3. HEALTH EFFECTS

3.1 INTRODUCTION

The primary purpose of this chapter is to provide public health officials, physicians, toxicologists, and other interested individuals and groups with an overall perspective on the toxicology of arsenic. It contains descriptions and evaluations of toxicological studies and epidemiological investigations and provides conclusions, where possible, on the relevance of toxicity and toxicokinetic data to public health.

A glossary and list of acronyms, abbreviations, and symbols can be found at the end of this profile.

3.2 DISCUSSION OF HEALTH EFFECTS BY ROUTE OF EXPOSURE

To help public health professionals and others address the needs of persons living or working near hazardous waste sites, the information in this section is organized first by route of exposure (inhalation, oral, and dermal) and then by health effect (death, systemic, immunological, neurological, reproductive, developmental, genotoxic, and carcinogenic effects). These data are discussed in terms of three exposure periods: acute (14 days or less), intermediate (15–364 days), and chronic (365 days or more).

Levels of significant exposure for each route and duration are presented in tables and illustrated in figures. The points in the figures showing no-observed-adverse-effect levels (NOAELs) or lowest-observed-adverse-effect levels (LOAELs) reflect the actual doses (levels of exposure) used in the studies. 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. ATSDR acknowledges that a considerable amount of judgment may be required in establishing whether an end point should be classified as a NOAEL, "less serious" LOAEL, or "serious" LOAEL, and that in some cases, there will be insufficient data to decide whether the effect is indicative of significant dysfunction. However, the Agency has established guidelines and policies that are used to classify these end points. ATSDR believes that there is sufficient merit in this approach to warrant an attempt at distinguishing between "less serious" and "serious" effects. The distinction between "less serious" effects and "serious" effects is considered to be important because it helps the users of the profiles to identify levels of exposure at which major health effects start to appear. LOAELs or NOAELs should also help in determining whether or not

the effects vary with dose and/or duration, and place into perspective the possible significance of these effects to human health.

The significance of the exposure levels shown in the Levels of Significant Exposure (LSE) tables and figures may differ depending on the user's perspective. Public health officials and others concerned with appropriate actions to take at hazardous waste sites may want information on levels of exposure associated with more subtle effects in humans or animals (LOAELs) or exposure levels below which no adverse effects (NOAELs) have been observed. Estimates of levels posing minimal risk to humans (Minimal Risk Levels or MRLs) may be of interest to health professionals and citizens alike.

Levels of exposure associated with carcinogenic effects (Cancer Effect Levels, CELs) of arsenic are indicated in Tables 3-1 and 3-3 and Figures 3-1 and 3-3. Because cancer effects could occur at lower exposure levels, Figures 3-1 and 3-3 also show a range for the upper bound of estimated excess risks, ranging from a risk of 1 in 10,000 to 1 in 10,000,000 (10^{-4} to 10^{-7}), as developed by EPA.

A User's Guide has been provided at the end of this profile (see Appendix B). This guide should aid in the interpretation of the tables and figures for Levels of Significant Exposure and the MRLs.

Chemical Forms of Concern. Analysis of the toxic effects of arsenic is complicated by the fact that arsenic can exist in several different oxidation states and many different inorganic and organic compounds. Most cases of human toxicity from arsenic have been associated with exposure to inorganic arsenic, so these compounds are the main focus of this profile.

The most common inorganic arsenical in air is arsenic trioxide (As_2O_3), while a variety of inorganic arsenates (AsO_4^{-3}) or arsenites (AsO_2^-) occur in water, soil, or food. A number of studies have noted differences in the relative toxicity of these compounds, with trivalent arsenites tending to be somewhat more toxic than pentavalent arsenates (Byron et al. 1967; Gaines 1960; Maitani et al. 1987a; Sardana et al. 1981; Willhite 1981). However, these distinctions have not been emphasized in this profile, for several reasons: (1) in most cases, the differences in the relative potency are reasonably small (about 2–3-fold), often within the bounds of uncertainty regarding NOAEL or LOAEL levels; (2) different forms of arsenic may be interconverted, both in the environment (see Section 6.3) and the body (see Section 3.4); and (3) in many cases of human exposure (especially those involving intake from water or soil, which are of greatest concern to residents near wastes sites), the precise chemical speciation is not known.

Gallium arsenide (GaAs) is another inorganic arsenic compound of potential human health concern, due to its widespread use in the microelectronics industry. Available toxicokinetic data suggest that although gallium arsenide is poorly soluble, it undergoes slow dissolution and oxidation to form gallium trioxide and arsenite (Webb et al. 1984, 1986). Therefore, the toxic effects of this compound are expected to be attributable to the arsenite that is liberated, plus the additional effects of the gallium species.

It is beyond the scope of this profile to provide detailed toxicity data on other less common inorganic arsenic compounds (e.g., As_2S_3), but these are expected to be of approximately equal or lesser toxicity than the oxycompounds, depending mainly on solubility (see Section 3.4).

Although organic arsenicals are usually viewed as being less toxic than the inorganics, several methyl and phenyl derivatives of arsenic that are widely used in agriculture are of possible human health concerns based on their toxicity in animal species (Arnold et al. 2003, 2006; NTP 1989b). Chief among these are monomethylarsonic acid (MMA) and its salts (monosodium methane arsonate [MSMA] and disodium methane arsonate [DSMA]), dimethylarsinic acid (DMA, also known as cacodylic acid) and its sodium salt (sodium dimethyl arsinite, or sodium cacodylate), and roxarsone (3-nitro-4-hydroxyphenylarsonic acid). However, it should be noted that food is the largest contributor to background intakes of organic arsenicals. Estimates on the concentration of organic arsenicals in the diet were not located; Cohen et al. (2006) estimated that the intake of DMA from food and water is <1 ng/kg/day. As with the inorganic compounds, there are toxicological differences between these various organic derivatives; because of these differences, the discussion of the health effects of MMA, DMA, and roxarsone are discussed separately. As discussed below, animals do not appear to be good quantitative models for inorganic arsenic toxicity in humans, but it is not known if this also applies to toxicity of organic arsenicals.

Several organic arsenicals are found to accumulate in fish and shellfish. These derivatives (mainly arsenobetaine and arsenocholine, also referred to as "fish arsenic") have been studied by several researchers and have been found to be essentially nontoxic (Brown et al. 1990; Cannon et al. 1983; Charbonneau et al. 1978; Kaise et al. 1985; Luten et al. 1982; Siewicki 1981; Tam et al. 1982; Yamauchi et al. 1986). Thus, these compounds are not considered further here.

Arsine (AsH_3) and its methyl derivatives, although highly toxic, are also not considered in this profile, since these compounds are either gases or volatile liquids that are unlikely to be present at levels of concern at hazardous waste sites.

Use of Animal Data. An additional complexity to the analysis of arsenic toxicity is that most laboratory animals appear to be substantially less susceptible to inorganic arsenic than humans. For example, chronic oral exposure of humans to inorganic arsenic at doses of 0.05–0.1 mg/kg/day is frequently associated with neurological (Barton et al. 1992; Goddard et al. 1992; Guha Mazumder et al. 1988; Haupert et al. 1996; Hindmarsh et al. 1977; Huang et al. 1985; Sass et al. 1993; Silver and Wainman 1952; Szuler et al. 1979; Tay and Seah 1975; Valentine et al. 1981) or hematological signs of arsenic toxicity (Glazener et al. 1968; Guha Mazumder et al. 1988; Prasad and Rossi 1995; Sass et al. 1993; Tay and Seah 1975), but no characteristic neurological or hematological signs of arsenism were detected in monkeys, dogs, or rats chronically exposed to arsenate or arsenite at doses of 0.7–2.8 mg As/kg/day (Byron et al. 1967; EPA 1980f; Heywood and Sortwell 1979). This may be because the studies were not conducted for a sufficient length of time, or because too few animals were used. Moreover, while there is good evidence that inorganic arsenic is carcinogenic in humans by both oral and inhalation routes, evidence of inorganic arsenic-induced carcinogenicity in animals is mostly negative, with the exception of studies in mice demonstrating transplacental carcinogenesis. For these reasons, quantitative dose-response data from animals are not judged to be reliable for determining levels of significant human exposure, and will be considered only briefly except when human data are lacking.

3.2.1 Inhalation Exposure

Most information on human inhalation exposure to arsenic derives from occupational settings such as smelters and chemical plants, where the predominant form of airborne arsenic is arsenic trioxide dust. One limitation to this type of study is that exposure data are usually difficult to obtain, especially from earlier time periods when exposure levels were higher than in recent years. This is further complicated by the fact that significant oral and dermal exposures are also likely to occur under these conditions and co exposure to other metals and chemicals is also common. Thus, studies of this type are, like virtually all epidemiological studies, subject to some limitations and uncertainties. Table 3-1 and Figure 3-1 summarize studies that provide the most reliable quantitative data on health effects in humans, along with several studies in animals exposed to arsenic trioxide and other inorganic arsenic compounds by the inhalation route. Data for DMA are shown in Table 3-2 and Figure 3-2. All exposure data are expressed as milligrams of arsenic (as the element) per cubic meter of air (mg As/m^3). These studies and others that provide useful qualitative information on health effects of inorganic and organic arsenicals are discussed below.

