pb_{tot} = 207 ppb (1 μM), C_{tot} = 18 ppm.

$PbO{\cdot}PbSO_4$, and PbS (Corrin and Natusch 1977; EPA 1986; Spear et al. 1998). Oil and coal combustion has also been found to release $PbCl_2$ and PbO (Nerin et al. 1999). Prior to 1990 nearly 90% of all lead emissions were the direct result of combustion of leaded gasoline. In 1984 the U.S. Environmental Protection Agency mandated the gradual phase-out of the use of lead alkyls in gasoline and banned their use entirely in 1996 (EPA 1996). Today, industrial emissions from metal smelting operations and battery manufacturing are the largest sources of lead to the atmosphere.

The vast majority of the lead emissions from leaded gasoline are in the form of inorganic particulates such as lead bromochloride. These halogenated lead compounds are formed from the combustion of gasoline as the tetra-alkyl lead additives react with halogenated lead scavenger compounds (EPA 1985). Less than 10% of the lead emitted from combustion of leaded gasoline is emitted as organolead compounds such as vapor phase lead alkyls. However, tetra-alkyl lead vapors are strongly photoreactive, and quickly react to form trialkyl and dialkyl lead compounds, and eventually inorganic lead oxides (Eisenreich et al. 1981). In direct sunlight the half-life of tetraethyl lead vapors is approximately 2 hours (DeJonghe and Adams 1986). These lead particles are then deposited to either land or water through both wet and dry deposition. Their fine particle size and solubility in acidic solutions make these particles a particularly potent source of bioaccessible lead.

Lead species in soils. Once deposited to soils, the speciation of lead can take several different forms depending on the soil type. Key factors controlling its speciation in soils include soil pH, the organic matter content, and the soil cation exchange capacity (NSF 1977; Reddy et al. 1995). All of these factors, along with the mineral form, particle size, and association with other mineral phases, affect speciation of lead in soils and its potential for solubilization, which would influence its bioaccessibility and bioavailability (Fig. 16; Ruby et al. 1999).

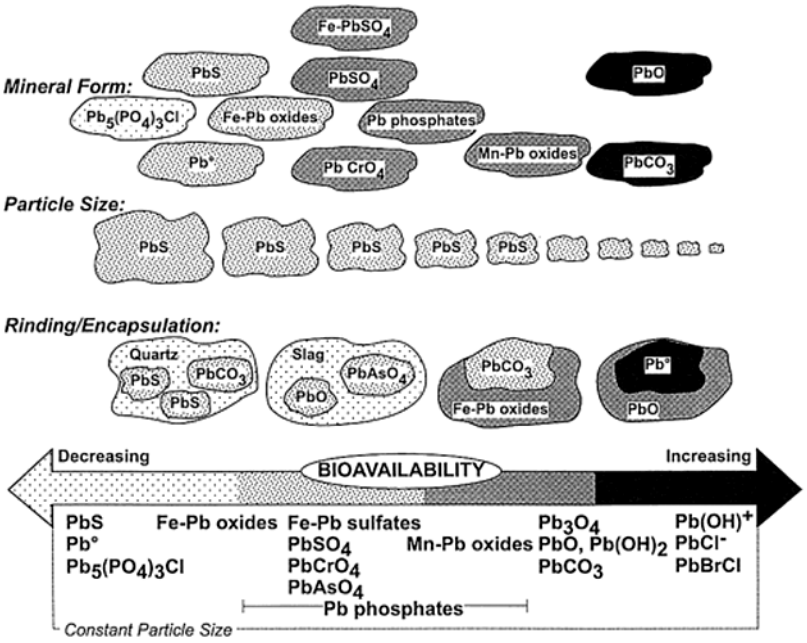

Figure 16. Properties of lead species and compounds and their effects on lead bioaccessibility and bioavailability in soils. [Reprinted with permission from Ruby et al. (1999). Copyright (1999) American Chemical Society.]

Sorption processes play a very important role in lead speciation in soils. In particular, iron(III) and manganese(III/IV) oxides and hydroxides, which are ubiquitous in most soils, are very effective sorbents for lead (O'Reilly and Hochella 2003). A number of studies (e.g., O'Reilly and Hochella 2003; Villalobos et al. 2005) have demonstrated that Mn-oxides in particular are very efficient sorbents of Pb. Figure 17 shows the measured lead sorption capacities for a variety of synthetic Fe- and Mn-oxides. Synthetic birnessite, in particular, can sorb large amounts of lead from solution, almost an order of magnitude more than Fe-oxides. EXAFS studies have indicated that Pb^{2+} forms inner-sphere complexes within the birnessite inter-layer (Morin et al. 1999; Matocha et al. 2001; Manceau et al. 2002).

Soil organic matter can also play a vital role in determining lead speciation in soils. Soil humic and fulvic substances, for example, have been shown to tightly bind lead (Pinheiro et al. 1999; Christl et al. 2005; Newton et al. 2006). Although humic substances contain several major functional groups (primarily carboxylic groups ~80% and phenolic groups ~20%), it is generally thought that Pb exhibits a strong affinity for the carboxylic groups (COOH) of the humic substances (Stevenson 1994; Xia et al. 1997). X-ray absorption studies of Pb-humate systems indicate that Pb can form inner-sphere complexes with the humic component and that the structure of the binding site does not change appreciably as a function of pH (Xia et al. 1997).

For example, Morin et al. (1999) examined smelter-contaminated soils from Evin-Malmaison, France, analyzing both tilled and wooded soil samples. They showed that although the source of Pb contamination was similar, differences in the chemical forms of Pb existed depending on soil type. In the wooded soil ~40% of the Pb was found to exist as outer-sphere complexes, ~10% as Pb^{2+} adsorbed on hydrous Fe oxides or oxyhydroxides, and ~50% as Pb^{2+} inner-sphere complexes bound to organic matter. By contrast, in the tilled soils Pb was present almost entirely as inner-sphere complexes adsorbed on hydrous Fe and Mn oxides. The higher amount of organo-Pb^{2+} and exchangeable outer-sphere Pb^{2+} complexes in the wooded soil was attributed to their higher organic matter content (6.4 wt% vs. 1.5 wt% TOC in the tilled soils).

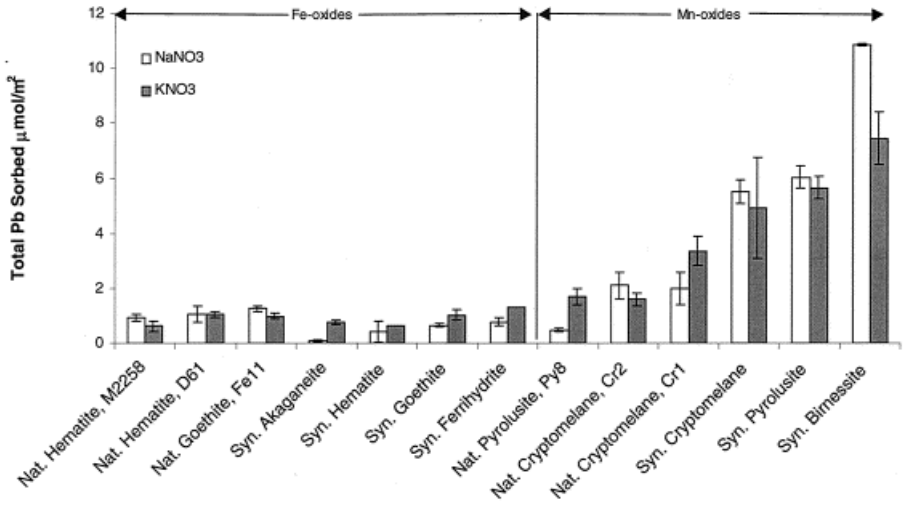

Figure 17. Average total lead sorption by various Mn and Fe-oxide minerals. Studies were conducted using $NaNO_3$ (white bars) and KNO_3 (gray bars) background electrolyte. [Reprinted with permission from O'Reilly and Hochella, *Geochimica et Cosmochimica Acta*, Vol. 67, Fig. 6, p. 4479. Copyright (2003) Elsevier.]

However, the binding of lead with humic components can be strongly affected by the presence of other major ions such as Ca^{2+}, Mg^{2+}, and Al^{3+}. Mota et al. (1996) demonstrated that by increasing Al^{3+} concentration, it was possible to also increase the concentration of free lead, showing that aluminum competes with lead for the binding sites of humic substances. This can be an important effect in soils containing gibbsite, where Al^{3+} concentrations may be high. Pinheiro et al. (1999) demonstrated a similar competitive binding of Pb^{2+} and Ca^{2+} to fulvic acid. Whereas solid organics can be quite efficient in diminishing lead mobility, aqueous organics, by contrast, can act to enhance lead mobility. Organic acids produced by plants and soil microorganisms can be particularly effective in chelating lead and enhancing mobility in soils.

Soil phosphate content also plays a role in controlling lead speciation. A number of studies of Pb-contaminated soils have shown that precipitation of Pb^{2+} as lead phosphate is effective in sequestering lead in soils (Davis et al. 1993; Cotter-Howells et al. 1994; Ruby et al. 1994; Juillot 1998; Traina and Laperche 1999; Morin et al. 2001). This is particularly important in evaluating lead bioaccessibility, inasmuch as members of the pyromorphite group [$(Pb,Ca)_5(PO_4)_3(OH,Cl,F)$] are among the most stable and least soluble lead minerals under surface environmental conditions (Nriagu 1973, 1974, 1984) and may form rapidly when adequate phosphate is present (Ruby et al. 1994). Their formation effectively limits the mobility of Pb in some soils. Cotter-Howells (1996) reported on the presence of lead phosphate in urban and roadside soils, demonstrating that these phases formed in the soil as a weathering product of Pb-bearing grains. The low solubility of pyromorphite has also advanced the use of phosphate amendments to lead-contaminated soils to promote pyromorphite formation.

There is also some indication that soil organisms may be able to biogenically precipitate pyromorphite in contaminated environments. Jackson et al. (2005) demonstrated that soil nematodes placed in a KCl-NaCl medium along with $Pb(NO_3)_2$ immobilize the Pb through pyromorphite precipitation. Given the high density and turnover of roundworms in soil, biogenic pyromorphite formation may influence Pb mobility and cycling in soils.

Lead species in paints and solders. The primary mode of lead exposure in children is through oral ingestion of Pb-containing products, such as lead-based paint and solder. Lead has been used as a pigment since ancient times and has been added to paint in a variety of forms. Perhaps the most common pigment is so-called "lead white," which contains a lead carbonate phase [$2PbCO_3 \cdot Pb(OH)_2$]. The popularity of lead white is due to its superior opacity. Other commonly used lead-based pigments include 'red lead' (Pb_3O_4 or $2PbO \cdot PbO_2$), lead-tin yellow (Pb_2SnO_4), and Naples yellow [$Pb(SbO_3)_2$ or $Pb(SbO_4)_2$]. The amount of lead in these pigments may be very high, commonly more than 38% of the dried weight of the paint.

In solders, lead is usually present as an alloy with tin. Electronic solders are usually 60% tin and 40% lead by weight in order to produce a near-eutectic mixture. Plumbing solders typically contain a higher proportion of lead. Corrosion of lead-containing plumbing materials in water distribution systems can release lead to drinking water, but the speciation of the lead in the water can vary dependent on municipal water chemistry. Renner (2004) highlights some of the complexities in evaluating the high levels of lead found in drinking water from some areas in Washington, D.C. In most natural waters, lead occurs in the Pb(II) oxidation state. Municipal drinking water, however, tends to be highly oxidizing due to the addition of significant amounts of chlorine. These oxidizing conditions promote the stability of Pb(IV) species such as PbO_2 (Fig. 15), which has been observed as a common scaling product in municipal water pipes in Washington, D.C. These scales remain stable and insoluble as long as the water remains oxidizing. However, many municipalities have switched from the use of chlorine to chloramine to comply with the EPA's 1998 Disinfection Byproducts Rule, which restricts disinfection byproducts in water. Chloramine use lowers the oxidizing potential of drinking water, so that PbO_2 scale may become soluble.

Lead in the body

Lead exposure levels. The toxicological effects of lead exposure have been well publicized and are generally familiar to most readers. Humans are often exposed to lead levels above that naturally occurring in soil or dust, and the most common contaminant sources and species have been discussed above. A 1999 Swedish study (Baecklund et al. 1999) characterized lead blood levels (PbB) in 176 men and 248 women, aged 49–92 years. Blood lead levels ranged from 0.56 to 15 μg Pb/dL (median 2.7 μg Pb/dL). The concentration of lead in the blood (PbB) is the most commonly used metric of absorbed dose for lead. The average PbB in the U.S. is ~2 μg/dL (0.1 μM or 20 ppb). The U.S. Centers for Disease Control (CDC) estimates that 890,000 children in the U.S. under 6 years of age may still have unsafe PbB and has established a 10 μg/dL threshold for elevated blood-lead level (CDC 1985).

Although elevated lead exposure is generally harmful, the greatest health risks from lead exposure are to preschool-age children and pregnant women and their fetuses. The most profound effects of lead exposure are neurological. Lead affects virtually every neurotransmitter system in the brain, and lead exposure has been linked to schizophrenia. Ancient Romans used lead acetate as a sweetener in wine, which has been thought to have caused a high incidence of dementia. Other medical conditions symptomatic of lead exposure include nephropathy (kidney damage), acute abdominal pain (lead colic or painter's colic), anemia, bluish discoloration of gums (gingival deposition of lead sulfide, *a.k.a.* Burtonian lines), gout (Saturnine gout), cardiac toxicity, endocrine effects, reproductive damage, and miscarriage. The EPA has classified elemental lead and inorganic lead compounds as probable human carcinogens.

Lead speciation and toxicokinetics. The toxicokinetics of lead in the human body have been extensively studied, but the role of speciation in toxicity is not completely understood. Studies have generally demonstrated that both organic and inorganic forms of lead are readily absorbed through inhalation, although factors such as particle size, solubility, and speciation may play important roles in determining lead absorption rates (ATSDR 2005b). Particles with diameters <1 μm are particularly well absorbed through inhalation exposure (~95% absorption for inorganic lead) since they can be deposited in alveolar regions of the lungs and can then be absorbed through extracellular dissolution or ingestion by phagocytic cells (ATSDR 2005b). Measurements of clearance rates for submicron particles of both inorganic lead, such as lead oxide and lead nitrate (Chamberlain et al. 1978), and organic species, such as tetraethyl or tetramethyl lead (Heard et al. 1979), are consistent with about 60-80% absorption 48 hours after initial deposition in the respiratory tract.