Table 3-1 Levels of Significant Exposure to Inorganic Arsenic - Inhalation

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/m³)	LOAEL Less Serious (mg/m³)	LOAEL Serious (mg/m³)	Reference Chemical Form	Comments
ACUTE EXPOSURE								
Immuno/ Lymphoret								
1	Mouse (CD-1)	3 hr		0.123 F	0.271 F (decreased pulmonary bactericidal activity and increased susceptibility to streptococcal infection)		Aranyi et al. 1985 As(+3)	
2	Mouse (CD-1)	5 d 3 hr/d		0.259 F	0.519 F (decreased pulmonary bactericidal activity and increased susceptibility to streptococcal infection)		Aranyi et al. 1985 As(+3)	
Developmental								
3	Mouse (CFLP)	Gd 9-12 4 hr/d		0.2	2.2 (10% decreased average fetal body weight)	21.6 (increased fetal deaths, skeletal malformations, and retarded growth)	Nagymajtenyi et al. 1985 As(+3)	
INTERMEDIATE EXPOSURE								
Death								
4	Rat (CD)	14 pmd- Gd 19 7 d/wk 6 hr/d				20 F (5/10 dams died)	Holson et al. 1999 As(+3)	
Systemic								
5	Rat (CD)	14 pmd- Gd 19 7 d/wk 6 hr/d	Resp	2 F	8 F (rales, dried red material around nose)		Holson et al. 1999 As(+3)	
			Bd Wt	2 F	8 F (decreased body weight gain during gestation)			

Table 3-1 Levels of Significant Exposure to Inorganic Arsenic - Inhalation (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/m³)	LOAEL Less Serious (mg/m³)	LOAEL Serious (mg/m³)	Reference Chemical Form	Comments
6	Rat (CD)	14 pmd- Gd 19 7 d/wk 6 hr/d	Resp	0.9 F	8 F (rales)	20 F (labored breathing, gasping)	Holson et al. 1999 As(+3)	
			Gastro	8 F		20 F (gross gastrointestinal lesions)		
			Bd Wt	8 F		20 F (drastic decrease body weight)		
Immuno/ Lymphoret								
7	Mouse (CD-1)	4 wk 5 d/wk 3 hr/d		0.126 F	0.245 F (decreased pulmonary bactericidal activity)		Aranyi et al. 1985 As(+3)	
Reproductive								
8	Rat (CD)	14 pmd- Gd 19 7 d/wk 6 hr/d		8 F			Holson et al. 1999 As(+3)	
9	Rat (CD)	14 pmd- Gd 19 7 d/wk 6 hr/d		20 F			Holson et al. 1999 As(+3)	
Developmental								
10	Rat (CD)	14 pmd- Gd 19 7 d/wk 6 hr/d		8			Holson et al. 1999 As(+3)	

Table 3-1 Levels of Significant Exposure to Inorganic Arsenic - Inhalation (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/m³)	LOAEL Less Serious (mg/m³)		LOAEL Serious (mg/m³)		Reference Chemical Form	Comments
11	Rat (CD)	14 pmd- Gd 19 7 d/wk 6 hr/d		8			20	(marked increase in post-implantation loss and marked decrease in viable fetuses)	Holson et al. 1999 As(+3)	
CHRONIC EXPOSURE										
Systemic										
12	Human	23 yr (avg) (occup)	Cardio				0.36 M	(increased incidence of vasospasticity and clinical Raynaud's phenomenon)	Lagerkvist et al. 1986 As(+3)	
13	Human	0.5-50 yr (occup)	Resp	0.613					Perry et al. 1948 As(+3)	
			Dermal		0.078	(mild pigmentation keratosis of skin)	0.613	(gross pigmentation with hyperkeratinization of exposed areas, wart formation)		
Neurological										
14	Human	28 yr (avg) (occup)			0.31 M	(decreased nerve conduction velocity)			Lagerkvist and Zetterlund 1994 As(+3)	
Cancer										
15	Human	1- >30 yr (occup)					0.213 M	(CEL: lung cancer)	Enterline et al. 1987a As(+3)	
16	Human	19.5 yr (avg) (occup)					0.069 M	(CEL: lung cancer)	Enterline et al. 1987b As(+3)	

Table 3-1 Levels of Significant Exposure to Inorganic Arsenic - Inhalation (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/m³)	LOAEL Less Serious (mg/m³)	LOAEL Serious (mg/m³)	Reference Chemical Form	Comments
17	Human	3 mo- >30 yr (occup)				0.2 M (CEL: lung cancer)	Jarup and Pershagen 1991 As(+3)	
18	Human	3 mo- >30 yr (occup)				0.05 M (CEL: lung cancer)	Jarup et al. 1989 As(+3)	
19	Human	1- >30 yr (occup)				0.38 M (CEL: lung cancer)	Lee-Feldstein 1986 As(+3)	
20	Human	>25 yr (occup)				0.29 M (CEL: lung cancer)	Lubin et al. 2000 As(+3)	
21	Human	14.8 yr (avg) (occup)				0.3 M (CEL: lung cancer)	Welch et al. 1982 As(+3)	

a The number corresponds to entries in Figure 3-1.

avg = average; Bd Wt = body weight; Cardio = cardiovascular; CEL = cancer effect level; d = day(s); F = female; Gastro = gastrointestinal; Gd = gestation day; hr = hour(s); Immuno/Lymphoret = immunological/lymphoreticular; LOAEL = lowest-observable-adverse-effect level; M = male; mo = month(s); NOAEL = no-observable-adverse-effect level; NS = not specified; occup = occupational; pmd = pre-mating day; Resp = respiratory; wk = week(s); yr = year(s)

Figure 3-1 Levels of Significant Exposure to Inorganic Arsenic - Inhalation

Acute (≤14 days)

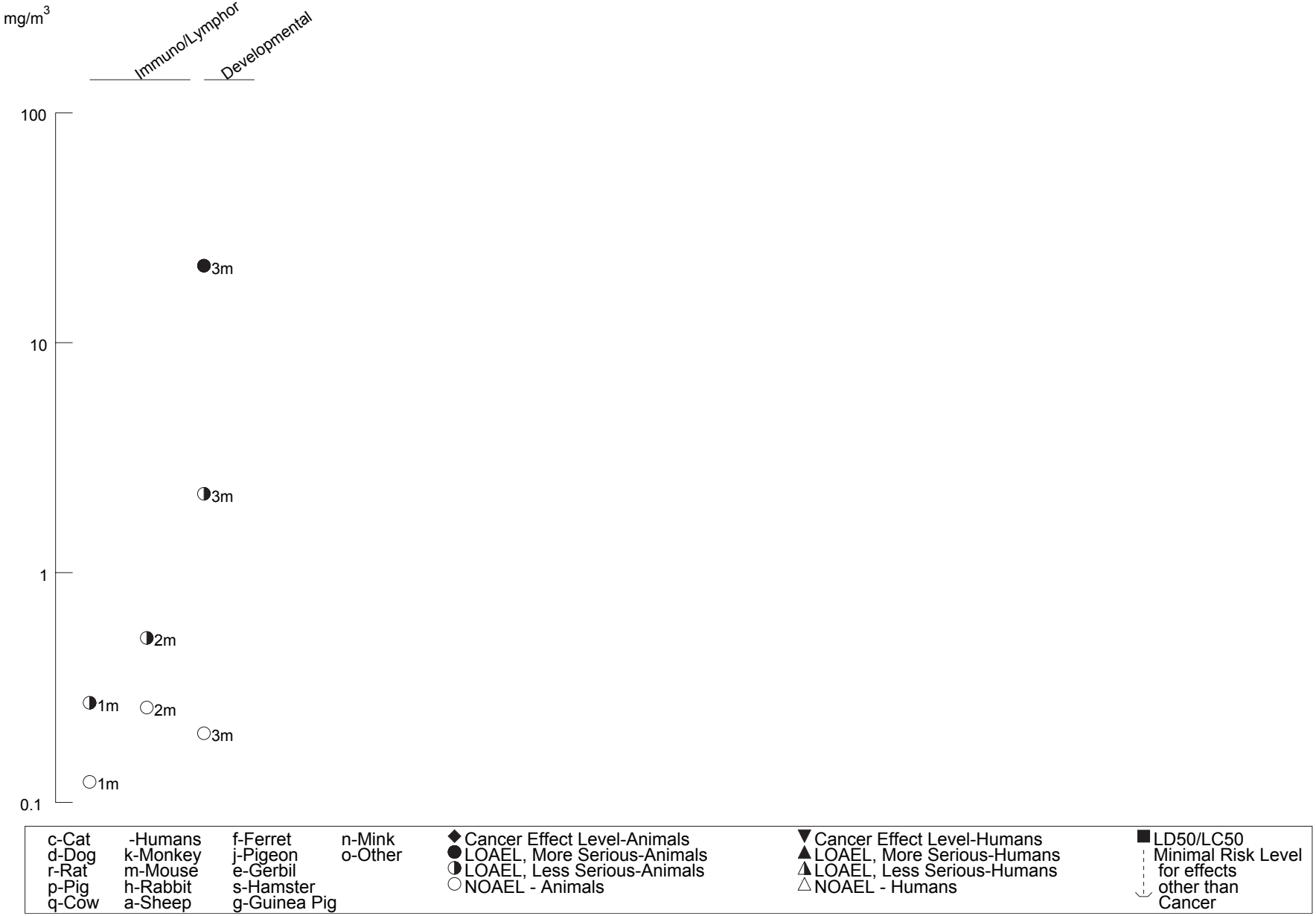


Figure 3-1 Levels of Significant Exposure to Inorganic Arsenic - Inhalation *(Continued)*

Intermediate (15-364 days)

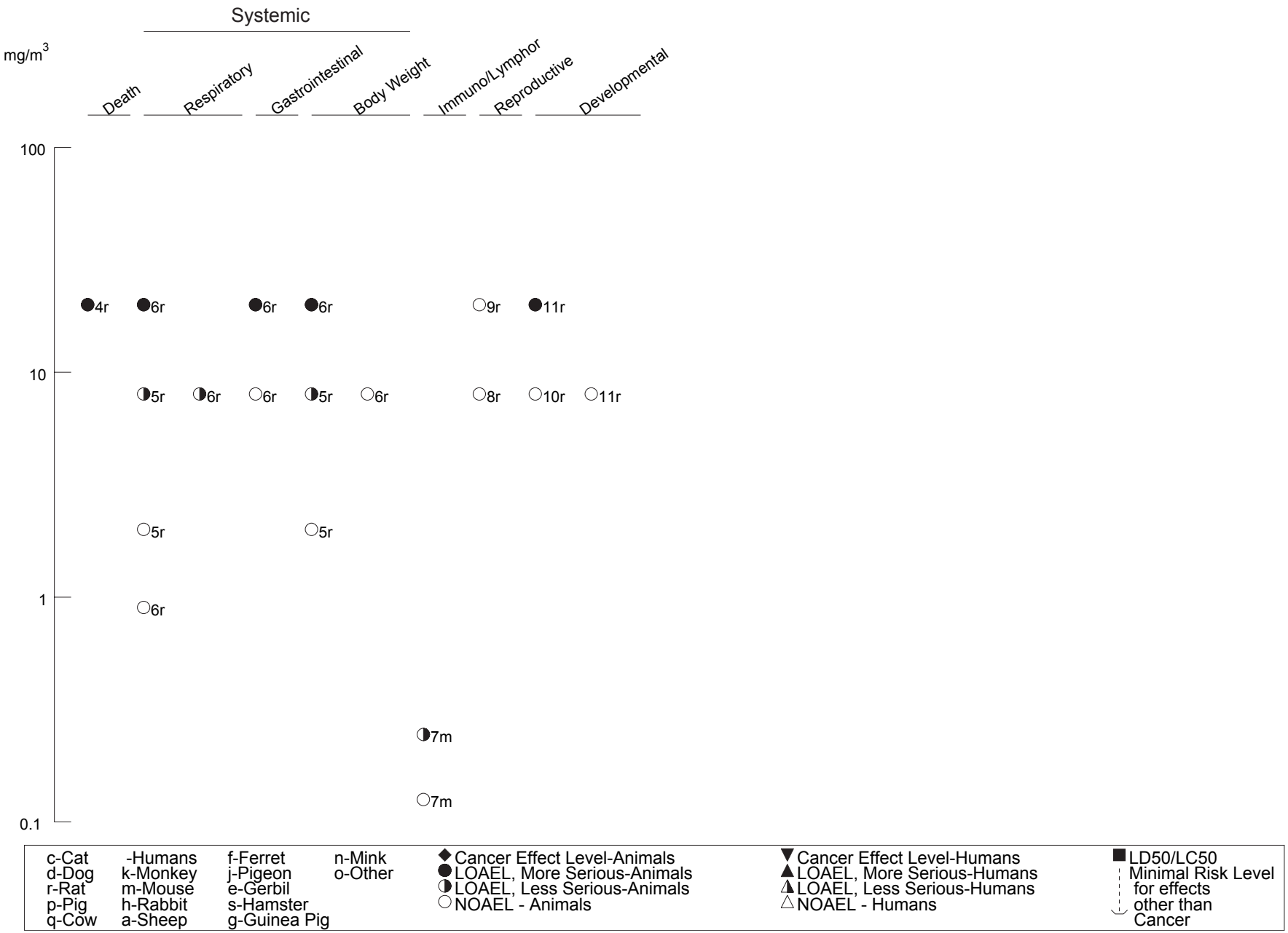


Figure 3-1 Levels of Significant Exposure to Inorganic Arsenic - Inhalation *(Continued)*

Chronic (≥365 days)

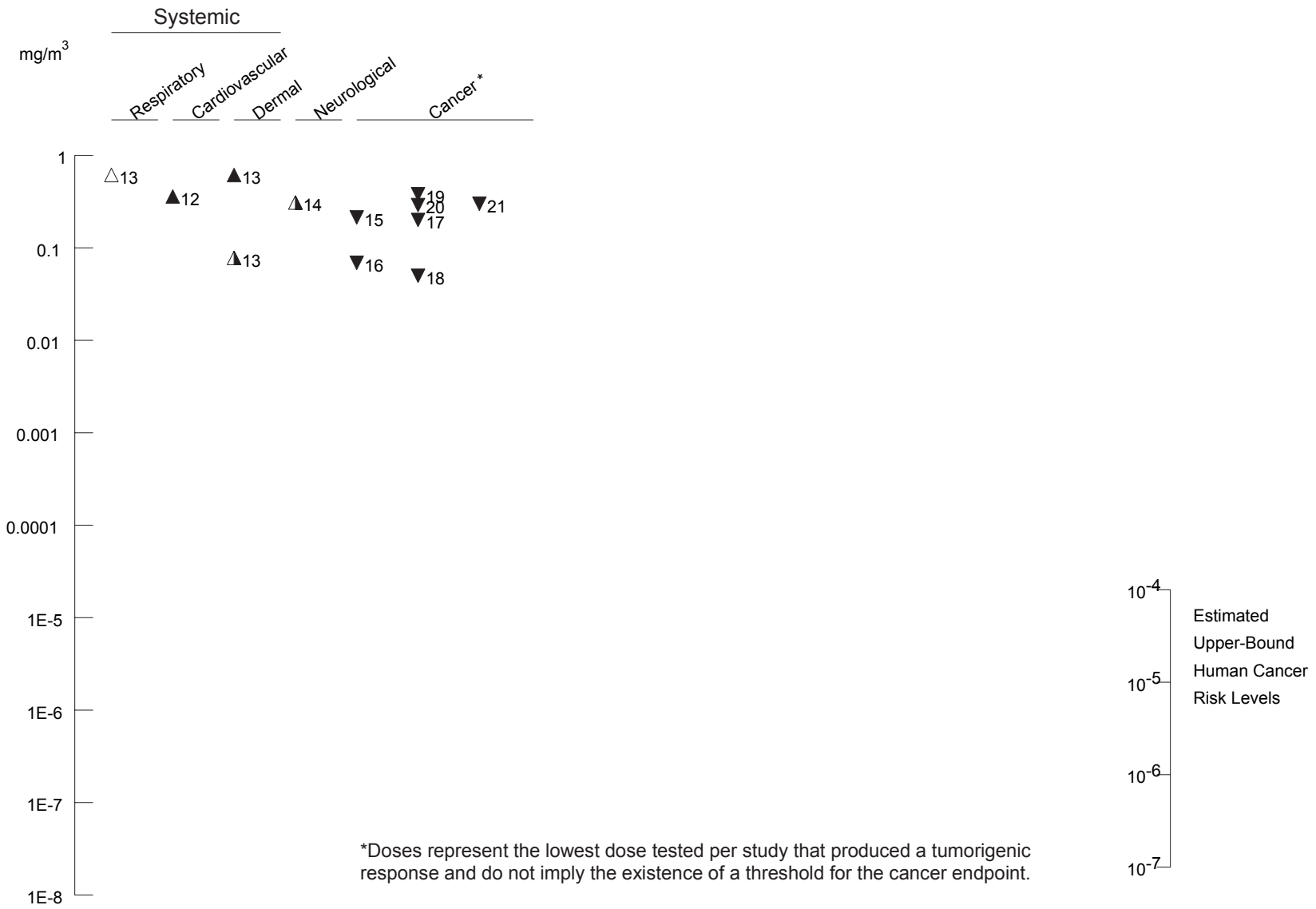


Table 3-2 Levels of Significant Exposure to Dimethylarsinic Acid - Inhalation

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/m³)	LOAEL Less Serious (mg/m³)	LOAEL Serious (mg/m³)	Reference Chemical Form	Comments
ACUTE EXPOSURE								
Death								
1	Rat (Sherman)	2 hr				3900 F (LC50)	Stevens et al. 1979 DMA	
Systemic								
2	Rat (Sherman)	2 hr	Resp			4000 (respiratory distress)	Stevens et al. 1979 DMA	
			Gastro		4000 (diarrhea)			
			Dermal	4100	6900 F (erythematous lesions of ears and feet)			
			Ocular		4000 (eye encrustation)			
			Bd Wt		4000 (unspecified decrease in body weight)			
3	Mouse (Swiss-Webster)	5 min	Resp		3150 M (RD50)		Stevens et al. 1979 DMA	
INTERMEDIATE EXPOSURE								
Systemic								
4	Rat (Sprague-Dawley)	6 hr/d 5 d/wk 67-68 exposures	Resp	10	34 (intracytoplasmic eosinophilic globules in nasal turbinates)		Whitman 1994 DMA	
			Cardio	100				
			Gastro	100				
			Hemato	100				
			Hepatic	100				
			Renal	100				
			Endocr	100				
			Dermal	100				

Table 3-2 Levels of Significant Exposure to Dimethylarsinic Acid - Inhalation (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/m^3)	LOAEL			Reference Chemical Form	Comments
					Less Serious (mg/m^3)	Serious (mg/m^3)			
			Bd Wt	100					

a The number corresponds to entries in Figure 3-2.