Ingestion is also a relatively effective pathway for lead absorption, and there are clear indications that the form of the ingested lead and the biochemistry of the gastrointestinal tract at the time of ingestion affect absorption rates. Dermal absorption appears not to be a particularly efficient pathway for the absorption of inorganic lead species. However, animal studies have shown that organic lead is well absorbed through the skin (ATSDR 2005b) and is much more readily absorbed in general. The available data on organic (i.e., alkyl) lead compounds indicate that some of the toxic effects of alkyl lead are mediated through metabolism to inorganic lead (EPA 1985).

Of the lead absorbed by the body, the majority ends up in mineralized tissues such as bone and teeth (Barry 1981, 1975): ~95% in adults and ~70% in children. The elimination half-life for inorganic lead in blood is approximately 30 days, whereas for bone it is approximately 27 years. Thus, PbB values only reflect exposure history for the previous few months, whereas lead in bone is considered a cumulative biomarker. These effects are a reflection of the proteins that bind lead in the body—proteins that naturally bind Ca and Zn (Godwin 2001). The two proteins that have seen the most study are synaptotagmin, a Ca^{2+} binding membrane protein widely expressed in the central and peripheral nervous system, and δ-aminolevulinic acid dehydratase

or ALAD, a zinc enzyme that catalyzes the second step of heme synthesis (Figure 18). Heme is the portion of hemoglobin that carries oxygen in the blood from the lungs to the rest of the body. In both these cases, the Pb^{2+} ion has a much higher binding affinity than either Ca^{2+} or Zn^{2+}. For example, consider that the CDC level for elevated PbB is 0.5 μM, whereas the average total concentration of zinc in human plasma is about 17 μM and calcium roughly 10^{-6} to 10^{-3} M (Godwin 2001). In the ALAD enzyme, lead binding to a three-cysteine site in the enzyme is ~500 times greater than zinc binding. When Pb^{2+} displaces the active Zn^{2+} ion present in the metalloenzyme, the enzyme is rendered useless, inhibiting hemoglobin production and resulting in anemia. In the synaptotagmin protein, Pb^{2+} appears to bind ~1000 times more strongly than Ca^{2+} (Bouton et al. 2001). Here, substitution of Pb^{2+} interferes with calcium-mediated signal transduction in neurotransmitters.

There are indications that at least for lead introduced via ingestion, its speciation in the presence of other metal components affects its absorption and toxicity. Simply relying on the total concentration level as a measure of potential threat is inadequate. For example, Ruby et al. (1999) examined lead gastrointestinal absorption using a juvenile swine model. Nineteen lead-bearing substrates containing similar amounts of total lead from eight hazardous waste sites were fed to test animals (Fig. 19), after which PbB was measured serially. At the completion of the study, samples of blood, bone (femur), liver, and kidney were collected and analyzed for lead concentration. This was then used to produce an estimate of relative lead bioavailability. Samples from carbonate-rich soils (Jasper, MO) yielded high lead bioavailability values. Soils derived from tailings, smelter slags, and soil from mining sites had the lowest relative bioavailability, while soils from the vicinity of smelters generally yielded higher values. It is important to bear in mind, however, that in large part this is a function of lead mineralogy; mine sites with high levels of cerrussite ($PbCO_3$) have tailings and soils with greater lead mobility than mine sites rich in galena (PbS).

Rat models also demonstrate differences in absorption depending on the form of lead ingested. Bone and tissue lead levels increase in a dose-dependent manner for animals receiving lead in their diet in the form of lead acetate, lead sulfide, or lead-contaminated soil. However, the bioavailability of lead sulfide was approximately 10% that of lead acetate (Freeman et al. 1996).

The presence of food in the gastrointestinal tract tends to decrease absorption of lead, but likely this is compositionally dependent as well as being a reflection of the biochemistry of the

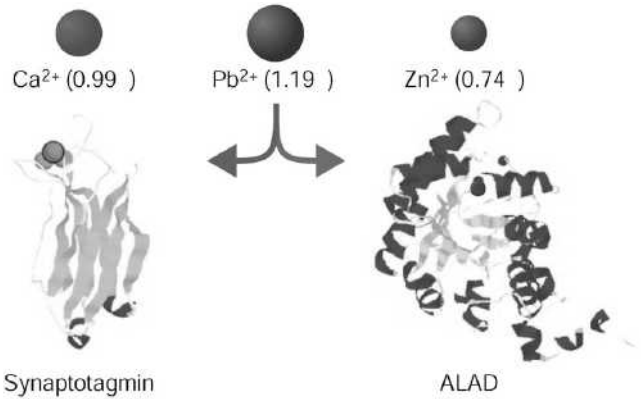

Figure 18. Lead binds with protiens that naturally bind calcium (synaptotagmin) and zinc (ALAD) in spite of their larger size. [Reprinted from Godwin, *Current Opinion in Chemical Biology,* Vol. 5, Fig. 1, p. 224. Copyright (2001) Elsevier.]

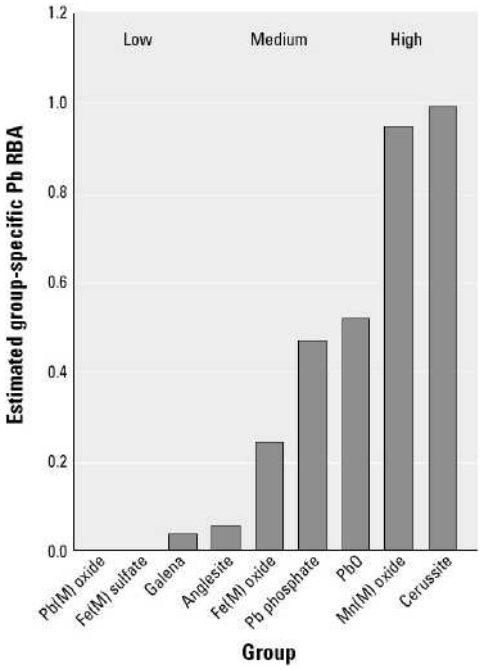

Figure 19. Relative lead bioavailabilities (Pb RBA) of various groups of solid phases, as determined from *in vivo* studies using a juvenile swine model. Pb(M) oxide = Pb oxide, Pb/As oxide, Pb silicate, Pb vanadate; Fe(M) sulfate = Fe/Pb sulfate, Pb sulfosalts; Fe(M) oxide = Fe/Pb oxide, Zn/Pb silicate; Mn(M) oxide = Mn/Pb oxide. [Reproduced with permission from *Environmental Health Perspectives*, Casteel et al. (2006).]

gastrointestinal tract during exposure (Rabinowitz et al. 1980; Heard and Chamberlain 1982; Blake and Mann 1983; Blake et al. 1983; James et al. 1985; Maddaloni et al. 1998). James et al. (1985) demonstrated that fasting adults given a tracer dose of lead acetate absorbed approximately 63% of the lead while subjects that were given a meal absorbed only ~3%. In particular, the presence of calcium and phosphate in the gastrointestinal tract tends to depress the absorption of ingested lead, as may the presence of oxalates and phytates (Heard and Chamberlain 1982; Blake and Mann 1983; Blake et al. 1983). For example, *in vitro* studies (Scheckel and Ryan 2003) have shown that when various Pb compounds ($PbCl_2$ and Pb paint) are introduced into a simulated stomach and gastrointestinal experimental system in the presence of cola soft drinks that contain phosphoric acid, rapid *in vitro* formation of pyromorphite occurs. The formation of pyromorphite within the stomach cavity may minimize Pb absorption through the higher pH gastrointestinal tract. Likewise, food in the stomach cavity may increase stomach pH so that less Pb is solubilized prior to reaching the GI tract. There are also animal studies indicating that zinc and iron deficiency may enhance lead absorption. Gastrointestinal absorption of lead occurs by acid solubilization (Ellenhorn and Barceloux 1988) and it seems that at least some lead transport across the digestive mucosa is similar to that of calcium (Gilman et al. 1990).

In considering gastrointestinal absorption of lead, researchers have developed a number of *in vitro* digestion models that compartmentalize the digestive process (Fig. 20; Oomen et al. 2003a,b). In order for lead to be absorbed within the gastrointestinal tract it must be mobilized from its matrix and present in a form capable of being transported across the intestinal epithelium. Gastric juices in the stomach can be quite acidic, with pH values as low as 1, which favors release of lead to a soluble form that makes it available for absorption (Fig. 15). Many studies assume that lead absorption primarily occurs in the small intestine, where pH values are 5-7.5 and lead is likely to be complexed (Oomen et al. 2003a). *In vitro* studies (Zhang et al. 1998; Oomen et al. 2003a) have found that the change from simulated stomach to intestinal conditions causes a rapid and complete transformation of lead to lead phosphates, such as chloropyromorphite [$Pb_5(PO_4)_3Cl_{[s]}$], and lead-bile complexes. As discussed earlier, lead phosphates would be expected to have very low solubilities and their formation could limit the concentration of soluble Pb; in fact these *in vitro* studies have demonstrated that the soluble Pb fraction in the simulated intestinal fluid is negligible. However, Oomen et al. (2003a) also demonstrated that the lead phosphates that are formed are labile, as also was shown by Feroci et al. (1995) for the lead-bile complexes.

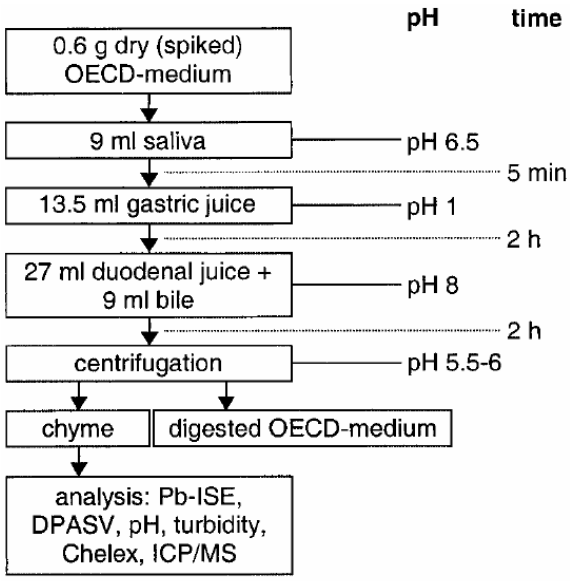

Figure 20. Schematic representation of an *in vitro* digestion procedure and subsequent analysis. [Reproduced with kind permission of Springer Science and Business Media from Oomen et al. (2003b).]

ROLE OF METAL SPECIATION: MERCURY

Mercury is an example of a metal that is far more toxic as an organometal species than in either metallic form or as an inorganic ionic species. Methylmercury (MeHg), in particular, poses a serious health threat. Monomethylmercury is the primary species of concern, although dimethylmercury has been detected in the marine environment. It has been proposed that one of the primary factors contributing to the high toxicity of MeHg is its high lipid solubility, which facilitates its transport across physiological membranes. Significantly, MeHg readily crosses the blood-brain barrier, where it has greatest impact. Unlike arsenic discussed above, the methylation of mercury occurs in the environment rather than within the body.

In this section we briefly summarize the sources of mercury in the environment, processes that control mercury speciation in the environment, their absorption and metabolism by humans, and the differences in toxicity between the methylated and inorganic forms. Detailed reviews of the environmental chemistry of mercury (Ullrich et al. 2001; Tchounwou et al. 2003; Fitzgerald and Lamborg 2004), the distribution and toxicity of MeHg (NRC 2000; Yokoo et al. 2003; Dopp et al. 2004; Norling et al. 2004), bioavailability of MeHg (NRC 2000; Tchounwou et al. 2003), and mode of action of mercury and mercury compounds have been published recently (Langford and Ferner 1999; NRC 2000; Schettler 2001; Tchounwou et al. 2003; Zheng et al. 2003; Fitsanakis and Aschner 2005; Garcia et al. 2005).

Mercury in the environment

Mercury, a Group IIb element in the periodic table, is rare in the Earth's crust, with average abundance estimated to be approximately 40 ppb (Wedepohl 1995). Organic-rich sediments and soils are enriched in mercury due to the uptake of mercury by biota, while sandstones and sedimentary carbonates contain on average 30 and 40 ppb Hg, respectively (Fitzgerald and Lamborg 2004). Plants take up atmospheric mercury through their leaves and soil-bound mercury through their roots (Tomiyasu et al. 2005). Once taken up, the mercury is essentially sequestered (Greger et al. 2005) and mercury remains associated with buried organic material. US coal contains on average 170 ppb mercury, much of it associated with pyrite and to a lesser extent the organic matrix (Tewalt et al. 2005). Most of the mercury in coal

pyrite is derived from degradation of organic compounds as the initial organic-rich sediment undergoes diagenesis. Mercury ores are formed from hydrothermal fluids. Cinnabar, HgS, is commonly the most abundant ore mineral in mercury deposits. Mercury has been used in ancient Chinese cultures and was mined by the Greeks and Romans for use as pigments and ointments. The modern-day, widespread use of mercury in gold mining, medical equipment, chemical processing, fluorescent lamps, and relays has led to a high demand for mercury.

Weathering of Hg-containing rocks and volcanic emissions are the most important natural processes that release inorganic mercury into the biosphere. Unlike weathering, volcanic emissions are episodic. For example, a recent study of an ice core retrieved from the Fremont Glacier in Wyoming, USA, shows a fivefold increase in atmospheric mercury deposition associated with the Krakatau eruption in 1883 (Schuster et al. 2002).

Mercury is a redox element, with Hg(0), Hg(I), and Hg(II) as stable oxidation states in the presence of water. Mercury is unusual in the fact that it is a liquid in its elemental state, a form in which it is readily volatilized. Furthermore, liquid mercury is stable over a wide range of redox conditions. The solubility of liquid mercury in water in the form of $Hg^0_{(aq)}$ is ~10^{-7} M at 25 °C (Glew and Hames 1971). However, $Hg^0_{(aq)}$ is volatile and readily lost to the atmosphere. In sulfur-rich, reducing environments the formation of insoluble mercury sulfide phases (cinnabar and metacinnabar) limits the solubility of mercury. In oxidizing environments the solubility of mercury is limited by the formation of mercury oxides. In natural environments, where the total concentrations of mercury are often vanishingly low, the solution concentration is controlled by complexation with organic ligands and sulfide on one hand and sorption onto organic matter and particulate matter on the other hand (Reddy and Aiken 2001).