Bd Wt = body weight; Cardio = cardiovascular; d = day(s); DMA = dimethylarsinic acid; F = female; Gastro = gastrointestinal; Hemato = hematological; hr = hour(s); LC50 = lethal concentration, 50% kill; LOAEL = lowest-observable-adverse-effect level; M = male; min = minute(s); NOAEL = no-observable-adverse-effect level; RD50 = 50% decrease in respiration rate; Resp = respiratory; wk = week(s); yr = year(s)

Figure 3-2 Levels of Significant Exposure to Dimethylarsinic Acid - Inhalation

Acute (≤14 days)

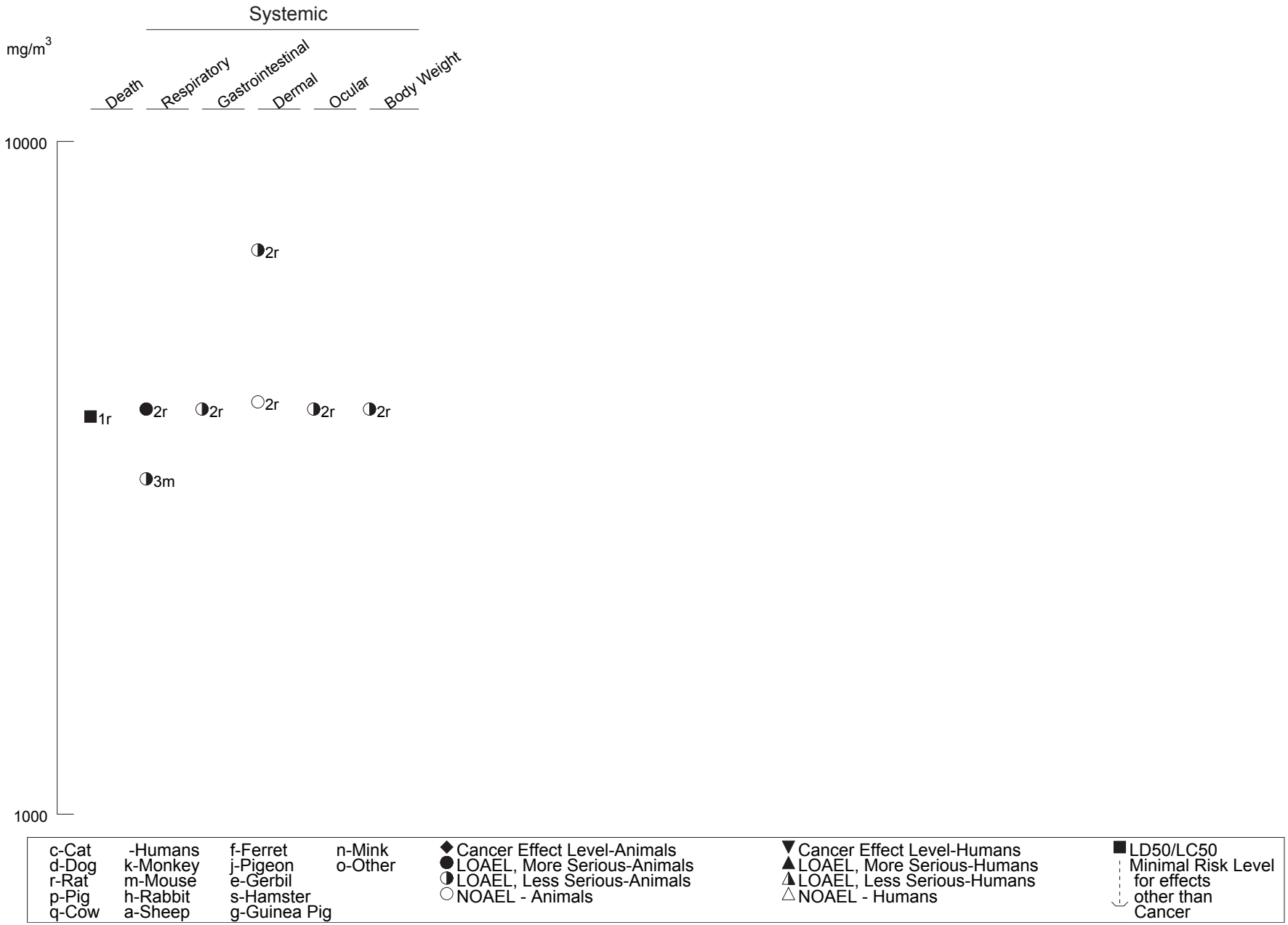
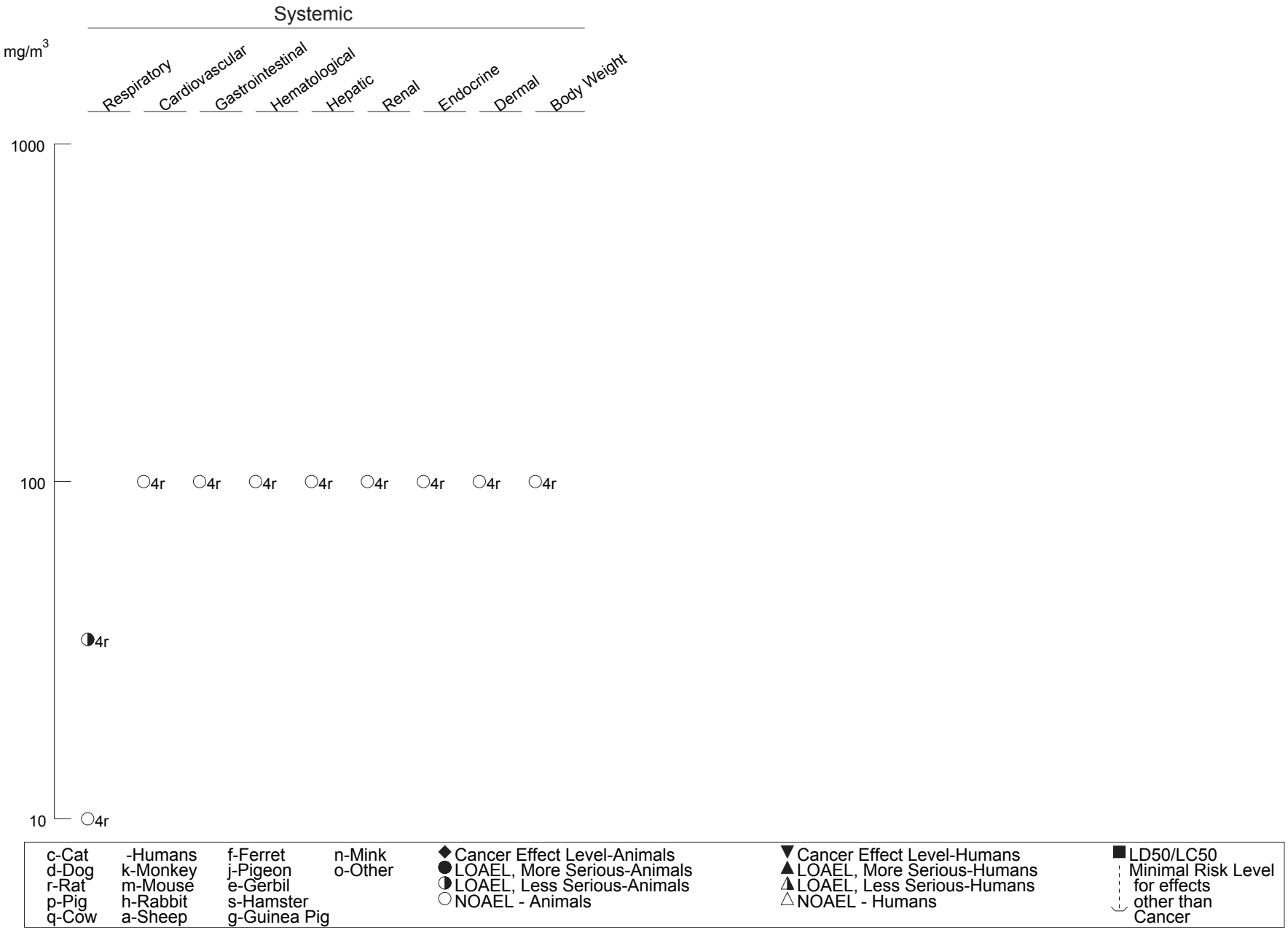


Figure 3-2 Levels of Significant Exposure to Dimethylarsinic Acid - Inhalation *(Continued)*

Intermediate (15-364 days)



3.2.1.1 Death

Inorganic Arsenicals. Although there are many studies of humans exposed to arsenic in air, no cases of lethality from short-term exposure were located. This suggests that death is not likely to be of concern following acute exposure, even at the very high exposure levels (1–100 mg As/m^3) found previously in the workplace (e.g., Enterline and Marsh 1982; Järup et al. 1989; Lee-Feldstein 1986). Delayed lethality from chronic exposure attributable to increased risk of cardiovascular disease or lung cancer is discussed below in Sections 3.2.1.2 and 3.2.1.7, respectively. The only report of a lethal effect of inhaled inorganic arsenic in animals was a developmental toxicology study in which four of nine pregnant rats died, and one rat was euthanized in extremis, between days 12 and 19 of gestation after 30–35 days of exposure to an aerosol of arsenic trioxide at an exposure concentration of 20 mg As/m^3 (Holson et al. 1999). These animals exhibited severe hyperemia and plasma discharge into the intestinal lumen at autopsy. In this same study, there was 100% mortality in groups of 10 pregnant rats after 1 day of exposure to concentrations ≥100 mg/m^3 (76 mg As/m^3).

Organic Arsenicals. No studies were located regarding death in humans after inhalation exposure to organic arsenicals. A 2-hour LC_{50} of 3,900 mg DMA/m^3 was calculated for DMA in female rats (Stevens et al. 1979). This LC_{50} is shown in Table 3-2 and Figure 3-2. Male rats and mice of both sexes were less susceptible, with only a few deaths after 2-hour exposures as high as 6,900 mg DMA/m^3 in rats and 6,400 mg DMA/m^3 in mice (Stevens et al. 1979). The cause of death was not specified, but was probably due to lung injury (see Section 3.2.1.2). No deaths were observed among rats and mice exposed to DSMA (the disodium salt of MMA) at concentrations up to 6,100 mg DSMA/m^3 in rats and 6,900 mg DSMA/m^3 in mice (Stevens et al. 1979). Chamber atmospheres at these high concentrations were so dense that it was difficult to see the animals clearly. These data indicate that there is no significant risk of acute lethality from concentrations of DMA or MMA that might be encountered in the environment or the workplace.

3.2.1.2 Systemic Effects

The highest NOAEL values and all reliable LOAEL values for systemic effects from inhalation exposure to inorganic arsenicals in each species and duration category are recorded in Table 3-1 and plotted in Figure 3-1, while the corresponding data for DMA are shown in Table 3-2 and Figure 3-2.

Respiratory Effects.

Inorganic Arsenicals. Workers exposed to arsenic dusts in air often experience irritation to the mucous membranes of the nose and throat. This may lead to laryngitis, bronchitis, or rhinitis (Dunlap 1921; Morton and Caron 1989; Pinto and McGill 1953), and very high exposures (characteristic of workplace exposures in the past) can cause perforation of the nasal septum (Dunlap 1921; Pinto and McGill 1953; Sandstrom et al. 1989). Despite the known respiratory irritant effects of arsenic, there have been few systematic investigations of respiratory effects in humans exposed to arsenic. Perry et al. (1948) found no difference in chest x-rays or respiratory performance (vital capacity and exercise-tolerance tests) between unexposed and exposed workers in a cross-sectional study at a factory where sodium arsenite was prepared. The NOAEL of 0.613 mg As/m^3 for respiratory effects in this study is shown in Table 3-1 and plotted in Figure 3-1.

Increased mortality due to respiratory disease has been reported in some cohort mortality studies of arsenic-exposed workers, but no conclusive evidence of an association with arsenic has been produced. In studies of workers exposed to arsenic trioxide at the Anaconda copper smelter in Montana, mortality due to noncancer respiratory disease (e.g., emphysema) was significantly increased compared to the general population (Lee-Feldstein 1983; Lubin et al. 2000; Welch et al. 1982). However, the data were not adjusted for smoking (a well-known confounder for respiratory disease), and analysis of the data with respect to arsenic exposure level did not show a clear dose-response. Similarly, Enterline et al. (1995) found a significant excess of nonmalignant respiratory disease mortality in workers at the ASARCO copper smelter in Tacoma, Washington, but only a slight negative relation to cumulative arsenic exposure. Xuan et al. (1993) found an increase in the relative risk of mortality from pneumoconiosis associated with arsenic exposure in a cohort of tin miners in China. However, this finding was based on a small number of observations (n=32), a clear exposure-response relationship with arsenic was not established, and the miners experienced confounding exposures to dust (a known risk factor for pneumoconiosis) and to radon. These studies were all considered to be inconclusive as to the relationship between inhaled inorganic arsenic and respiratory disease.

Respiratory symptoms were observed in a study of developmental effects in rats. Pregnant female rats exposed to arsenic trioxide dust starting 14 days prior to mating and continuing through mating and gestation exhibited rales at 8 mg As/m^3 and labored breathing and gasping at 20 mg As/m^3, with no symptoms at 2 mg As/m^3 (Holson et al. 1999). The lungs were examined by gross necropsy and no lesions were found. Intratracheal instillation of arsenic trioxide (13 mg As/kg) or gallium arsenide (1.5–

52 mg As/kg) can cause marked irritation and hyperplasia in the lungs of rats and hamsters (Goering et al. 1988; Ohyama et al. 1988; Webb et al. 1986, 1987). Since this sort of response is produced by a number of respirable particulate materials, it is likely that the inflammatory response is not specifically due to the arsenic.

Organic Arsenicals. No studies were located regarding respiratory effects in humans exposed to organic arsenicals. Short-term exposure of rats and mice to high concentrations (≥4,000 mg/m^3) of DMA caused respiratory distress, and necropsy of animals that died revealed bright red lungs with dark spots (Stevens et al. 1979). Respiratory distress was also observed in rats and mice exposed to high levels (≥6,100 mg/m^3) of the disodium salt of MMA (Stevens et al. 1979), although none of the MMA-exposed animals died. Respiratory distress appears to be associated with inhalation of very high concentrations of organic arsenicals. In 5-minute whole-body plethysmography trials, DMA and the disodium salt of MMA had RD_{50} (concentration calculated to produce a 50% decrease in respiration rate) values of 3,150 and 1,540 mg/m^3, respectively (Stevens et al. 1979). Based on these RD_{50} values, neither DMA nor MMA is considered to be a potent respiratory irritant. At low concentrations of DMA (34 or 100 mg DMA/m^3), an increase in intracytoplasmic eosinophilic globules were found in the nasal turbinates of rats exposed to DMA 6 hours/day, 5 days/week for 67–68 exposures (Whitman 1994).

Cardiovascular Effects.

Inorganic Arsenicals. There is some evidence from epidemiological studies that inhaled inorganic arsenic can produce effects on the cardiovascular system. Cardiovascular effects following oral exposure to arsenic are well known (see Section 3.2.2.2). A cross-sectional study of workers exposed to an estimated time-weighted average of 0.36 mg As/m^3 (as arsenic trioxide) at the Ronnskar copper smelter in Sweden for an average of 23 years showed that smelter workers had significantly increased incidences of Raynaud's phenomenon (a peripheral vascular disease characterized by spasm of the digital arteries and numbness of the fingers) and showed increased vasospasticity (constriction of blood vessels) in response to cold when tested in the fingers (Lagerkvist et al. 1986). A follow-up study conducted 2–3 years later found that vasospasticity measurements in exposed workers had improved concurrent with a reduction in arsenic exposure levels, although symptoms of peripheral vascular effects (cold hands or feet, white fingers, numbness in fingers or feet) were still common (Lagerkvist et al. 1988). A cross-sectional study including 46 workers in Denmark with varying, unquantified occupational exposure to arsenic in different occupations found that systolic blood pressure was significantly increased in the arsenic workers (median=125 mmHg) compared with controls (median=117 mmHg) (Jensen and Hansen 1998). Diastolic

pressure was also increased in this study (77.9 vs. 74.7 mmHg), although the difference from controls was not statistically significant.

Cohort mortality studies of arsenic-exposed workers at the ASARCO copper smelter in Tacoma, Washington (Enterline et al. 1995), Anaconda copper smelter in Montana (Lee-Feldstein 1983; Welch et al. 1982), Ronnskar copper smelter in Sweden (Wall 1980), orchard workers in Washington state (Tollestrup et al. 1995), and tin miners in China (Qiao et al. 1997; Xuan et al. 1993) have all reported increased risk of mortality from cardiovascular disease, specifically ischemic heart disease and cerebrovascular disease, in the cohorts studied. However, none of these studies provided conclusive evidence that the observed increase in risk was due to arsenic exposure. The studies in the ASARCO and Anaconda copper smelter workers failed to find a clear dose-response relationship with arsenic (Enterline et al. 1995; Welch et al. 1982), while a follow-up study of the Ronnskar smelter workers not only found lack of a dose-response, but also that the risk of cardiovascular disease was no longer elevated in the cohort (Järup et al. 1989). The studies in orchard workers and tin miners were limited by confounding exposures to copper, lead, and radon, respectively (Qiao et al. 1997; Tollestrup et al. 1995). The risk of cardiovascular disease mortality in the tin miners not only showed no dose-response relationship with arsenic exposure, but was positively associated with radon exposure, suggesting that radon may have been responsible for the increased cardiovascular risk in this cohort (Xuan et al. 1993).

The LOAEL for Raynaud's phenomenon and vasospasticity identified by Lagerkvist et al. (1986) is shown in Table 3-1 and Figure 3-1. No studies were located regarding cardiovascular effects in animals after inhalation exposure to inorganic arsenic.

Organic Arsenicals. No studies were located regarding cardiovascular effects in humans after inhalation exposure to organic arsenicals. No histological alterations were observed in the hearts of rats exposed to 100 mg DMA/m^3 for 67–68 exposures (Whitman 1994).

Gastrointestinal Effects.

Inorganic Arsenicals. Several case studies have reported nausea, vomiting, and diarrhea in workers with acute arsenic poisoning following occupational inhalation exposure (Beckett et al. 1986; Bolla-Wilson and Bleecker 1987; Ide and Bullough 1988; Morton and Caron 1989; Pinto and McGill 1953). Although gastrointestinal effects are not typically associated with arsenic poisoning by inhalation (Pinto and McGill 1953), such effects are a common feature of oral ingestion of high doses of arsenic (see Section 3.2.2.2),

and it is possible that mucociliary transport of arsenic dust from the lungs to the gut could be responsible for the effects in these cases. Exposure levels were not reliably estimated for any of these cases.