Once mercury enters aquatic systems—either as dissolved species, associated with particulate matter, or as elemental mercury—it can become methylated (Fitzgerald and Lamborg 2004). An idealized cycle illustrating the major processes of mercury methylation in freshwater lakes is shown in Figure 21. Methylation of mercury is closely tied to the sulfur redox cycle. In anaerobic environments, sulfate-reducing bacteria promote methylation. It is thought that the formation of sulfide by sulfate-reducing bacteria leads to the formation of a suite of mercury-sulfide complexes (Ullrich et al. 2001). As shown in Figure 22, species such as $Hg(HS)_2^0{}_{(aq)}$ and $HgS_2^{2-}{}_{(aq)}$ account for as much as 10% of the aqueous mercury

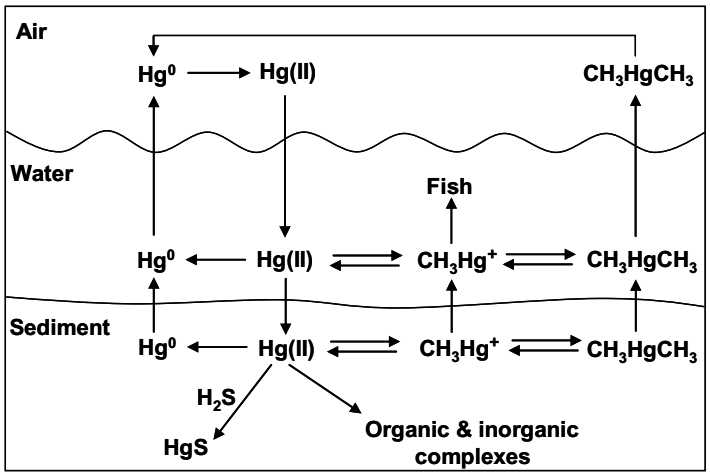

Figure 21. Idealized cycle of mercury methylation in freshwater lake environments. [Adapted from Winfrey and Rudd (1990).]

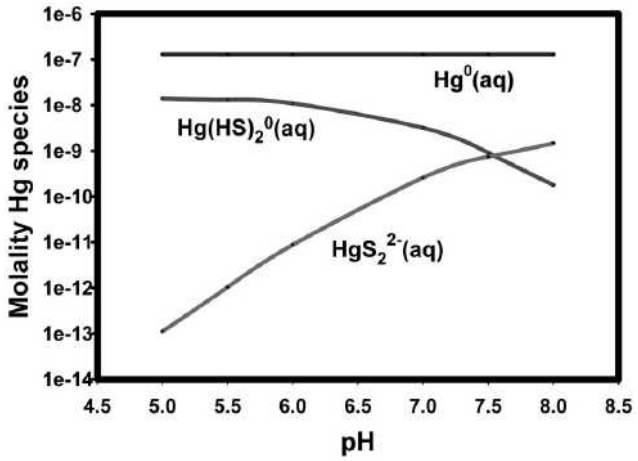

Figure 22. Aqueous inorganic Hg speciation in equilibrium with liquid metallic mercury in the presence of 0.1 mM sulfidic sulfur at 25 °C. Calculation based on MinteqV4 database provided with Phreeqc 2.12.5. The formation of metacinnabar and cinnabar has been suppressed for these calculations.

species in an anaerobic, sulfide-containing system in equilibrium with liquid mercury. Some of the neutral inorganic complexes, such as $Hg(HS)_2^0{}_{(aq)}$ and $Hg^0{}_{(aq)}$, can readily diffuse into the cells of sulfate-reducing bacteria where they are subsequently methylated as part of a cellular defense mechanism (Fitzgerald and Lamborg 2004). Methylation takes place under reducing conditions where hydrogen sulfide or bisulfide is the dominant sulfur species (see Fig. 23). Methylated mercury species are labile and can be demethylated leading to a complex, dynamic mercury cycle with significant fluxes of mercury between the atmospheric and aquatic reservoirs. A fraction of the methylated mercury is taken up in the aquatic food chain, bioaccumulates, and resists degradation (Norling et al. 2004).

While there is consensus that the global cycles of mercury have been significantly altered by anthropogenic emissions, there is considerable uncertainty about the magnitude of some

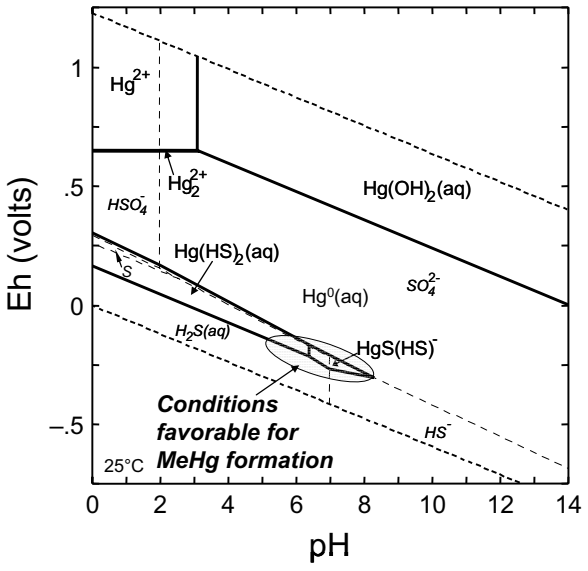

Figure 23. Eh-pH predominance diagram for mercury in the presence of sulfur at 25 °C; $Hg_{tot} = 10^{-9}$ M, $S_{tot} = 10^{-4}$ M. Formation of cinnabar and metacinnabar have been suppressed. Calculations performed with Geochemist's Workbench using LNLL database supplemented with thermodynamic data for $Hg^0{}_{(aq)}$, $Hg(OH)_{2(aq)}$, $Hg(HS)_2^0{}_{(aq)}$, $HgS(HS)^-{}_{(aq)}$, and $HgS_2^{2-}{}_{(aq)}$ taken from MinteqV4 database.

of the mercury fluxes (Fitzgerald and Lamborg 2004). It is estimated that anthropogenic mercury emissions account for about 66% of the total emissions. Table 5 summarizes natural and human inputs to the environment. Among anthropogenic inputs, the single most important process by far is fossil fuel burning, particularly burning of coal (Nriagu and Pacyna 1988; Pacyna and Pacyna 2002). While mercury emissions from coal power plants in Europe and the US have dropped substantially over the last decade, the rapid economic development in India and China has led to a significant increase in emissions in Asia (Pacyna and Pacyna 2002). Mercury released by fossil fuel burning is predominantly in the form of gaseous elemental mercury. A complex set of reactions in the atmosphere lead to the conversion of elemental mercury into ionic mercury, which is deposited into aquatic systems via a combination of wet

Table 5. Summary of relevant toxicokinetic data for three important mercury species[#].

MeHg	Hg(0)	$Hg(II)_{(aq)}$
Exposure		
Fish, marine mammals, crustaceans, animals and poultry fed fish meal	Dental amalgams, fossil fuels, occupational exposure, incinerators	Oxidation of elemental mercury or demethylation of MeHg; deliberate or accidental poisoning with $HgCl_2$
Absorption		
Inhalation: Vapors of MeHg absorbed Oral: Approximately 95% of MeHg in fish readily absorbed from GI tract Dermal: In guinea pigs, 3-5% of applied dose absorbed in 5 hr	Inhalation: ~80% of inhaled vapor Hg^0 dose readily absorbed in lungs Oral: GI absorption of metallic Hg is poor; any released vapor in GI tract converted to mercuric sulfide and excreted Dermal: Absorption of Hg^0 vapor through human skin is very low relative to inhalation absorption	Inhalation: Aerosols of $HgCl_2$ absorbed Oral: 7-15% of ingested dose of $HgCl_2$ absorbed from the GI tract; absorption proportional to water solubility of mercuric salt; uptake by neonates greater than adults Dermal: In guinea pigs, 2-3% of applied dose of $HgCl_2$ absorbed
Distribution		
Distributed throughout body since lipophilic; ~1-10% of absorbed oral dose of MeHg distributed to blood; 90% of blood MeHg in red blood cells; readily crosses blood-brain and placental barriers	Rapidly distributed throughout the body since it is lipophilic. Readily crosses blood-brain and placental barriers	Highest accumulation in kidney; fraction of dose retained in kidney is dose dependent. Does not readily penetrate blood-brain or placental barriers
Metabolism		
Slowly demethylated to Hg^{2+}	Hg(0) is oxidized in tissue and blood to Hg^{2+}	Hg^{2+} not methylated in tissue, but process may proceed in GI tract mediated by gut microorganisms
Excretion		
About 1% of burden released, mostly via bile and feces.	Excreted as Hg^0 in exhaled air, sweat, and saliva, and as mercuric Hg in feces and urine	Excreted in urine and feces; also excreted in saliva, bile, sweat, exhaled air, and breast milk

#Abbreviated from Table 2-2 in (NRC 2000)

and dry deposition (Fitzgerald and Lamborg 2004). Disposal of mercury-containing devices and incineration of mercury-containing waste contribute to its release into atmosphere. The use of liquid mercury in the extraction of gold by amalgamation has led to the release of large quantities of mercury directly into aquatic environments (Eisler 2004). A recent study has shown that methylation of mercury in wetlands increases with atmospheric sulfate loading (Jeremiason et al. 2006). This finding suggests that acid rain deposition is a confounding factor in the bioaccessibility of mercury in the environment.

Mercury in the body

Consumption of fish is the main source of exposure to mercury for humans. An estimated 95% of methylmercury contained in fish or shellfish is absorbed, with a significant fraction accumulating in the brain (Table 5). MeHg is slowly demethylated in the body to inorganic Hg(II), which is more readily excreted, primarily in the bile and then into feces. The ratio of MeHg to inorganic Hg(II) in tissue varies with duration of exposure and length of time after absorption. Postmortem studies have shown that as much as 80% of the mercury in the brain is inorganic Hg(II) (IPCS 1990).

Oral ingestion of liquid mercury results in very low absorption in the gastrointestinal tract. Less than 10% absorption is reported for oral ingestion of inorganic Hg(II) contained in food (Elinder et al. 1988). Inhalation of mercury vapor results in high absorption in the alveoli of the lungs, as much as 80%. Once dissolved in the blood, Hg(0) is enzymatically oxidized to Hg(II) and is partly associated with red blood cells. Hence, metallic mercury is considered a Fenton metal (Valko et al. 2005).

The ability to cross the blood-brain barrier contributes to the toxicity of methylmercury (Garcia et al. 2005). The mechanism(s) of toxicity are not fully understood (IPCS 1990). Poisoning by MeHg is known as Minamata Disease, named after an infamous incident in which thousands of people were exposed to fish contaminated with MeHg released from an industrial source. Currently, informal gold mining in the Amazon is affecting native Indians who rely on fish as a source of protein (Lodenius and Malm 1998; Hylander et al. 2000; Maurice-Bourgoin et al. 2000). An estimated 2000 tonnes of mercury has been released into the Amazon since 1980 (Malm 1998). Some of this mercury is transformed into MeHg which enters into the aquatic food chain. Population studies among Amazonian Indian communities show that individuals in communities eating contaminated fish are more likely to develop neurological diseases (Dolbec et al. 2000; Counter 2003) and some have mild symptoms of Minamata disease (Lodenius and Malm 1998).

METHODS FOR CHARACTERIZATION OF METAL SPECIATION

The choice of methods for speciation analysis typically depends on which metals need to be analyzed, which species are expected, and the physical forms of the materials to be analyzed (e.g., liquids vs. solids). The approaches that can be used in speciation analysis vary broadly since distinct chemical species encompass chemical compounds that can "differ in isotopic composition, conformation, oxidation or electronic state, or in the nature of their complexed or covalently bound substituents" (Templeton et al. 2000). The following overview of commonly used techniques is not meant to be either definitive or comprehensive. The number of techniques available to the researcher today is truly staggering and the techniques are rapidly evolving. The techniques that we highlight here reflect some of the research examples presented in this chapter. These brief descriptions should serve as a starting point for readers in assessing the techniques that will be most suitable for their given problems. It is also important to recognize that often it is necessary to use several techniques to obtain a more complete view of metal speciation (e.g., Scheinost et al. 2002).

For more comprehensive descriptions readers are referred to the "Handbook of Elemental Speciation: Techniques and Methodology" by Cornelis et al. (2003) and to review articles by Tack and Verloo (1995), Caruso et al. (2003), and D'Amore et al. (2005). But it must be emphasized that in applying any of these techniques particular attention should be given to ensure that sampling, sample storage, and sample preparation do not alter either the metal concentrations or other chemical properties for which the speciation analysis is being conducted.

Equilibrium modeling

Frequently, equilibrium modeling is a critical step in establishing the identity of chemical species likely to be present under given conditions, particularly in aquatic systems or in solutions. Equilibrium modeling generally assumes that all reactions in a system have gone to completion and is thus not typically suited to evaluating reaction kinetics. However, it can be very powerful in modeling equilibria among dissolved, adsorbed, solid, and gas phases as a function of pH and ionic strength. For many metal species, with just a measurement of solution pH and component concentrations a researcher can readily model the equilibrium abundances of metal ion species in the system. Numerous computer software programs exist to facilitate such modeling, more than can be described here. Some of the most commonly used programs include MINTEQA2 (U.S. EPA), PHREEQC (Parkhurst and Appelo 1999), and The Geochemists Workbench (Bethke 2002) (*http://www.rockware.com*). The user can typically input the total component concentrations and any invariant system parameters such as pH, p*e*, or gas partial pressures. In some cases you can also specify if given mineral phases are presumed to be present at equilibrium. The results of model calculations are not only dependent on user-defined constraints, but also the thermodynamic database used. In programs such as PHREEQC the user can select among several databases. The databases differ in the number of elements and species for which thermodynamic data are included. A substantial caution exists in that such modeling is only as good as the thermodynamic data used and the assumptions made to constrain the modeling.