The only report of gastrointestinal effects of inhaled inorganic arsenic in animals was a developmental toxicology study in which four of nine pregnant rats died, and one rat was euthanized *in extremis*, between days 12 and 19 of gestation after 30–35 days of exposure to an aerosol of arsenic trioxide at an exposure concentration of 20 mg As/m^3 (Holson et al. 1999). These animals exhibited severe hyperemia and plasma discharge into the intestinal lumen at autopsy. Exposure to 8 mg As/m^3 did not produce gross gastrointestinal lesions.

Organic Arsenicals. Data regarding gastrointestinal effects in people exposed to organic arsenic in the air are limited. The frequency of gastrointestinal complaints was no higher than controls in workers exposed to arsanilic acid (i.e., 4-aminophenyl arsonic acid) at mean concentrations up to 0.17 mg/m^3 in a chemical factory (Watrous and McCaughey 1945). However, this sort of data might easily be biased by workers who chose not to complain about minor symptoms, so no conclusion can be reached. Rats and mice exposed to very high levels (above 3,000 mg/m^3) of MMA (disodium salt) or DMA experienced diarrhea (Stevens et al. 1979). The diarrhea could be due to transport of inhaled particulate material from the lungs to the gastrointestinal system or to direct ingestion of the compound (e.g., from grooming of the fur). No gastrointestinal effects were observed in rats repeatedly exposed to 100 mg DMA/m^3 6 hours/day, 5 days/week for 67–68 exposures (Whitman 1994).

Hematological Effects.

Inorganic Arsenicals. Although anemia is a common feature of arsenic poisoning following oral exposure in humans (see Section 3.2.2.2), case studies of workers with arsenic poisoning from occupational inhalation exposure reported no effects on red blood cell count (Beckett et al. 1986; Bolla-Wilson and Bleecker 1987; Ide and Bullough 1988; Morton and Caron 1989). The reason for this apparent route specificity is not clear, but might simply be related to dose. No studies were located regarding hematological effects in animals after inhalation exposure to inorganic arsenicals.

Organic Arsenicals. No effect on levels of hemoglobin, red cells, or white cells was detected in the blood of manufacturing workers (323 counts in 35 workers) exposed to airborne arsanilic acid dusts at a mean concentration of 0.17 mg/m^3 in the workplace (Watrous and McCaughey 1945). Controls were an unspecified number of unexposed manufacturing workers with 221 complete blood counts. No

hematological alterations were observed in rats exposed to 100 mg DMA/m^3 for an intermediate duration (Whitman 1994).

Musculoskeletal Effects.

Inorganic Arsenicals. Few data were located regarding musculoskeletal effects associated with inhalation exposure to inorganic arsenic, and none to suggest the existence of any such effects. Electromyographic examination of the calves and feet showed no differences between control and arsenic-exposed workers in a cross-sectional study of workers at the Ronnskar copper smelter in Sweden (Blom et al. 1985). No studies were located regarding musculoskeletal effects in animals after inhalation exposure to inorganic arsenicals.

Organic Arsenicals. No studies were located regarding musculoskeletal effects in humans or animals after inhalation exposure to organic arsenicals.

Hepatic Effects.

Inorganic Arsenicals. There is no evidence that inhaled inorganic arsenic produces effects on the liver, although few data are available. Case studies of workers with inhalation arsenic poisoning that included liver function tests did not find any evidence of hepatic dysfunction (Bolla-Wilson and Bleecker 1987; Ide and Bullough 1988). No studies were located regarding hepatic effects in animals after inhalation exposure to inorganic arsenicals.

Organic Arsenicals. No studies were located regarding hepatic effects in humans after inhalation exposure to organic arsenicals. No histological alterations were observed in the livers of rats exposed to 100 mg DMA/m^3 for 67–68 exposures (Whitman 1994).

Renal Effects.

Inorganic Arsenicals. The limited data available do not suggest any relationship between inhalation of inorganic arsenic and kidney effects. A cross-sectional study of renal function parameters in glass factory workers exposed to arsenic (concentrations unknown) found no meaningful differences from controls in urinary levels of several proteins (albumin, retinol binding protein, β_2-microglobulin, brush-border antigen) used as markers of glomerular damage or tubular cell exfoliation (Foà et al. 1987). Routine

clinical urinalysis was normal when included in case studies of workers with inhalation arsenic poisoning (Ide and Bullough 1988; Morton and Caron 1989). No studies were located regarding renal effects in animals after inhalation exposure to inorganic arsenicals.

Organic Arsenicals. No studies were located regarding renal effects in humans after inhalation exposure to organic arsenicals. No renal effects were reported in rats exposed to 100 mg DMA/m^3 6 hours/day, 5 days/week for 67–68 exposures (Whitman 1994).

Dermal Effects.

Inorganic Arsenicals. Dermatitis has frequently been observed in industrial workers exposed to inorganic arsenic in the air, with the highest rates occurring in the workers with the greatest arsenic exposure (Cöl et al. 1999; Dunlap 1921; Holmqvist 1951; Lagerkvist et al. 1986; Pinto and McGill 1953). Limited quantitative information is available regarding the exposure levels that produce dermatitis, and the high likelihood of co-exposure by the dermal route makes dose-response analysis difficult. A cross-sectional study of workers at a factory where sodium arsenite was prepared found that workers with the highest arsenic exposure (mean air levels ranging from 0.384 to 1.034 mg As/m^3 and estimated to average 0.613 mg As/m^3) tended to be grossly pigmented with hyperkeratinization of exposed skin and to have multiple warts (Perry et al. 1948). In the same study, workers with lower arsenic exposure (estimated to average 0.078 mg As/m^3) were much less affected, but still had a higher incidence of pigmentation keratosis than controls. LOAEL values identified by Perry et al. (1948) and Mohamed (1998) are shown in Table 3-1 and Figure 3-1. NOAEL values for dermal irritation have not been identified. Dermal effects (hyperkeratoses, hyperpigmentation) are also very common in people exposed to inorganic arsenic by the oral route (see Section 3.2.2.2). No studies were located on dermal effects in animals after inhalation exposure to inorganic arsenicals.

Organic Arsenicals. Data regarding dermal effects in people exposed to organic arsenic in the air are limited. Complaints of keratosis were roughly 2-fold higher than unexposed controls in female packaging workers exposed to arsanilic acid at an average concentration of 0.065 mg/m^3 and in male manufacturing workers exposed to an average concentration of 0.17 mg/m^3 in a chemical factory (Watrous and McCaughey 1945). Limitations in study methodology (e.g., alternate sources of effects were not investigated, workers might choose not to report minor complaints to company officials) make the reliability of this observation uncertain. Female rats exposed to DMA at 6,900 mg/m^3 developed erythematous lesions on the feet and ears (Stevens et al. 1979); these lesions did not develop in females

exposed at lower concentrations (4,100 mg/m^3) or males. It seems likely that these effects were due to direct irritation from dermal contact with the dust. No dermal effects were observed in rats repeatedly exposed to lower levels of DMA (100 mg/m^3) (Whitman 1994).

Ocular Effects.

Inorganic Arsenicals. Chemical conjunctivitis, characterized by redness, swelling, and pain, has been observed in workers exposed to arsenic dusts in air, usually accompanied by facial dermatitis (Dunlap 1921; Pinto and McGill 1953). No information was located regarding air levels of arsenic that produce this effect. No studies were located on ocular effects in animals after inhalation exposure to inorganic arsenicals.

Organic Arsenicals. No studies were located on ocular effects in humans after inhalation exposure to organic arsenicals. Rats and mice exposed to high concentrations of DMA (≥4,000 mg/m^3) developed an encrustation around the eyes (Stevens et al. 1979). It seems likely that these effects were due to direct irritation from ocular contact with the dust.

Body Weight Effects.

Inorganic Arsenicals. No studies were located on body weight effects in humans after inhalation exposure to inorganic arsenicals. Female rats exposed to arsenic trioxide dust starting 14 days before mating and continuing through mating and gestation showed a marked decrease in body weight and food consumption at 20 mg As/m^3 (preliminary study) and a smaller decrease at 8 mg As/m^3 (definitive study), with no effect at 2 mg As/m^3 (Holson et al. 1999).

Organic Arsenicals. No studies were located on body weight effects in humans after inhalation exposure to organic arsenicals. Rats and mice exposed to high concentrations of DMA (≥4,000 mg/m^3) for 2 hours had an unspecified decrease in body weight gain during the subsequent 14 days (Stevens et al. 1979). No alterations in body weight gain were observed in rats exposed to 100 mg DMA/m^3 for 67–68 exposures (Whitman 1994).

3.2.1.3 Immunological and Lymphoreticular Effects

Inorganic Arsenicals. A single study was located regarding the immunological and lymphoreticular effects of inhaled inorganic arsenic in humans. Bencko et al. (1988) detected no abnormalities in serum levels of immunoglobins in 47 workers exposed to arsenic (exposure levels not measured) in a coal-burning power plant. However, serum levels of other proteins such as transferrin, orosomucoid, and ceruloplasmin were significantly elevated compared to levels in a group of 27 workers from a different plant in which the arsenic content in the coal was 10 times lower. The investigators suggested that the increased levels of ceruloplasmin might be related to higher cancer mortality rates found among these workers.

The immune effects of inhaled arsenic in animals were studied by Aranyi et al. (1985). Female mice exposed to arsenic trioxide aerosol for 3 hours showed a concentration-related decrease in pulmonary bactericidal activity (presumably as a result of injury to alveolar macrophages) and a corresponding concentration-related increase in susceptibility to introduced respiratory bacterial pathogens. Similar results were found when the exposure was repeated over 1- and 4-week periods. The NOAEL and LOAEL values for this study are shown in Table 3-1 and Figure 3-1.

Intratracheal studies in animals offer some support for an immune effect of inhaled inorganic arsenic. Decreases in humoral response to antigens and in several complement proteins were noted in mice given an intratracheal dose of 5.7 mg As/kg as sodium arsenite (Sikorski et al. 1989), although these changes were not accompanied by any decrease in resistance to bacterial or tumor cell challenges. Animals given an intratracheal dose of GaAs (25 mg As/kg or higher) also displayed a variety of changes in numerous immunological end points (some increased, some decreased) (Burns and Munson 1993; Sikorski et al. 1989). Whether these effects were due to a direct effect on the immune system or were secondary to the inflammatory effect of GaAs on the lung (see Section 3.2.1.2, above) is uncertain.

Organic Arsenicals. No studies were located regarding immunological and lymphoreticular effects in humans or animals after inhalation exposure to organic arsenicals.

3.2.1.4 Neurological Effects

Inorganic Arsenicals. There is evidence from epidemiological studies that inhaled inorganic arsenic can produce neurological effects. A study by Gerr et al. (2000) reported an elevated incidence of peripheral neuropathy in subjects who lived near an arsenic-using pesticide plant (13/85=15.3%; odds ratio

[OR]=5.1, p=0.004), relative to subjects who lived farther from the plant (4/118=3.4%). Concentrations of arsenic in soil and house dust were elevated (~30–300 µg As/g) for residences near the plant, according to 1993–1995 monitoring data. Studies of copper smelter workers at the ASARCO smelter in Tacoma, Washington (Feldman et al. 1979), a power station in Slovakia (Buchancová et al. 1998), and the Ronnskar smelter in Sweden (Blom et al. 1985; Lagerkvist and Zetterlund 1994) have demonstrated peripheral neurological effects in workers associated with arsenic trioxide exposure. At the ASARCO smelter, the prevalence of clinically diagnosed peripheral neuropathy was markedly higher in arsenic-exposed workers (26/61=43%) than controls (4/33=12%), and although the difference in mean nerve conduction velocities (NCV) was not statistically significant, mean peroneal motor NCV was lower in arsenic-exposed workers than controls and all 12 cases of abnormally low NCV occurred in the arsenic group (Feldman et al. 1979). In the study of 70 workers in Slovakia, the investigators described 16 cases of arsenic intoxication. Among these, 13 had signs and symptoms of sensory and motor polyneuropathy on both upper and lower extremities, 10 were diagnosed with pseudoneurasthenic syndrome, and 6 suffered from toxic encephalopathy (Buchancová et al. 1998). The average length of exposure was 22.3 years (SD ±8.4 years) and the average arsenic exposure in inhaled air ranged from 4.6 to 142.7 µg/m^3. Similar results were observed at the Ronnskar smelter, where Blom et al. (1985) reported significantly increased prevalence of workers with abnormally low NCV in the exposed group, and lower, but not statistically significant, mean NCV in five peripheral nerves. A follow-up study on the Ronnskar workers 5 years later found that the prevalence of abnormally low NCV remained significantly increased in the exposed workers, but that the decrease in mean NCV was now also statistically significant in the tibial (motor) and sural (sensory) nerves (Lagerkvist and Zetterlund 1994). Blood lead was monitored in this study as a potential confounder, but levels were low and not considered likely by the researchers to have had any influence on the results. The follow-up Ronnskar study provided enough information to estimate that mean arsenic exposure was 0.31 mg As/m^3 and lasted an average of 28 years in the exposed group, and this LOAEL is shown in Table 3-1 and Figure 3-1.

The literature also contains several case studies of workers with inhalation arsenic poisoning who developed neurological symptoms. Although these studies do not provide reliable information on exposure levels or conclusive evidence that the observed effects were related to arsenic, the findings are suggestive. Symptoms in these cases included not only indicators of peripheral neuropathy (numbness, loss of reflexes, muscle weakness, tremors) (Ide and Bullough 1988; Morton and Caron 1989), but also frank encephalopathy (hallucinations, agitation, emotional lability, memory loss) (Beckett et al. 1986; Bolla-Wilson and Bleecker 1987; Morton and Caron 1989). Both peripheral neuropathy and encephalopathy are associated with oral exposure to inorganic arsenic (see Section 3.2.2.4).

The possible association between arsenic in air and neurological effects in children has also been examined. A study by Bencko et al. (1977) reported that children of approximately 10 years of age (n=56) living near a power plant burning coal of high arsenic content showed significant hearing losses (increased threshold) compared to a control group of children (n=51) living outside the polluted area (Bencko et al. 1977). The effect was most marked at low frequencies. The precise site affected within the auditory pathway was not determined and could have been in the periphery, centrally-located, or both. A small study of children in Mexico reported a significant negative correlation between tests of verbal IQ and urinary arsenic in children (n=41) living in an urban area near a smelter complex (Calderón et al. 2001). Exposure concentrations were not available in either study.

No studies were located regarding neurological effects in animals after inhalation exposure to inorganic arsenicals. Mice given a single intratracheal dose of 200 mg/kg of GaAs displayed a decrease in overall activity 6–8 hours later, but no additional neurological evaluations were conducted on these animals (Burns and Munson 1993).

Organic Arsenicals. Data regarding neurological effects in people exposed to organic arsenic in the air are limited to a single study. The frequency of central nervous system complaints was no higher than controls in workers at a chemical factory exposed to arsanilic acid at mean concentrations up to 0.17 mg/m^3 (Watrous and McCaughey 1945). Although peripheral nerve complaints were higher in arsenic packaging workers (mean exposure=0.065 mg/m^3) than in unexposed controls, this was not the case in manufacturing workers with higher arsenic exposure (mean=0.17 mg/m^3). This suggests that the effects on the peripheral nerves in the exposed packaging workers were not due to arsenic. The reliability of these data is limited by shortcomings in the study methodology (e.g., the data might easily be biased by workers who chose not to complain about minor symptoms). No studies were located regarding neurological effects in animals after inhalation exposure to organic arsenicals.

3.2.1.5 Reproductive Effects

Inorganic Arsenicals. No studies were located regarding reproductive effects in humans after inhalation exposure to inorganic arsenicals. Reproductive performance was evaluated in female rats exposed to 0.08–20 mg As/m^3 (preliminary study) or 0.2–8 mg As/m^3 (definitive study) as As_2O_3 6 hours daily from 14 days prior to mating through gestation day 19 (Holson et al. 1999). No changes occurred in the precoital interval (time to mating), mating index (percentage of rats mated), or fertility index (percentage

of matings resulting in pregnancy). The NOAEL values for this study are shown in Table 3-1 and Figure 3-1.

Organic Arsenicals. No studies were located regarding reproductive effects in humans or animals after inhalation exposure to organic arsenicals.

3.2.1.6 Developmental Effects

Inorganic Arsenicals. Developmental effects associated with occupational and environmental exposure to airborne arsenic have been investigated in a series of studies at the Ronnskar copper smelter in northern Sweden (Nordström et al. 1978a, 1978b, 1979a, 1979b). In comparison to a northern Swedish reference population, female employees of the smelter had a significantly increased incidence of spontaneous abortion (Nordström et al. 1979a), and their children had a significantly increased incidence of congenital malformations (Nordström et al. 1979b) and significantly decreased average birth weight (Nordström et al. 1978a). Increased incidence of spontaneous abortion and decreased average birth weight of children were also found in populations living in close proximity to the smelter (Nordström et al. 1978a, 1978b, 1979b). While these data are suggestive of developmental effects associated with occupational and environmental exposure from the smelter, the reported effects are not large, the analyses include only limited consideration of potential confounders (e.g., smoking), and there are no data relating the apparent effects specifically to arsenic exposure.

Ihrig et al. (1998) conducted a case-control study of stillbirths in the vicinity of a Texas arsenic pesticide factory that included estimation of environmental arsenic exposures using atmospheric dispersion modeling and multiple regression analysis considering arsenic exposure, race/ethnicity, maternal age, median income, and parity as explanatory variables. There was a statistically significant increase in the risk of stillbirth in the highest exposure category (>100 ng As/m^3, midpoint=682 ng/m^3). Further analysis showed that this increase in risk was limited to people of Hispanic descent, who the researchers speculated may be an especially sensitive population due to a genetic impairment in folate metabolism. Interpretation of this study is limited by small numbers of cases and controls in the high exposure group, lack of data on smoking, potential confounding exposures to other chemicals from the factory, and failure to take into account previous years of deposition in the exposure estimates.

Arsenic has been shown to produce developmental effects by inhalation exposure in laboratory animals, although it is unclear whether or not the effects occur only at maternally toxic doses. Mice exposed to

22 mg As/m^3 (as As_2O_3) for 4 hours on days 9–12 of gestation had serious developmental effects (significant increases in the percentage of dead fetuses, skeletal malformations, and the number of fetuses with retarded growth), while those exposed to 2.2 mg As/m^3 had only a 10% decrease in average fetal body weight, and those exposed to 0.20 mg As/m^3 had no effects (Nagymajtényi et al. 1985). The study was limited by failure to quantify malformations on a litter basis, discuss the nature and severity of the observed malformations, or report on the occurrence of maternal effects. No increases in fetal resorptions, fetal mortality, or malformations, and no decreases in fetal body weight occurred when rats were exposed to 0.2–8 mg As/m^3 (as As_2O_3), 6 hours daily from 14 days prior to mating through gestation day 19 (Holson et al. 1999). At the 8 mg/m^3 exposure level, toxicity was observed in the dams, including rales, a dried red exudate at the nose, and lower gains in net body weight than controls. In a preliminary dose-range study, there was a marked significant increase in postimplantation loss (primarily early resorptions) and consequent marked significant decrease in viable fetuses per litter at 20 mg As/m^3, a concentration that also produced severe maternal effects including mortality (Holson et al. 1999).