Sequential extraction of solid species

Sequential extraction protocols in and of themselves are not methods of directly assessing speciation, but rather a means of isolating chemical species for analysis. For some solids, extraction protocols are necessary for effectively measuring their abundance. The number of sequential extraction protocols available for various metal species are numerous and are readily found by searching published articles. Many of these techniques are modifications of the procedure developed by Tessier et al. (1979) for the partitioning of particulate trace metals into five fractions: exchangeable, acid soluble (bound to carbonates), reducible (bound to Fe-Mn oxides), oxidizable (bound to organic matter or sulfides), and residual. In particular, an emphasis is placed on the choice of extraction reagents and their selectivity in each leaching solution. For example ammonium acetate is commonly used to liberate exchangeable metals, and sodium acetate or acetic acid are effective at acidic pH values for selective dissolution of the carbonate fraction.

Caution is needed, however, in applying extraction procedures to systems other than those for which the procedures were initially developed. Several studies have shown that results from sequential extraction procedures can vary dramatically when applied to terrestrial soils and sediments. For example, Scheckel et al. (2003) noted the problem of extracted metal species alteration before, during, and after separation of solids from solution. For soils spiked with Pb and amended with calcium phosphate, they observed significant shifts of extractable Pb to the residual phase during the sequential extraction steps, which were attributed to pyromorphite formation. Similarly, in the evaluation of As species in soils Mihaljevič et al (2003) demonstrated how the use of extraction techniques can be complicated by the occurrence of As extracts in the form of anionic complexes resulting from the dissociation of H_3AsO_4. Moreover, sequential extraction protocols are generally found to be problematic for As and Se (Gruebel et al. 1988).

Electrochemical methods: voltammetry and polarography

Voltammetry allows metal concentrations in solutions to be measured at extremely low levels, often at parts per trillion concentrations. Some organic compounds can also be measured. It is also an excellent tool for measuring metal oxidation states and differentiating between free and bound metal ions in solution. Voltammetry measures the current at a working electrode as a function of the applied potential, where the current is proportional to the analyte concentration in solution. Working electrodes can be made of several conducting materials such as gold, platinum, or carbon. In polarography, a dropping mercury electrode is used to continuously renew the electrode surface. The method is really only directly applicable to the analysis of species in solution and care must be taken to ensure that sample preparation does not induce changes in speciation. Nevertheless, it is a technique well suited for examining metal speciation in aquatic systems and has been widely applied to *in situ* measurements on natural waters (Van Leeuwen et al. 2005). It is also applicable to analysis of metals in bodily fluids, for example the analysis of blood lead levels (Bannon and Chisolm 2001). In cyclic voltammetry the scan rate of the experiment can be "tuned" to the kinetics of the electrochemical reaction, making it also useful for examining redox kinetics.

Liquid membrane techniques

As with voltammetry, liquid membrane techniques are suitable for measuring the concentration and distribution of chemical species in liquids, including natural waters, plant extracts, and bodily fluids. Detection limits are typically on the order of micrograms per liter. Dialysis is a familiar liquid-membrane technique. Different membrane techniques exist based on the type of membrane material. The permeation liquid membrane (PLM) technique, for example, is based on liquid-liquid extraction principles. A polymer membrane is used that supports a hydrophobic ligand as a carrier molecule. The membrane separates the solution to be analyzed (containing a natural ligand) from a receiving, strip solution, setting up a chemical potential gradient. The flux through the membrane can then be measured and related to the concentration of the free ion or labile metal complex. The Donnan membrane technique (DMT) is similar, but uses a cation exchange membrane to measure free ion activity using the Donnan principle (Lampert 1982). Detection limits are typically on the order of milligrams per liter. Here, the analyte solution is separated from an anion containing receiving solution by a negatively charged semipermeable membrane (Zhang and Young 2006). Because of the negative charge of the membrane, cations move across the membrane to the acceptor solution.

Liquid chromatography

There are numerous chromatographic methods used for speciation analysis. In chromatography an analyte is passed through a stationary phase (such as an ion exchange resin) as a gas or liquid, with each component having a distinct retention time. In essence, all chromatographic methods are separation techniques that take advantage of differences in partitioning, adsorption, ion exchange behavior, or molecular size of the analyte species relative to the stationary phase. For the speciation analysis of environmental samples the most commonly employed chromatographic techniques are typically based on some form of liquid chromatography such as high-performance liquid chromatography (HPLC) or ion chromatography. The exact technique to be used is dependent on the species of interest. Liquid and gas chromatographs are also commonly interfaced to element specific detection systems such as mass spectrometers (LCMS and GCMS) or nuclear magnetic resonance systems (LC/NMR). Such coupled devices ('hyphenated' techniques) have detection limits at low-femtomole levels. Such combined systems can elute species sequentially from the chromatographic column directly to the mass spectrometer for compound-specific identification. For example, such techniques have been used in measuring Se speciation in Se-enriched garlic (Dumont et al. 2006).

Capillary electrophoresis

Capillary electrophoresis (CE) can be used for the separation of a wide variety of species, of inorganic and organic ions and of varying ionic radius. The technique relies on ion migration of charged electrical species in an electrolyte in the presence of an electric current. The use of capillaries facilitates automated analysis and peak detection. A number of variants of the technique exist that allow for separation based on size and charge differences between species. As with chromatography, CE systems are typically coupled to detector systems such as UV absorbance detectors, diode detectors, fluorescence detectors, or as hyphenated techniques coupled to mass spectrometers. Advantages of the technique are high separation efficiency, high speed of analysis, and the ability to use very small amounts of sample. As an example, Forte et al. (2005) were able to use CE analysis to attain the simultaneous separation of seven arsenical species: arsenite (As III), arsenate (As V), monomethylarsonic acid (MMA), dimethylarsinic acid (DMA), arsenobetaine (AsBet), arsenocholine (AsCh) and p-arsanilic acid (pAs), as well as quantify the abundance of each species.

Inductively coupled mass spectrometry (ICP-MS)

As described above, ICP-MS systems are now commonly coupled to GC and HPLC separation systems. In fact, HPLC-ICP-MS may arguably be the most widely used technique for speciation analysis in aqueous media. In ICP-MS, plasma is used to atomize and ionize the species in a sample. The species are then identified by their mass-to-charge ratio. Detection limits for ICP-MS are typically better than sub ng/L. ICP-MS can detect a wide range of species containing trace elements, with a very large dynamic range (Rosen and Hieftje 2004). It can thus be used for analysis of a wide range of both trace and major elements from the same sample dilution. However, in this mode serious interference problems can exist, which must be evaluated during analysis. For example, low levels of arsenic cannot be analyzed in high-chloride solutions, such as sea water, using standard ICP-MS due to interferences; as a result, specialized ICP-MS systems, such as high-resolution ICP-MS or dynamic-reaction chamber (DRC) ICP-MS techniques, are needed for analysis of low levels of As, Mo, Se, and some other elements in sea water. There can also be problems with the composition of the solvent injected into the nebulizer leading to plasma instability.

Magnetic spectroscopies (NMR, EPR, and Mössbauer spectroscopy)

Magnetic spectroscopies exploit the magnetic properties of nuclei. Nuclear Magnetic Resonance (NMR) spectroscopy, Electron Paramagnetic Resonance (EPR) spectroscopy, and Mössbauer spectroscopy are the three most familiar magnetic techniques used for speciation analysis. NMR spectroscopy uses the fact that when NMR-active nuclei are placed in a magnetic field of a given strength, they resonate at a specific frequency, dependent on strength of the magnetic field. Since different chemical groups in a compound resonate at slightly different frequencies, the "chemical shift" imparted to the proton spectrum by each component can be used to identify the compound. NMR techniques are restricted to the analysis of non-ferromagnetic elements with spin ½ nuclear isotopes. Detection sensitivities are on the order of 10-10,000 ppm (D'Amore et al. 2005). Electron paramagnetic resonance (EPR) or electron spin resonance (ESR) is analogous to NMR, but detects species that have unpaired electrons. Thus transition-metal and rare-earth species that contain unpaired electrons can readily be analyzed. Organic molecules that contain unpaired electrons can also be analyzed. EPR usually requires microwave-frequency radiation (GHz), while NMR is observed at lower radio frequencies (MHz). NMR and EPR are generally nondestructive techniques as they utilize non-ionizing radiation. Mössbauer spectroscopy utilizes the fact that when some solid samples are exposed to a beam of gamma radiation, the intensity of the beam transmitted through the sample will change as a function of gamma-ray absorption because there is a lack of recoil. Mössbauer analysis is restricted to elements that display this behavior. Most

notably for environmental analysis, Fe, Cs, Ba, Ni, Zn, Sn, Sb, and Hg can be analyzed. Some actinides such as Th, Pa, U, Np, and Pu can also be analyzed. Detection limits are on the order of 1-1000 ppm, and since the technique can be used to detect shifts in transition energy around the absorbing atom, it is well suited for detecting changes in valance and site substitutions (D'Amore et al. 2005).

X-ray diffraction and X-ray absorption spectroscopy

A number of X-ray techniques can be used to evaluate metal speciation in environmental and biological materials. The three most commonly used techniques are X-ray diffraction (XRD), X-ray fluorescence (XRF), and X-ray absorption spectroscopy (XAS). Many readers will be familiar with XRD, which provides information on crystal structure and phase identity from the Bragg scattering vectors from X-rays interacting with a periodic array of atoms. It is worth noting that neutron diffraction offers benefits for certain materials, particularly where structural information for protons or water is needed. Although most commonly used for crystalline materials, scattering techniques may also be useful for characterization of non-crystalline materials.

We also assume that many readers are familiar with X-ray fluorescence (XRF), which provides quantitative elemental analysis of materials in solid, liquid, and even gaseous forms. The increased availability of synchrotron X-ray sources, with their extremely high brightness and brilliance, has allowed improvements in spatial resolution and sensitivity of X-ray methods by orders of magnitude relative to laboratory X-ray tube sources (Sutton et al. 2003). Synchrotron-based XRF can detect metal concentrations with a sensitivity of approximately 1 fg. Synchrotron-based XRD with spatial resolution better than 10 μm is now possible, allowing for *in situ* phase identification of complex, heterogeneous materials. X-ray absorption spectroscopy (XAS) is an element-specific technique. Largely restricted to synchrotron sources, XAS is now routinely performed on samples with metal abundances as low as 1 to a few tens of ppm, depending on the metal and the nearest-neighbor atoms. X-ray absorption near-edge structure spectroscopy (XANES) provides direct information on oxidation state and coordination, whereas extended X-ray absorption fine structure spectroscopy (EXAFS) allows characterization of the number and type of neighboring atoms and their distance from the absorbing element. One of the primary advantages of these synchrotron-based X-ray techniques for assessing speciation (compared to those discussed above) is that samples need not be disturbed or destroyed for study; XAS allows direct, *in situ* characterization (Manceau et al. 2003). For example, synchrotron-based XAS methods can identify the chemical speciation and elemental associations of arsenic in dilute (<100 mg kg^{-1}) heterogeneous materials, such as poultry litter, with high spatial resolution (Arai et al. 2003), or measure the speciation of lead within organisms such as soil nematodes *in situ* with no need for species separation, pre-concentration, or pre-treatment (Jackson et al. 2005). Most of the methods previously described require separation or processing (e.g., dissolution for the analysis of non-liquid samples). It is not surprising, therefore, that synchrotron-based techniques have become so widely used in speciation analysis.

FUTURE RESEARCH AND ROLE OF GEOCHEMISTS

Geochemistry and mineralogy have been integral parts of exposure studies for many years. In particular the insight provided into the speciation of metals and the processes that control speciation, mobility, and ultimately bioaccessibility represent important contributions. A new frontier will be the integration of geochemical concepts and methods with molecular toxicology to better understand the mechanisms by which metals cause adverse health effects. One of the challenges is to identify and incorporate all relevant biological ligands into models. For example, ligands such as glutathione (Fig. 24) may be present at

millimolar concentrations in different physiological fluids and are known to have a high affinity for certain metal species. However, in many cases the necessary metal complexation equilibria and redox equilibria are not yet known. One possible path forward is to use approximation and extrapolation techniques that have served geochemist well over the last half century to obtain estimates of the most relevant equilibria.

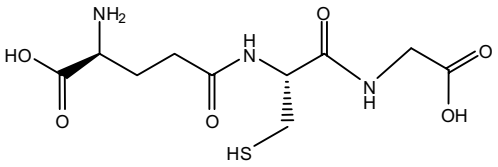

Figure 24. Glutathione is one of many biologically important molecules that complex metal species, with the sulfhydryl group being a primary binding site.

Microspatial speciation analysis of biological tissue samples represents another major challenge. While total metal analysis of tissues is routine, microspatial and in many cases redox-specific microspatial analysis is expected to offer new insights into the role of metals in disease. Instrumental techniques familiar to many geochemists and mineralogists, such as synchrotron-based micro-XRF and micro-XAS, are now being used more commonly for speciation analysis in biological tissues. With continued improvements in spatial resolution and signal quality provided by new generation synchrotron sources, further advances in speciation of metals will offer new opportunities for research.

ACKNOWLEDGMENTS

We thank our many colleagues and students who share an interest in multidisciplinary research. Support for RJR and MAAS was partially provided by the Center for Environmental Molecular Science (NSF grant CHE-0221934). AL was supported by Department of Energy (DOE) - Geosciences (grant DE-FG02-92ER14244). We thank Geoff Plumlee, Blair Jones, and an anonymous reviewer for helpful comments that improved this work. We also thank Nita Sahai for her patience during manuscript preparation.

REFERENCES

Alzieu C (1996) Biological effects of tributyltin on marine organisms. *In:* Tributyltin: Case Study of an Environmental Contaminant. de Mora SJ (ed) Cambridge University Press, p 167-211

Anderson LCD, Bruland KW (1991) Biogeochemistry of arsenic in natural waters: the importance of methylated species. Environ Sci Technol 25:420-427

Anderson MA, Rubin AJ (1981) Adsorption of Inorganics at Solid-Liquid Interfaces. Ann Arbor Sciences

Anderson RA (1986) Chromium metabolism and its role in disease processes in man. Clin Physiol Biochem 4:31-41

Arai Y, Elzinga EJ, Sparks DL (2001) X-ray absorption spectroscopic investigation of arsenite and arsenate adsorption at the aluminum oxide-water interface. J Colloid Interface Sci 235:80-88

Arai Y, Lanzirotti A, Sutton S, Davis J, Sparks D (2003) Arsenic speciation and reactivity in poultry litter. Environ Sci Technol 37:4083-4090

ATSDR (2005a) Toxicological Profile for Arsenic. U.S. Department of Health and Human Services-Agency for Toxic Substances and Disease Registry, *http://www.atsdr.cdc.gov/toxprofiles/tp2.html*

ATSDR (2005b) Toxicological Profile for lead. (Draft for Public Comment). U.S. Department of Health and Human Services-Agency for Toxic Substances and Disease Registry, *http://www.atsdr.cdc.gov/toxprofiles/tp13.html*

ATSDR (2005c) Toxicological profile for tin and tin compounds U.S. Department of Health and Human Services-Agency for Toxic Substance and Disease Registry, *http://www.atsdr.cdc.gov/toxprofiles/tp55.html*

ATSDR (2000) Toxicological Profile for Chromium. U.S. Department of Health and Human Services-Agency for Toxic Substances and Disease Registry, *http://www.atsdr.cdc.gov/toxprofiles/tp7.html*

Badmaev V, Prakash S, Majeed M (1999) Vanadium: a review of its potential role in the fight against diabetes. J Altern Complement Med 5:273-291
Baecklund M, Pedersen NL, Bjorkman L, Vahter M (1999) Variation in blood concentrations of cadmium and lead in the elderly. Environ Res 80:222-230
Bannon DI, Chisolm JJ (2001) Anodic stripping voltammetry compared with graphite furnace atomic absorption spectrophotometry for blood lead analysis. Clin Chem 47:1703-1704
Barceloux DG (1999a) Molybdenum. J Toxicol Clin Toxicol 37:231-237
Barceloux DG (1999b) Nickel. J Toxicol Clin Toxicol 37:239-258
Barry PS (1981) Concentrations of lead in the tissues of children. Br J Ind Med 38:61-71
Barry PS (1975) Letter: Lead levels in blood. Nature 258:775-775
Bartlett RJ, James BR (1993) Redox chemistry of soils. Adv Agronomy 50:151-208
Baxter D, Frech W (1995) Speciation of lead in environmental and biological samples. Pure Appl Chem 67: 615-648
Bernhard M, Brinckman FE, Irgolic KJ (1986) Why Speciation? *In:* The Importance Of Chemical Speciation in Environmental Processes. Bernhard M, Brinckman FE, Sadler PJ (eds) Springer-Verlag, p 7-14
Bethke CM (2002) The Geochemist's Workbench - release 4.0, A user's guide to Rxn, Act2, Tact, React and Gtplot, University of Illinois
Blake KC, Mann M (1983) Effect of calcium and phosphorus on the gastrointestinal absorption of ^{203}Pb in man. Environ Res 30:188-194
Blake KCH, Barbezat GO, Mann M (1983) Effect of dietary constituents on the gastrointestinal absorption of ^{203}Pb in man. Environ Res 30:182-187
Bothe JV, Brown PW (1999) Arsenic immobilization by calcium arsenate formation. Environ Sci Technol 33: 3806-3811
Bouton CM, Frelin LP, Forde CE, Arnold Godwin H, Pevsner J (2001) Synaptotagmin I is a molecular target for lead. J Neurochem 76:1724-1735
Bragt PC, van Dura EA (1983) Toxicokinetics of hexavalent chromium in the rat after intratracheal administration of chromates of different solubilities. Ann Occup Hyg 27:315-322
Bright DA, Dodd M, Reimer KJ (1996) Arsenic in subarctic lakes influenced by gold mine effluent: the occurrence of organoarsenicals and arsenic. Sci Total Environ 180:165-182
Brown GE, Foster AL, Ostergren JD (1999) Mineral surfaces and bioavailability of heavy metals: A molecular-scale perspective. Proc Nat Acad Sci 96:3388-3395
Brown GE, Parks GA (2001) Sorption of trace elements on mineral surfaces: Modern perspectives from spectroscopic studies, and comments on sorption in the marine environment. Int Geol Rev 43:963-1073
Brown GE (1990) Spectroscopic studies of chemisorption reaction mechanisms at oxide-water interfaces. Rev Mineral 23:309-363
Buchet JP, Lauwerys R, Roels H (1981) Comparison of the urinary excretion of arsenic metabolites after a single oral dose of sodium arsenite, monomethylarsonate or dimethylarsinate in man. Int Arch Occup Environ Health 48:71-79
Buerge IJ, Hug SJ (1999) Influence of mineral surfaces on chromium(VI) reduction by iron(II). Environ Sci Technol 33:4285-4291
Campbell PGC (1995) Interactions between trace metals and aquatic organisms: A critique of the free-ion activity model. *In*: Metal Speciation and Bioavailability in Aquatic Systems. Tessier A, Turner DR (eds) John Wiley and Sons, 45-102
Caruso JA, Klaue B, Michalke B, Rocke DM (2003) Group assessment: elemental speciation. Ecotox Environ Safety 56:32-44
Casteel SW, Weis CP, Henningsen GM, Brattin WJ (2006) Estimation of relative bioavailability of lead in soil and soil-like materials using young swine. Environ Health Perspect 114:1162–1171
CDC (1985) Preventing lead poisoning in young children, a statement by the Centers for Disease Control. US Department of Health and Human Services, 44, *http://aepo-xdv-www.epo.cdc.gov/wonder/prevguid/p0000029/p0000029.asp*
Chamberlain A, Heard C, Little MJ (1978) Investigations into lead from motor vehicles. Harwell, United Kingdom. Phil Trans Royal Soc London A 290:557-589
Charlet L, Manceau A (1993) Structure, formation and reactivity of hydrous oxide particles; insights from X-ray absorption spectroscopy. *In:* Environmental Particles. Buffle J, Leeuwen HPV (eds) Lewis Publishers, p 117-164
Christl I, Metzger A, Heidmann I, Kretzschmar R (2005) Effect of humic and fulvic acid concentrations and ionic strength on copper and lead binding. Environ Sci Technol 39:5319-5326
Cornelis R, Caruso J, Crews H, Heumann K (2003) Handbook of Elemental Speciation, Wiley
Corrin ML, Natusch DFS (1977) Physical and chemical characteristics of environmental lead. *In*: Lead in the Environment. Boggess WR, Wixson BG (eds) National Science Foundation, p 7-31
Cotter-Howells J (1996) Lead phosphate formation in soils. Environ Pollution 93:9-16

Cotter-Howells J, Champness P, Chamock J, Pattrick R (1994) Identification of pyromorphite in mine-waste contaminated soils by ATEM and EXAFS. Eur J Soil Sci 45:393-402

Counter SA (2003) Neurophysiological anomalies in brainstem responses of mercury-exposed children of Andean gold miners. J Occup Environ Med 45:87-95

Cullen WR, Reimer KJ (1989) Arsenic speciation in the environment. Chem Rev 89:713-764

D'Amore JJ, Al-Abed SR, Scheckel KG, Ryan JA (2005) Methods for speciation of metals in soils: A review. J Environ Qual 34:1707–1745

Davies BE, Bowman C, Davies TC, Selinus O (2005) Medical geology: perspectives and prospects. *In*: Essentials of Medical Geology. Selinus O (ed) Elsevier, p 1-14

Davis A, Drexler JW, Ruby MV, Nicholson A (1993) Micromineralogy of mine wastes in relation to lead bioavailability, Butte. Environ Sci Technol 27:1415-1425

Davis JA, Kent DB (1990) Surface complexation modeling in aqueous geochemistry. Rev Mineral Geochem 23:177-260

Dawson DC, Ballatori N (1995) Membrane transporters as sites of action and routes of entry for toxic metals. *In*: Toxicology of metals: biochemical aspects. Goyer RA, Cherian MG (eds) Springer-Verlag, p 53-76

De Flora S, Badolati GS, Serra D (1987) Circadian reduction of chromium in the gastric environment. Mutation Research 192:169-174

de Mora SJ (1996) The tributyltin debate: ocean transportation versus seafood harvesting. *In*: Tributyltin: Case Study of an Environmental Contaminant. de Mora SJ (ed) Cambridge University Press, p 1-20

De Voss JJ, Rutter K, Schroeder BG, Barry CEI (1999) Iron acquisition and metabolism by mycobacteria. J Bacteriol 181:4443-4451

DeJonghe WRA, Adams FC (1986) Biogeochemical cycling of organic lead compounds. Adv Environ Sci Technol 17:561-594

Diez S, Abalos M, Bayona JM (2002) Organotin contamination in sediments from the Western Mediterranean enclosures following 10 years of TBT regulation. Water Res 36:905-918

DOE (1995) Molecular Environmental Science: Speciation, Reactivity, and Mobility of Environmental Contaminants: An Assessment of Research Opportunities and the Need for Synchrotron Radiation Facilities. Stanford Synchrotron Radiation Laboratory, SLAC-R-477

Dolbec J, Mergler D, Sousa Passos CJ, Sousa de Morais S, Lebel J (2000) Methylmercury exposure affects motor performance of a riverine population of the Tapajós river, Brazilian Amazon. Int Arch Occup Environ Health 73:195-203

Dopp E, Hartmann LM, Florea AM, Rettenmeier AW, Hirner AV (2004) Environmental distribution, analysis, and toxicity of organometal(loid) compounds. Crit Rev Toxicol 34:301-333

Duffus JH (2002) "Heavy metals"—A meaningless term? Pure Appl Chem 74:793–807

Dumont E, Ogra Y, Vanhaecke F, Suzuki KT, Cornelis R (2006) Liquid chromatography-mass spectrometry (LC-MS): a powerful combination for selenium speciation in garlic (Allium sativum). Anal Bioanal Chem 384:1196-1206

Eary LE, Rai D (1988) Chromate removal from aqueous wastes by reduction with ferrous ion. Environ Sci Technol 22:972-977

Ehlers LJ, Luthy RG (2003) Contaminant bioavailability in soil and sediment. Environ Sci Technol 37:295A-302A

Eisenreich SJ, Looney BB, Thornton JD (1981) Airborne organic contaminants in the Great Lakes ecosystem. Environ Sci Technol 15:30-38

Eisler R (2004) Mercury hazards from gold mining to humans, plants, and animals. Rev Environ Contam Toxicol 181:139-198

Elbaz-Poulichet F, Holliger P, Huang WW (1984) Lead cycling in estuaries, illustrated by the Gironde Estuary, France. Nature 308:409-414

Elinder CG, Gerhardsson L, Oberdörster G (1988) Biological monitoring of toxic metals — Overview. *In:* Biological Monitoring of Toxic Metals. Clarkson TW, Friberg L, Nordberg GF, Sager PR (eds), Plenum Press, p 1-72

Ellenhorn M, Barceloux D (1988) Medical Toxicology: Diagnosis and Treatment of Human Poisoning. Elsevier

Elzinga EJ, Reeder RJ (2002) X-ray absorption spectroscopy study of Cu^{2+} and Zn^{2+} adsorption complexes at the calcite surface. Geochim Cosmochim Acta 66:3943-3954

EPA (2005) Estimation of Relative Bioavailability of Arsenic in Soil and Soil-Like Materials by In Vivo and In Vitro Methods - USEPA Review Draft. U.S. Environmental Protection Agency, Region 8, Denver, CO

EPA (1998a) Toxicological Review of Hexavalent Chromium. U.S. Environmental Protection Agency

EPA (1998b) Toxicological Review Of Trivalent Chromium. U.S. Environmental Protection Agency

EPA (1996) U.S. Environmental Protection Agency, Federal Registry # 61:3832

EPA (1986) Air Quality Criteria for Lead. U.S. Environmental Protection Agency, Office of Research and Development, Office of Health and Environmental Assessment, Environmental Criteria and Assessment Office

EPA (1985) Lead Exposures in the Human Environment. Environmental Criteria and Assessment Office, U.S. Environmental Protection Agency

Fendorf S, Wielinga BW, Hansel CM (2000) Chromium transformations in natural environments: The role of biological and abiological processes in chromium(VI) reduction. Int Geol Rev 42:691-701

Fendorf SE (1995) Surface reactions of chromium in soils and waters. Geoderma 67:55-71

Fendorf SE, Fendorf M, Sparks DL, Gronsky R (1992) Inhibitory mechanisms of Cr(III) oxidation by δ-MnO_2. J Colloid Interface Sci 153:37-54

Feroci G, Fini A, Fazio G (1995) Interaction between dihydroxy bile salts and divalent heavy metal ions studied by polarography. Anal Chem 67:4077-4085

Finley BL, Scott PK, Norton RL (1996) Urinary chromium concentrations in humans following ingestion of safe doses of hexavalent and trivalent chromium: Implications for biomonitoring. J Toxicol Environ Health 48:479-499

Fitsanakis VA, Aschner M (2005) The importance of glutamate, glycine, and gamma-aminobutyric acid transport and regulation in manganese, mercury and lead neurotoxicity. Toxicol Appl Pharmacol 204: 343-354

Fitzgerald WF, Lamborg CH (2004) Geochemistry of mercury in the environment. *In*: Treatise on Geochemistry. Lollar SB (ed) Elsevier, p 107-148

Fordyce F (2005) Selenium deficiency and toxicity in the environment. *In*: Essentials of Medical Geology. Selinus O (ed) Elsevier, p 373-415

Forte G, D'Amato M, Caroli S (2005) Capillary electrophoresis speciation analysis of various arsenical compounds. Microchem J 79:15-19

Foster AL (2003) Spectroscopic investigations of arsenic species in solid phases. *In*: Arsenic in Ground Water. Welch AH, Stollenwerk KG (eds) Kluwer Academic Publishers, p 27-65

Foster AL, Brown GE, Parks GA (1998) X-ray absorption fine-structure spectroscopy study of photocatalyzed, heterogeneous As(III) oxidation on kaolin and anatase. Environ Sci Technol 32:1444-1452

Foulkes EC (2000) Transport of toxic heavy metals across cell membranes. Proc Soc Exp Biol Med 223:234-240

Fredrickson JK, Zachara JM, Kennedy DW, Duff MC, Gorby YA, Li S-mW, Krupka KM (2000) Reduction of U(VI) in goethite (α-FeOOH) suspensions by a dissimilatory metal-reducing bacterium. Geochim Cosmochim Acta 64:3085-3098

Freeman GB, Dill JA, Johnson JD, Kurtz PJ, Parham F, Matthews HB (1996) Comparative absorption of lead from contaminated soil and lead salts by weanling Fischer 344 rats. Fundam Appl Toxicol 33:109-119

Fuller CC, Davis JA, Waychunas GA (1993) Surface chemistry of ferrihydrite: Part 2. Kinetics of arsenate adsorption and coprecipitation. Geochim Cosmochim Acta 57:2271-2282

Gaetke LM, Chow CK (2003) Copper toxicity, oxidative stress, and antioxidant nutrients. Toxicology 189: 147-163

Gaggelli E, Berti F, D'Amelio N, Gaggelli N, Valensin G, Bovalini L, Paffetti A, Trabalzini L (2002) Metabolic pathways of carcinogenic chromium. Environ Health Perspect Suppl 110:733-738

Gailer J, George GN, Pickering IJ, Prince RC, Ringwald SC, Pemberton JE, Glass RS, Younis HS, DeYoung DW, Aposhian HV (2000) A metabolic link between arsenite and selenite: The seleno-bis(*S*-glutathionyl) arsenium ion. J Am Chem Soc 122:4637-4639

Gailer J, George GN, Pickering IJ, Prince RC, Younis HS, Winzerling JJ (2002) Biliary excretion of $[(GS)_2AsSe]^-$ after intravenous injection of rabbits with arsenite and selenate. Chem Res Toxicol 15: 1466-1471

Garcia JJ, Martínez-Ballarín E, Millán-Plano S, Allué JL, Albendea C, Fuentes L, Escanero JF (2005) Effects of trace elements on membrane fluidity. J Trace Elem Med Biol 19:19-22

Gargas ML, Norton RL, Paustenbach DJ, Finley BL (1994) Urinary excretion of chromium by humans following ingestion of chromium picolinate. Implications for biomonitoring. Drug Metab Dispos 22: 522-529

Gilman A, Rall T, Nies A, Taylor P (1990) Goodman and Gilman's The Pharmacological Basis of Therapeutics. Pergamon Press

Glew DN, Hames DA (1971) Aqueous nonelectrolyte solutions. Part X. Mercury solubility in water. Canadian J Chem 49:3114-3118

Godwin H (2001) The biological chemistry of lead. Curr Opinion Chem Biol 5:223-227

Goldberg S (2002) Competitive adsorption of arsenate and arsenite on oxides and clay minerals. Soil Sci Soc Am J 66:413-421

Goldhaber SB (2003) Trace element risk assessment: essentiality vs. toxicity. Reg Toxicol Pharmacol 38: 232-242

Greger M, Wang Y, Neuschutz C (2005) Absence of Hg transpiration by shoot after Hg uptake by roots of six terrestrial plant species. Environ Pollut 134:201-208

Gruebel K, Davis J, Leckie J (1988) The feasibility of using sequential extraction techniques for arsenic and selenium in soils and sediments. Soil Sci Soc Am J 52:390-397

Guidotti TL (2005) Toxicology. *In:* Essentials of Medical Geology. Selinus O (ed) Elsevier, p 595-608
Gurzau ES, Neagu C, Gurzau AE (2003) Essential metals--case study on iron. Ecotoxicol Environ Safety 56: 190-200
Hartikainen H (2005) Biogeochemistry of selenium and its impact on food chain quality and human health. J Trace Elements Med Biol 18:309-318
Heard MJ, Chamberlain AC (1982) Effect of minerals and food on uptake of lead from the gastrointestinal tract in humans. Human Toxicol 1:411-415
Heard MJ, Wells AC, Newton D (1979) Human uptake and metabolism of tetraethyl and tetramethyl lead vapour labeled with ^{203}Pb. *In:* International Conference on Management and Control of Heavy Metals in the Environment. CEP Consultants, Ltd., p 103-108
Hering JG, Kneebone PE (2002) Biogeochemical controls on arsenic occurrence and mobility in water supplies. *In:* Environmental Chemistry of Arsenic. Frankenberger WT (ed) Marcel Dekker, p 155-181
Hingston FJ (1981) A review of anion adsorption. *In:* Adsorption of Inorganics at Solid-Liquid Interfaces. Anderson MA, Rubin AJ (eds) Ann Arbor Science, p 51-90
Hochella MF, White AF (1990) Mineral-Water Interface Geochemistry. Reviews in Mineralogy, Vol. 23. Mineralogical Society of America
Hong SM, Candelone JP, Patterson CC, Boutron CF (1994) Greenland ice evidence of hemispheric lead pollution 2-millennia ago by Greek and Roman civilizations. Science 265:1841-1843
Hudson RJM (1998) Which aqueous species control the rates of trace metal uptake by aquatic biota? Observations and predictions of non-equilibrium effects. Sci Total Environ 219:95-115
Hylander LD, Pinto FN, Guimarães JR, Meili M, Oliveira LJ, de Castro e Silva E (2000) Fish mercury concentration in the Alto Pantanal, Brazil: influence of season and water parameters. Sci Total Environ 261:9 - 20
Ilton ES, Veblen DR (1994) Chromium sorption by phlogopite and biotite in acidic solutions at 25 °C: Insights from X-ray photoelectron spectroscopy and electron microscopy. Geochim Cosmochim Acta 58:2777-2788
Inskeep WP, McDermott TR, Fendorf S (2002) Arsenic (V)/(III) cycling in soils and natural waters: Chemical and microbial processes. *In:* Environmental Chemistry of Arsenic. Frankenberger WT (ed) Marcel Dekker, p 183-215
IPCS (2001) Arsenic and Arsenic Compounds. *In:* Environmental Health Criteria 224. World Health Organization
IPCS (1990) Methylmercury. *In:* Environmental Health Criteria 101. World Health Organization
IPCS (1988) Chromium. *In:* Environmental Health Criteria 61. World Health Organization
Jackson B, Williams P, Lanzirotti A, Bertsch P (2005) Evidence for biogenic pyromorphite formation by the nematode Caenorhabditis elegans. Environ Sci Technol 39:5620-5625
Jain A, Loeppert RH (2000) Effect of competing anions on the adsorption of arsenate and arsenite by ferrihydrite. J Environ Qual 29:1422-1430
James BR, Bartlett RJ (1983a) Behavior of chromium in soils. VI. Interactions between oxidation-reduction and organic complexation. J Environ Qual 12:173-176
James BR, Bartlett RJ (1983b) Behavior of chromium in soils. VII. Adsorption and reduction of hexavalent forms. J Environ Qual 12:177-181
James HM, Hilburn ME, Blair JA (1985) Effects of meals and meal times on uptake of lead from the gastrointestinal tract in humans. Human Toxicol 4:401-407
Jeremiason JD, Engstrom DR, Swain EB, Nater EA, Johnson BM, Almendinger JE, Monson BA, Kolka RK (2006) Sulfate addition increases methylmercury production in an experimental wetland. Environ Sci Technol 40:3800-3806
Johnson CC, Ge X, Green KA, Liu X (2000) Selenium distribution in the local environment of selected villages of the Keshan Disease belt, Zhangjiakou District, Hebei Province, People's Republic of China. Appl Geochem 15:385-401
Juillot F (1998) Localisation et speciation de l'arsenic, du plomb et du zinc dans des sites et sols contaminés. Comparaison avec un sol développé sur une anomalie géochimique naturelle en plomb. Ph.D. Dissertation, Université Paris, Paris
Kafer A, Zoltzer H, Krug HF (1992) The stimulation of arachidonic acid metabolism by organic lead and tin compounds in human HL-60 leukemia cells. Toxicol Appl Pharmacol 116:125-132
Kimbrough DE, Cohen Y, Winer AM, Creelman L, Mabuni C (1999) A critical assessment of chromium in the environment. Crit Rev Environ Sci Technol 29:1-46
Köhrl J, Brigelius-Flohé R, Böck A, Gärtner R, Meyer O, Flohé L (2000) Selenium in biology: facts and medical perspectives. Biol Chem 381:849-864
Lampert JK (1982) Measurement of trace cation activities by Donnan membrane equilibrium and atomic adsorption analysis. Ph.D. Dissertation, University of Wisconsin, Madison
Langford N, Ferner R (1999) Toxicity of mercury. J Hum Hypertens 13:651-656

Le XC (2002) Arsenic speciation in the environment and humans. *In:* Environmental Chemistry of Arsenic. Frankenberger WT (ed) Marcel Dekker, p 95-116

Le XC, Ma M, Lu X, Cullen WR, Aposhian HV, Zheng B (2000) Determination of monomethylarsonous acid, a key arsenic methylation intermediate, in human urine. Environ Health Perspect 108:1015-1018

Levander OA (1977) Metabolic interrelationships between arsenic and selenium. Environ Health Perspect 19: 159-164

Levina A, Zhang L, Lay PA (2003) Structure and reactivity of a chromium(V) glutathione complex. Inorg Chem 42:767-784

Liger E, Charlet L, Cappellen PV (1999) Surface catalysis of uranium(VI) reduction by iron(II). Geochim Cosmochim Acta 63:2939-2955

Lin JH, Lu AYH (1997) Role of pharmacokinetics and metabolism in drug discovery and development. Pharmacol Rev 49:403-449

Lindh U (2005) Biological function of the elements. *In:* Essentials of Medical Geology. Selenius O, Alloway B, Centeno JA, Finkelman RB, Fuge R, Lindh U, Smedley P (eds) Elsevier Academic Press, p 115-160

Lodenius M, Malm O (1998) Mercury in the Amazon. Rev Environ Contam Toxicol 157:25-52

Long DT, Angino EE (1977) Chemical speciation of Cd, Cu, Pb, and Zn in mixed freshwater, seawater, and brine solutions. Geochim Cosmochim Acta 41:1183-1191

Lovley DR, Phillips EJP, Gorby YA, Landa ER (1991) Microbial reduction of uranium. Nature 350:413-416

Maddaloni M, Lolacono N, Manton W, Blum C, Drexler J, Graziano J (1998) Bioavailability of soilborne lead in adults, by stable isotope dilution. Environ Health Perspect Suppl 106:1589-1594

Maguire RJ (1996) The occurrence, fate, and toxicity of tributyltin and its degradation products in freshwater environments. *In:* Tributyltin: Case Study of an Environmental Contaminant. de Mora SJ (ed) Cambridge University Press, p 94-138

Malm O (1998) Gold mining as a source of mercury exposure in the Brazilian Amazon. Environ Res 77:73-78

Manceau A, Lanson B, Drits VA (2002) Structure of heavy metal sorbed birnessite. Part III: Results from powder and polarized extended X-ray absorption fine structure spectroscopy. Geochim Cosmochim Acta 66:2639-2663

Manceau A, Marcus M, Tamura N (2003) Quantitative speciation of heavy metals in soils and sediments by synchrotron X-ray techniques. Rev Mineral Geochem 49:341-428

Manning BA, Fendorf SE, Bostick B, Suarez DL (2002) Arsenic(III) oxidation and arsenic(V) adsorption reactions on synthetic birnessite. Environ Sci Technol 36:976-981

Manning BA, Goldberg S (1997) Adsorption and stability of arsenic(III) at the clay mineral-water interface. Environ Sci Technol 31:2005-2011

Matocha CJ, Elzinga EJ, Sparks DL (2001) Reactivity of Pb(II) at the Mn(III,IV) (oxyhydr)oxide-water interface. Environ Sci Technol 35:2967-2972

Maurice-Bourgoin L, Quiroga I, Chincheros J, Courau P (2000) Mercury distribution in waters and fishes of the upper Madeira rivers and mercury exposure in riparian Amazonian populations. Sci Total Environ 260:73-86

Mertz W, Roginski EE (1971) Chromium metabolism: The glucose tolerance factor. *In:* Newer Trace Elements in Nutrition. Mertz W, Cornatzer WE (eds) Dekker, p 123–153

Mihaljevič M, Poňavič M, Ettler V, Šebek O (2003) A comparison of sequential extraction techniques for determining arsenic fractionation in synthetic mineral mixtures. Anal Bioanal Chem 377:723-729

Moon DH, Dermatas D, Menounou N (2004) Arsenic immobilization by calcium-arsenic precipitates in lime treated soils. Sci Total Environ 330:171-185

Morel FMM (1983) Principles of Aquatic Chemistry. Wiley-Interscience

Morin G, Juillot F, Ildefonse P, Calas G, Samama J, Chevallier P, Brown G (2001) Mineralogy of lead in a soil developed on a Pb-mineralized sandstone (Largentière, France). Am Mineral 86:92–104

Morin G, Ostergren J, Juillot F, Ildefonse P, Calas G, Brown G (1999) XAFS determination of the chemical form of lead in smelter-contaminated soils and mine tailings: Importance of adsorption processes. Am Mineral 84:420-434

Mota AM, Rato A, Brazia C, Goncalves MLS (1996) Competition of Al^{3+} in complexation of humic matter with Pb^{2+}: A comparative study with other ions. Environ Sci Technol 30:1970-1974

Mundell JA, Hill KR, Weaver JW (1989) In situ case history: leachable lead required precipitation immobilization. Hazard Waste Manage 12:23-27

Myneni SCB, Traina SJ, Logan TJ, Waychunas GA (1997) Oxyanion behavior in alkaline environments: Sorption and desorption of arsenate in ettringite. Environ Sci Technol 31:1761-1768

Nerin C, Domeno C, Garcia JI, et al. (1999) Distribution of Pb, V, Cr, Ni, Cd, Cu and Fe in particles formed from the combustion of waste oils. Chemosphere 38:1533-1540

Newton K, Amarasiriwardena D, Xing B (2006) Distribution of soil arsenic species, lead and arsenic bound to humic acid molar mass fractions in a contaminated apple orchard. Environ Pollut 143:197-205

Nordstrom DK, Archer DG (2003) Arsenic thermodynamic data and environmental geochemistry. *In:* Arsenic in Ground Water. Welch AH, Stollenwerk KG (eds) Kluwer Academic Publishers, p 1-25
Norling P, Wood-Black F, Masciangioli TM (2004) Water and Sustainable Development. National Academies Press
NRC (2003) Bioavailability of Contaminants in Soils and Sediments: Processes Tools, and Applications. National Academies Press
NRC (2001) Arsenic in Drinking Water: 2001 Update. National Academies Press
NRC (2000) Toxicological Effects of Methylmercury. National Academies Press
NRC (1999) Arsenic in Drinking Water. National Academies Press
Nriagu J (1984) Formation and stability of base metal phosphates in soils and sediments. *In:* Phosphate Minerals. Nriagu JO, Moore PB (eds) Springer-Verlag, p 318-329
Nriagu JO (1983) Occupational exposure to lead in ancient-times. Sci Total Environ 31:105-116
Nriagu JO (1974) Lead orthophosphates-IV. Formation and stability in the environment. Geochim Cosmochim Acta 38:887-898
Nriagu JO (1973) Lead orthophosphates-II. Stability of chloropyromorphite at 25 °C. Geochim Cosmochim Acta 37:367-377
Nriagu JO, Pacyna JM (1988) Quantitative assessment of worldwide contamination of air, water and soils by trace metals. Nature 333:134-139
NSF (1977) Transport and distribution in a watershed ecosystem. *In:* Lead in the environment: Chapter 6. Boggess W (ed) National Science Foundation, p 105-133
O'Day PA (2006) Chemistry and mineralogy of arsenic. Elements 2:77-83
O'Day PA (1999) Molecular environmental geochemistry. Rev Geophys 37:249–274
O'Flaherty EJ (1998) Physiologically based models of metal kinetics. Crit Rev Toxicol 28:271-317
O'Flaherty EJ (1996) A physiologically based model of chromium kinetics in the rat. Toxicol Appl Pharmacol 138:54-64
O'Flaherty EJ, Kerger BD, Hays SM, Paustenbach DJ (2001) A physiologically based model for the ingestion of chromium(III) and chromium(VI) by humans. Toxicol Sci 60:196-213
O'Reilly SE, Hochella MF (2003) Lead sorption efficiencies of natural and synthetic Mn and Fe-oxides. Geochim Cosmochim Acta 67:4471–4487
Oomen AG, Tolls J, Sips AJAM, Groten JP (2003a) In vitro intestinal lead uptake and transport in relation to speciation. Arch Environ Contam Toxicol 44:116-124
Oomen AG, Tolls J, Sips AJAM, Van den Hoop MAGT (2003b) Lead speciation in artificial human digestive fluid. Arch Environ Contam Toxicol 44:107-115
Oteiza PI, Mackenzie GG (2005) Zinc, oxidant-triggered cell signaling, and human health. Mol Aspects Med 26:245-255
Pacyna EG, Pacyna JM (2002) Global emission of mercury from anthropogenic sources in 1995. Water, Air, Soil Pollution 137:149-165
Parkhurst DL, Appelo CAJ (1999) User's guide to PHREEQC (version 2)--A computer program for speciation, batch-reaction, one-dimensional transport, and inverse geochemical calculations. Report 99-4259, US Geological Survey
Patterson RR, Fendorf S, Fendorf M (1997) Reduction of hexavalent chromium by amorphous iron sulfide. Environ Sci Technol 31:2039-2044
Peraza MA, Ayala-Fierro F, Barber DS, Casarez E, Rael LT (1998) Effects of micronutrients on metal toxicity. Environ Health Perspect Suppl 106:203-216
Peterson ML, Brown GE, Parks GA (1996) Direct XAFS evidence for heterogeneous redox reaction at the aqueous chromium/magnetite interface. Colloids Surf A 107:77-88
Peterson ML, Brown GE, Parks GA, Stein CL (1997) Differential redox and sorption of Cr (III/VI) on natural silicate and oxide minerals: EXAFS and XANES results. Geochim Cosmochim Acta 61:3399-3412
Pinheiro JP, Mota AM, van Leeuwen HP (1999) On lability of chemically heterogeneous systems: Complexes between trace metals and humic matter. Colloids Surf A 151:181-187
Plant JA, Kinniburgh DG, Smedley PL, Fordyce FM, Klinck BA (2005) Arsenic and Selenium. *In:* Environmental Geochemistry. Lollar BS (ed) Elsevier, p 17-66
Plumlee GS, Morman SA, Ziegler TL (2006) The toxicological geochemistry of earth materials: an overview of processes and the interdisciplinary methods used to understand them. Rev Mineral Geochem 64:5-57
Plumlee GS, Ziegler TL (2003) The medical geochemistry of dusts, soils and other earth materials. *In:* Treatise on Geochemistry. Lollar BS (ed) Elsevier, p 263-310
Pomroy C, Charbonneau SM, McCullough RS, Tam GKH (1980) Human retention studies with ^{74}As. Toxicol Appl Pharmacol 53:550-556
Rabinowitz MB, Kopple JD, Wetherill GW (1980) Effect of food intake and fasting on gastrointestinal lead absorption in humans. Am J Clin Nutrit 33:1784-1788
Rai D, Eary LE, Zachara JM (1989) Environmental chemistry of chromium. Sci Total Environ 86:15-23

Rai D, Sass BM, Moore DA (1987) Chromium(III) hydrolysis constants and solubility of chromium(III) hydroxide. Inorg Chem 26:345-349
Reddy KJ, Wang L, Gloss SP (1995) Solubility and mobility of copper, zinc and lead in acidic environments. Plant Soil 171:53-58
Reddy MM, Aiken GR (2001) Fulvic acid-sulfide ion competition for mercury ion binding in the Florida Everglades. Water, Air, Soil Pollution 132:89-104
Reeder RJ (1996) Interaction of divalent cobalt, zinc, cadmium, and barium with the calcite surface during layer growth. Geochim Cosmochim Acta 60:1543-1552
Renner R (2004) Plumbing the depths of D.C.'s drinking water crisis. Environ Sci Technol 38:224A-227A
Richmond WR, Loan M, Morton J, Parkinson GM (2004) Arsenic removal from aqueous solution via ferrihydrite crystallization control. Environ Sci Technol 38:2368-2372
Rochette EA, Bostick BC, Li G, Fendorf S (2000) Kinetics of arsenate reduction by dissolved sulfide. Environ Sci Technol 34:4714-4720
Rosen AL, Hieftje GM (2004) Inductively coupled plasma mass spectrometry and electrospray mass spectrometry for speciation analysis: applications and instrumentation. Spectrochim Acta B 59:135-146
Ruby MV, Davis A, Nicholson A (1994) In situ formation of lead phosphates in soils as a method to immobilize lead. Environ Sci Technol 28:646-654
Ruby MV, Davis A, Schoof R, Eberle S, Sellstone CM (1996) Estimation of lead and arsenic bioavailability using a physiologically based extraction test. Environ Sci Technol 30:422-430
Ruby MV, Schoof R, Brattin W, Goldade M, Post G, Harnois M, Mosby DE, Casteel SW, Berti W, Carpenter M, Edwards D, Cragin D, Chappell W (1999) Advances in evaluating the oral bioavailability of inorganics in soil for use in human health risk assessment. Environ Sci Technol 33:3697-3705
Rudel H (2003) Case study: bioavailability of tin and tin compounds. Ecotoxicol Environ Safety 56:180-189
Ryan JA, Scheckel KG, Berti WR, Brown SL, Casteel SW, Chaney RL, Hallfrisch J, Doolan M, Grevatt P, Maddaloni M, D. M (2004) Reducing children's risk from lead in soil. Environ Sci Technol 38:18A-24A
Sadiq M (1997) Arsenic chemistry in soils: An overview of thermodynamic predictions and field observations. Water, Air, Soil Pollution 93:117-136
Sass BM, Rai D (1987) Solubility of amorphous chromium(III)-iron(III) hydroxide solid solutions. Inorg Chem 26:2228 - 2232
Scheckel KG, Impellitteri CA, Ryan JA, McEvoy T (2003) Assessment of a sequential extraction procedure for perturbed lead-contaminated samples with and without phosphorus amendments. Environ Sci Technol 37:1892-1898
Scheckel KG, Ryan JA (2003) In vitro formation of pyromorphite via reaction of Pb sources with soft-drink phosphoric acid. Sci Total Environ 302:253-265
Scheckel KG, Ryan JA, Allen D, Lescano NV (2005) Determining speciation of Pb in phosphate-amended soils: Method limitations. Sci Total Environ 350:261-272
Scheinost A, Kretzscmar R, Pfister S, Roberts D (2002) Combining selective sequential extractions, X-ray absorption spectroscopy, and principal component analysis for quantitative zinc speciation in soil. Environ Sci Technol 36:5021-5028
Schettler T (2001) Toxic threats to neurologic development of children. Environ Health Perspect Suppl 109: 813-816
Schoolmeester WL, White DR (1980) Arsenic poisoning. Southern Med J 73:198-208
Schoonen MAA, Cohn CA, Roemer E, Laffers R, Simon SR, O'Riordan T (2006) Mineral-induced formation of reactive oxygen species. Rev Mineral Geochem 64:179-221
Schoonen MAA, Strongin DR (2005) Catalysis of electron transfer reactions at mineral surfaces. *In:* Environmental Catalysis. Grassian V (ed) CRC Press, p 37-60
Schuster PF, Krabbenhoft DP, Naftz DL, Cecil LD, Olson ML, Dewild JF, Susong DD, Green JR, Abbott ML (2002) Atmospheric mercury deposition during the last 270 years: A glacial ice core record of natural and anthropogenic sources. Environ Sci Technol 36:2303-2310
Scott N, Hatelid KM, MacKenzie NE, Carter DE (1993) Reactions of arsenic(III) and arsenic(V) species with glutathione. Chem Res Toxicol 6:102-106
Semple KT, Doick KJ, Jones KC, Burauel P, Craven A, Harms H (2004) Defining bioavailability and bioaccessibility of contaminated soil and sediment is complicated. Environ Sci Technol 38:228A-231A
Senko JM, Istok JD, Suflita JM, Krumholz LR (2002) *In situ* evidence for uranium immobilization and remobilization. Environ Sci Technol 36:1491-1496
Shim H, Harris ZL (2003) Genetic defects in copper metabolism. J Nutr 133:1527S-1531S
Smedley PL, Kinniburgh DG (2005) Arsenic in Groundwater and the Environment. *In:* Essentials of Medical Geology. Selinus O (ed) Elsevier, p 263-299
Smedley PL, Kinniburgh DG (2002) A review of the source, behaviour and distribution of arsenic in natural waters. Appl Geochem 17:517-568

Smith KS, Huyck HLO (1999) An overview of the abundance, relative mobility, bioavailability, and human toxicity of metals. *In:* The Environmental Geochemistry of Mineral Deposits: Part A Processes, Techniques and Health Issues. Plumlee GS, Logsdon MJ (eds) Society of Economic Geologists, p 29-70

Sparks DL (2003) Environmental Soil Chemistry, 2nd ed. Academic Press

Spear TM, Svee W, Vincent JH, Stanisich N (1998) Chemical speciation of lead dust associated with primary lead smelting. Environ Health Perspect 106:565-571

Spliethoff HM, Mason RP, Hemond HF (1995) Interannual variability in the speciation and mobility of arsenic in a dimictic lake. Environ Sci Technol 29:2157-2161

Sposito G (2004) The Surface Chemistry of Natural Particles. Oxford University Press

Sposito G (1984) The Surface Chemistry of Soils. Oxford University Press

Steinmaus C, Carrigan K, Kalman D, Atallah R, Yuan Y, Smith AH (2005) Dietary intake and arsenic methylation in a U.S. population. Environ Health Perspect 113:1153-1159

Stevenson F (1994) Humus Chemistry: Genesis, Composition, Reactions. Wiley

Stollenwerk KG (2003) Geochemical processes controlling transport of arsenic in groundwater: A review of adsorption. *In:* Arsenic in Ground Water. Welch AH, Stollenwerk KG (ed) Kluwer Academic Publishers, p 67-100

Stumm W (1992) Chemistry of the Solid-Water Interface. Wiley

Styblo M, Del Razo LM, Vega L, Germolec DR, LeCluyse EL, Hamilton GA, Reed W, Wang C, Cullen WR, Thomas DJ (2000) Comparative toxicity of trivalent and pentavalent inorganic and methylated arsenicals in rat and human cells. Arch Toxicol 74:289-299

Sudaryanto A, Takahashi S, Iwata H, Tanabe S, Ismail A (2004) Contamination of butyltin compounds in Malaysian marine environments. Environ Pollut 130:347-358

Sudaryanto A, Takahashi S, Iwata H, Tanabe S, Muchtar M, Razak H (2005) Organotin residues and the role of anthropogenic tin sources in the coastal marine environment of Indonesia. Marine Pollut Bull 50: 226-235

Sunda WG, Huntsman SA (1998) Processes regulating cellular metal accumulation and physiological effects: Phytoplankton as model systems. Sci Total Environ 219:165-181

Sutton SR, Bertsch PM, Newville M, Rivers M, Lanzirotti A, Eng P (2003) Microfluorescence and microtomography analyses of heterogeneous earth and environmental materials. Rev Mineral Geochem 49:429-483

Tack FM, Verloo MG (1995) Chemical speciation and fractionation in soil and sediment heavy metal analysis: A review. Int J Environ Anal Chem 59:225-238

Tam GKH, Charbonneau SM, Bryce F, Pomroy C, Sandi E (1979) Metabolism of inorganic arsenic (^{74}As) in humans following oral ingestion. Toxicol Appl Pharmacol 50:319-322

Tchounwou PB, Ayensu WK, Ninashvili N, Sutton D (2003) Environmental exposure to mercury and its toxicopathologic implications for public health. Environ Toxicol 18:149-175

Templeton D, Ariese F, Cornelis R, Danielsson L, Muntau H, Leeuwen HV, Lobinski R (2000) Guidelines for terms related to chemical speciation and fractionation of elements. Definitions, structural aspects, and methodological approaches. Pure Appl Chem 72:1453-1470

Tessier A, Campbell PGC, Bisson M (1979) Sequential extraction procedure for the speciation of particulate trace metals. Anal Chem 51:844-851

Tewalt SJ, Bragg LJ, Finkelman RB (2005) Mercury in U.S. Coal—Abundance, Distribution, and Modes of Occurrence. US Geological Survey Fact Sheet 095-01

Tomiyasu T, Matsuo T, Miyamoto J, Imura R, Anazawa K, H. S (2005) Low level mercury uptake by plants from natural environments--mercury distribution in Solidago altissima L. Environ Sci 12:231-238

Traina SJ, Laperche V (1999) Contaminant bioavailability in soils, sediments, and aquatic environments. Proc Nat Acad Sci 96:3365-3371

Tudor R, Zalewski PD, Ratnaike RN (2005) Zinc in health and chronic disease. J Nutr Health Aging 9:45-51

Ullrich SM, Tanton TW, Abdrashitovab SA (2001) Mercury in the aquatic environment: A review of factors affecting methylation. Crit Rev Environ Sci Technol 31:241–293

Uriu-Adams JY, Keen CL (2005) Copper, oxidative stress, and human health. Mol Aspects Med 26:268-298

Vahter M (2002) Mechanisms of arsenic biotransformation. Toxicol 181-182:211-217

Vahter M (2000) Genetic polymorphism in the biotransformation of inorganic arsenic and its role in toxicity. Toxicol Lett 112-113:209-217

Valko M, Morris H, Cronin MTD (2005) Metals, toxicity and oxidative stress. Curr Med Chem 12:1161-1208

Van Leeuwen HP, Town RM, Buffle J, Cleven RFMJ, Davison W, Puy J, van Riemsdijk WH, Sigg L (2005) Dynamic speciation analysis and bioavailability of metals in aquatic systems. Environ Sci Technol 39: 8545-8556

Villalobos M, Bargar J, Sposito G (2005) Mechanisms of Pb(II) sorption on a biogenic manganese oxide. Eviron Sci Technol 39:569-576

Voegelin A, Hug SJ (2003) Catalyzed oxidation of arsenic(III) by hydrogen peroxide on the surface of ferrihydrite: An *in situ* ATR-FTIR study. Environ Sci Technol 37:972-978

Vyskocil A, Viau C (1999) Assessment of molybdenum toxicity in humans. J Appl Toxicol 19:185 - 192

Waite TD, Davis JA, Payne TE, Waychunas GA, Xu N (1994) Uranium(VI) adsorption to ferrihydrite: Application of a surface complexation model. Geochim Cosmochim Acta 58:5465-5478

Walsh CT, Sandstead HH, Prasad AS, Newberne PM, Fraker PJ (1994) Zinc: health effects and research priorities for the 1990s. Environ Health Perspect Suppl 102:5 - 46

Warren LA, Haack EA (2001) Biogeochemical controls on metal behaviour in freshwater environments. Earth-Science Rev 54:261-320

Waychunas GA, Rea BA, Fuller CC, Davis JA (1993) Surface chemistry of ferrihydrite: Part 1. EXAFS studies of the geometry of coprecipitated and adsorbed arsenate. Geochim Cosmochim Acta 57:2251-2269

Wedepohl HK (1995) The composition of the continental crust. Geochim Cosmochim Acta 59:1217-1232

WHO (2003) Chromium in Drinking-water. World Health Organization, 8. *http://www.who.int/water_sanitation_health/dwq/chemicals/chromium.pdf*

Wiegand HJ, Ottenwalder H, Bolt HM (1985) Fast uptake kinetics in vitro of $^{51}Cr(VI)$ by red blood cells of man and rat. Arch Toxicol 57:31-34

Wilkin RT, Wallschlaeger D, Ford RG (2003) Speciation of arsenic in sulfidic waters. Geochem Trans 4:1-7

Winfrey MR, Rudd JWM (1990) Environmental factors affecting the formation of methylmercury in low pH lakes. Environ Toxicol Chem 9:853-869

Xia K, Bleam W, Helmke P (1997) Studies of the nature of Cu^{2+} and Pb^{2+} binding sites in soil humic substances using X-ray absorption spectroscopy. Geochim Cosmochim Acta 61:2211-2221

Yamauchi H, Takahashi K, Yamamura Y (1986) Metabolism and excretion of orally and intraperitoneally administered gallium arsenide in the hamster. Toxicol 40:237-246

Yokoo E, Valente J, Grattan L, Schmidt S, Platt I, Silbergeld E (2003) Low level methylmercury exposure affects neuropsychological function in adults. Environ Health: A Global Access Sci Source 2:8

Zachara JM, Ainsworth CC, Brown GE, Catalano JG, McKinley JP, Qafoku O, Smith SC, Szecsody JE, Traina SJ, Warner JA (2004) Chromium speciation and mobility in a high level nuclear waste vadose zone plume. Geochim Cosmochim Acta 68:13-30

Zachara JM, Ainsworth CC, Cowan CE, Resch CT (1989) Adsorption of chromate by subsurface soil horizons. Soil Sci Soc Am J 53:364-373

Zachara JM, Girvin DC, Schmidt RL, Resch CT (1987) Chromate adsorption on amorphous iron oxyhydroxide in the presence of major groundwater ions. Environ Sci Technol 21:589-594

Zakharyan R, Wu Y, Bogdan GM, Aposhian HV (1995) Enzymatic methylation of arsenic compounds: Assay, partial purification, and properties of arsenite methyltransferase and monomethylarsonic acid methyltransferase of rabbit liver. Chem Res Toxicol 8:1029-1038

Zhang H, Young S (2006) Characterizing the availability of metals in contaminated soils. II. The soil solution. Soil Use Manage 21:459-467

Zhang P, Ryan J, Yang J (1998) In vitro soil Pb solubility in the presence of hydroxyapatite. Environ Sci Technol 32:2763-2768

Zheng W, Aschner M, Ghersi-Egea JF (2003) Brain barrier systems: a new frontier in metal neurotoxicological research. Toxicol Appl Pharmacol 192:1-11

Zhou B, Westaway SK, Levinson B, Johnson MA, Gitschier J, Hayflick SJ (2001) A novel pantothenate kinase gene (PANK2) is defective in Hallervorden-Spatz syndrome. Nat Genet 28:345-349

APPENDIX 1.

Heavy metals and metalloids that are known or thought to be essential for human health.

Metal	Forms in body	Biological Roles	Deficiency-related diseases	Exposure-related diseases
Arsenic	Inorganic forms of arsenic are transformed into methylated arsenic compounds (MMA and DMA). The transformation of As(III) to MMA and DMA takes place primarily in the liver.	Unclear if As is an essential element.	No cases of arsenic deficiency in humans have ever been reported. Animals fed a diet with unusually low concentrations of arsenic did not gain weight normally. They also became pregnant less frequently than animals fed a diet containing a normal amount of arsenic. Furthermore, the offspring from these animals tended to be smaller than normal, and some died at an early age.	Inorganic forms of arsenic are far more toxic than methylated forms. As(III) reacts with sulfhydryl groups in proteins and inactivates enzymes. It is thought that As(III) interferes with DNA repair. As(V) is also genotoxic, but its toxicity may be related to its similarity to phosphate. Arsenic exposure is known to lead to skin lesions, bladder cancer, and neurological diseases. As(III) and As(V) can cross the placenta and cause fetotoxicity, decreased birth weight, and congenital malformations.
Chromium	Cr(III) in the form of a Cr(III)-oligopeptide compound (chromodulin) (Lindh 2005) Cr(VI) is reduced to Cr(III) after uptake.	Required for normal glucose, fat, and protein metabolism	Deficiency leads to impaired glucose tolerance, fasting hyperglycemia, glucosuria, elevated percent body fat, decreased lean body mass, maturity-onset diabetes, cardiovascular disease, decreased sperm count, and impaired fertility.	Cr(VI) enters cells through sulfate channels and reacts with antioxidants. Reactions with glutathione lead to a Cr(V) compound that promotes formation of reactive oxygen species within cell, which promotes carcinogenesis.
Cobalt	Co(II) in several metalloenzymes	Cofactor in vitamin B_{12}, component of metal center in several metalloenzymes	Little documentation of diseases related to lack of Co, except amnesia, lack of red blood cells. Deficiency in vitamin B_{12} may lead to depression and adversely affect the nervous system. .	Inhalation can lead to respiratory irritation, diminished pulmonary function, wheezing, asthma, pneumonia, fibrosis, and lung cancer. Dermal contact can lead to eczema. May cause memory loss and other neurological problems. Cobalt dissolved in cells is thought to promote the formation of reactive oxygen species. Exposure to alloys containing Co and W appear to increase toxicity. Co may also interfere with DNA repair system.

Metal	Forms in body	Biological Roles	Deficiency-related diseases	Exposure-related diseases
Copper	Cu(II)/Cu(I) in 30+ enzymes	Copper in several important enzymes and proteins. For example, Cu is active component in Superoxide Dismutase (SOD), which protects against reactive oxygen species. The Cu-containing protein Ceruloplasmin accounts for 95 % of Cu in human serum.	Copper deficiency is rare. A case involving children in Peru showed that copper deficiency leads to low counts in neutrophils (type of white blood), which puts individuals at a higher risk for bacterial infections (Goldhaber 2003). In addition, other symptoms included anemia and bone mineralization. Animal studies suggest that copper deficiency leads to poor growth, anemia, and degeneration of the central nervous system. Individuals without ceruplasmin protein (a genetic disorder) develop diabetes, retinal degeneration and neurodegeneration (Shim and Harris 2003).	Inhalation can lead to respiratory irritation, including coughing, sneezing, thoracic pain, runny nose, and fibrosis. Exposure to copper dust can lead to headaches, vertigo, and drowsiness. Excessive accumulation as a result of a genetic abnormality is known as Wilson's disease. Excessive copper exposure inhibits zinc absorption and leads to zinc deficiency. Copper is suspected to be a Fenton metal on the basis of its ability to cycle between Cu(II) and Cu(I) oxidation states (Gaetke and Chow 2003; Uriu-Adams and Keen 2005). Excess Cu may play a role in the development of Alzheimer disease (Uriu-Adams and Keen 2005).
Iron	Ferritin protein, which contains a nano-size Fe^{III} oxyhydroxide core, hemoglobin, and several other iron-containing metalloenzymes	Major redox cycling element in body. Hemoglobin is critical to the transfer of oxygen to tissue (Gurzau et al. 2003).	Iron deficiency is common and leads to anemia, or low red blood cell counts, which impairs efficient oxygenation of tissue. Low-iron status leads also to increased uptake of other metals, such as Mn and Ni. Hence, low iron status increases Ni and Mn toxicity for a given exposure.	Acute excessive iron exposure by ingestion leads to vomiting and diarrhea (Goldhaber 2003). Genetically-induced excessive accumulation of iron Hallervorden-Spatz Syndrome leads to hemochromatosis (Zhou et al. 2001). High iron exposure leads to oxidative stress and promotes TB (De Voss et al. 1999).
Manganese	Component of metalloenzymes. Mn present as Mn(II), Mn(III), and Mn(IV) in enzymes.	Mn is an essential nutrient and plays a role in bone mineralization, protein and energy metabolism, metabolic regulation, cellular protection from damaging free radical species, and the formation of glycosaminoglycans.	Mn deficiency is rare. Studies in which human subjects received a low-Mn diet showed slow hair and nail growth, a decrease in clotting proteins, and onset of dermatitis.	High acute exposures lead to manganism, a Parkinson-like, neurological disease.

Metal	Forms in body	Biological Roles	Deficiency-related diseases	Exposure-related diseases
Molybdenum	Mo(IV), Mo(V), Mo(VI)	Co-factor in several enzymes that regulate metabolism of carbon, nitrogen, and sulfur (Barceloux 1999a).	Molybdenum is a trace essential metal and deficiencies are uncommon. Animal studies suggest that Mo deficiency leads to reduced weight gain, decreased food consumption, and impaired reproduction. (Goldhaber 2003)	Little data are available on the human toxicity of molybdenum. A gout-like syndrome (arthritis) and pneumoconiosis (black lung disease) have been associated with excessive concentrations of molybdenum (Barceloux 1999a). Kidney failure has been observed in rats exposed to a high-Mo diet. (Goldhaber 2003). Individuals with inadequate intake of copper are at higher risk for adverse effects of molybdenum exposures (Vyskocil and Viau 1999)
Nickel	Ni-containing enzymes	Component of several metalloenzymes, including Superoxide Dismutase (SOD), which protects against reactive oxygen species.	Trace essential metal. No reports of human nickel deficiency could be located. Rats and chicks on a Ni-deficient diet developed liver problems. (Barceloux 1999b)	Genotoxic metal. Thought to interact with DNA, leading to inhibition of gene expression. Complexation of nickel with low-molecular weight ligands, as well as proteins and peptides, may convert dissolved Ni(II) into a Fenton metal and induce the formation of ROS. Allergic contact dermatitis is a common allergic response among humans to dermal exposure to nickel metal (Barceloux 1999b)
Selenium	Selenoprotein	Antioxidant, Selenoproteins, including glutathione peroxidase	Disturbance of selenoprotein expression or function is associated with deficiency syndromes (Keshan and Kashin-Beck disease), might contribute to tumorigenesis and atherosclerosis, is altered in several bacterial and viral infections, and leads to infertility in male rodents (Köhrl et al. 2000)	High chronic exposure leads to selenosis: symptoms include hair loss and brittle nails.(Goldhaber 2003)
Tin	unknown	Component of gastrine, a stomach-stimulating peptide hormone (Rudel 2003).	No studies addressing Sn deficiencies could be located.	Organotin compounds are lipid soluble and far more toxic than inorganic tin compounds. Exposure to organotin compounds at high dose levels leads to acute neurological effects, such as memory loss. Organotin compounds are thought to cause disruption of cell-signaling in the brain, programmed cell death, and cell death with increasingly high dose exposures. Little is known about the effect of lower dose levels. Dissolved inorganic tin is thought to be a Fenton metal. Inorganic tin is sequestered in bone.

Metal	Forms in body	Biological Roles	Deficiency-related diseases	Exposure-related diseases
Tungsten	W(IV)	Component of some metalloenzymes; role in humans not clear.	No studies addressing W deficiencies could be located.	Tungsten oxide fibers are capable of generating hydroxyl radicals in human lung cells in vitro, thought to contribute to the development of pulmonary fibrosis in hard metal workers.
Vanadium	V(V): VO_3^- V(IV): VO^{2+}	Unclear if V is an essential metal (Badmaev et al. 1999); possibly an antioxidant (Lindh 2005) and may be involved in development of skeleton and teeth.	Rats and chicks showed reduced growth and impairment of reproductive system. V deficiency-related diseases are not known to occur in humans	In rats, mice, and hamsters, it has been established that V(IV) and V(V) compounds are developmental and reproductive toxicants.
Zinc	Zn(II); over 300 known Zn-containing enzymes	Zinc-containing enzymes are important in respiration (SOD), in gene expression, DNA repair, and in programmed cell death (Tudor et al. 2005)	Chronic dietary zinc deficiency is common; it is estimated that 4 million people in the U.S. are affected. Initial symptoms are loss of taste and smell. Chronic severe deficiency leads to immune disorders (Walsh et al. 1994). Zinc deficiency leads to oxidative stress (Oteiza and Mackenzie 2005). Suboptimal zinc status may lead to bronchial asthma, rheumatoid arthritis, and Alzheimer's disease (Tudor et al. 2005).	Highly excessive inhalation exposures can lead to metal fume fever. Excessive Zn exposure may be neurotoxic (Walsh et al. 1994)