The NOAEL and LOAEL values for increased risk of stillbirth in humans identified by Ihrig et al. (1998) and those for developmental effects in rodents found by Nagymajtényi et al. (1985) and Holson et al. (1999) are shown in Table 3-1 and Figure 3-1.

Organic Arsenicals. No studies were located regarding developmental effects in humans or animals after inhalation exposure to organic arsenicals.

3.2.1.7 Cancer

Inorganic Arsenicals. There is convincing evidence from a large number of epidemiological studies that inhalation exposure to inorganic arsenic increases the risk of lung cancer. Most studies involved workers exposed primarily to arsenic trioxide dust in air at copper smelters (Axelson et al. 1978; Brown and Chu 1982, 1983a, 1983b; Enterline and Marsh 1982; Enterline et al. 1987a, 1987b, 1995; Ferreccio et al. 1996; Järup and Pershagen 1991; Järup et al. 1989; Lee and Fraumeni 1969; Lee-Feldstein 1983, 1986; Lubin et al. 2000; Mazumdar et al. 1989; Pinto et al. 1977, 1978; Sandstrom et al. 1989; Viren and Silvers 1999; Wall 1980; Welch et al. 1982) and mines (Liu and Chen 1996; Qiao et al. 1997; Taylor et al. 1989; Xuan et al. 1993), but increased incidence of lung cancer has also been observed at chemical plants where exposure was primarily to arsenate (Bulbulyan et al. 1996; Mabuchi et al. 1979; Ott et al. 1974; Sobel et al. 1988). In addition, several studies suggest that residents living near smelters or arsenical chemical plants may also have increased risk of lung cancer (Brown et al. 1984; Cordier et al. 1983; Matanoski et

al. 1981; Pershagen 1985), although the increases are small and are not clearly detectable in all cases (e.g., Frost et al. 1987). The strongest evidence that arsenic is responsible for the observed lung cancer comes from quantitative dose-response data relating specific arsenic exposure levels to lung cancer risk. These data are available for arsenic-exposed workers at the ASARCO copper smelter in Tacoma, Washington (Enterline and Marsh 1982; Enterline et al. 1987a, 1995; Mazumdar et al. 1989), the Anaconda copper smelter in Montana (Lee-Feldstein 1986; Welch et al. 1982), eight other U.S. copper smelters (Enterline et al. 1987b), and the Ronnskar copper smelter in Sweden (Järup and Pershagen 1991; Järup et al. 1989). A common limitation of these studies is confounding exposure to other chemicals, such as sulfur dioxide, and cigarette smoking.

Enterline and Marsh (1982) reported a significant increase in respiratory cancer mortality (standard mortality ratio [SMR]=189.4) based on 104 observed respiratory cancer deaths and only 54.9 expected over the years 1941–1976 in a cohort of 2,802 male workers employed for ≥1 year between 1940 and 1964 at the ASARCO smelter. When the cohort was separated into low and high arsenic exposure groups, with mean estimated time-weighted average arsenic exposures of 0.054 and 0.157 mg As/m^3, respectively (based on work history, historical urinary arsenic measurements, and an experimentally derived relationship between urinary and inhaled arsenic), respiratory cancer mortality was significantly increased in both groups in a concentration-related fashion (SMR=227.7 and 291.4 in the low and high groups, respectively). Enterline et al. (1987a) re-analyzed these data using improved exposure estimates that incorporated historical measurements of arsenic in the ambient air and personal breathing zone of workers. Respiratory cancer mortality was significantly increased in a concentration-related fashion in the low (SMR=213.0), medium (SMR=312.1), and high (SMR=340.9) arsenic exposure groups, which had mean estimated time-weighted average arsenic exposures of 0.213, 0.564, and 1.487 mg As/m^3, respectively. An alternative analysis of these data by Mazumdar et al. (1989) produced similar results. Enterline et al. (1995) extended the mortality follow-up from 1976 to 1986, but reported findings similar to the earlier study in a less thorough analysis. The CEL from Enterline et al. (1987a), the most complete analysis of the ASARCO cohort with the best exposure estimates, is presented in Table 3-1 and Figure 3-1.

Respiratory cancer mortality was significantly increased (SMR=285) based on 302 observed respiratory deaths between 1938 and 1977 in a cohort of 8,045 white male workers employed for at least 1 year between 1938 and 1956 at the Anaconda smelter (Lee-Feldstein 1986). When workers were categorized according to cumulative arsenic exposure and date of hire, lung cancer mortality was significantly increased in all groups hired between 1925 and 1947. Workers in the lowest cumulative exposure group

(<10 mg-mo/m^3) were reported to have had <2 years of exposure at an average arsenic concentration of 0.38 mg/m^3. An alternative analysis of a subset of the Anaconda cohort (n=1,800, including all 277 employees with heavy arsenic exposure and 20% of the others) that included information on smoking and other occupational exposures was performed by Welch et al. (1982). This analysis showed that lung cancer mortality increased with increasing time-weighted average arsenic exposure, with a small nonsignificant increase in the low group (SMR=138) exposed to 0.05 mg/m^3 and significant increases in the medium (SMR=303), high (SMR=375), and very high (SMR=704) groups exposed to 0.3, 2.75, and 5.0 mg/m^3, respectively. Cohort members were more likely to be smokers than U.S. white males, but smoking did not differ among the arsenic exposure groups. Exposure-response analysis of smokers was similar to the analysis based on the full subcohort, while analysis of nonsmokers (limited by small group sizes) also showed a similar pattern, but with lower SMRs. In a followup analysis of the same cohort, Lubin et al. (2000) re-weighted the exposure concentrations based on duration and time of exposure and re-evaluated the effects of exposure. Relative risks for respiratory cancer increased with increasing duration in each arsenic exposure area (light, medium, and heavy) after adjustment for duration in the other two exposure areas. SMRs were significantly elevated following exposure to 0.58 mg/m^3 (medium; SMR=3.01, 95% CI=2.0–4.6) or 11.3 mg/m^3 (high; SMR=3.68, 95% CI=2.1–6.4) for 10 or more years, and following exposure to 0.29 mg/m^3 (low; SMR=1.86, 95% CI=1.2–2.9) for 25 or more years. The CELs from the analyses of the Anaconda cohort are presented in Table 3-1 and Figure 3-1.

Enterline et al. (1987b) studied the mortality experience from 1949 to 1980 of a cohort of 6,078 white males who had worked for 3 years or more between 1946 and 1976 at one of eight U.S. copper smelters in Arizona, Utah, Tennessee, and Nevada. Lung cancer mortality was significantly increased only in the Utah smelter (SMR=226.7), which had the highest average arsenic exposure concentration (0.069 mg/m^3 vs. 0.007–0.013 mg/m^3 in the other smelters) and also contributed the largest number of cohort members (n=2,288 vs. 189–965 from the other smelters). A nested case-control study showed that arsenic exposure and cigarette smoking were significant risk factors for lung cancer in the smelter workers. Smoking was lower in the Utah smelter workers than in the other smelter workers, but still higher than in the referent Utah population, suggesting that the risk attributable to arsenic in this study population is somewhat lower than indicated by the SMR reported above. The CEL from this study is presented in Table 3-1 and Figure 3-1.

Järup et al. (1989) reported significantly increased lung cancer mortality (SMR=372, 95% confidence interval [CI]=304–450) based on 106 lung cancer deaths in a cohort of 3,916 male workers employed for ≥3 months between 1928 and 1967 at the Ronnskar smelter and followed for mortality through 1981.

Workers were separated into low, medium, and high arsenic exposure groups with mean time-weighted average exposure estimates of 0.05, 0.2, and 0.4 mg/m^3, respectively. Lung cancer mortality was significantly increased in all three exposure groups in a concentration-related fashion (SMR=201, 353, and 480, respectively). A nested case-control analysis of 102 lung cancer cases and 190 controls from the cohort showed that lung cancer risk increased with increasing arsenic exposure in nonsmokers, light smokers, and heavy smokers (Järup and Pershagen 1991). The results demonstrated that arsenic is a risk factor for lung cancer in the smelter workers, but also suggested a greater-than-additive interaction between smoking and arsenic exposure. In this analysis, in contrast to the cohort study, lung cancer risk due to arsenic was increased only in the higher arsenic-exposure groups. Potential explanations for this difference between the cohort and case-control analyses include a higher proportion of smokers in the smelter workers than in the regional referent population in the cohort study, and limited power to detect increased risk in the case-control study due to small group sizes in the dose-response analysis. The CELs from both the cohort and case-control studies are presented in Table 3-1 and Figure 3-1.

Several researchers have examined the histological cell types of lung cancer (epidermoid carcinoma, small cell carcinoma, adenocarcinoma) in arsenic-exposed workers (e.g., Axelson et al. 1978; Newman et al. 1976; Pershagen et al. 1987; Qiao et al. 1997; Wicks et al. 1981). Although the incidence of the various cell types varied from population to population, all studies found an increase in several tumor types. This indicates that arsenic does not specifically increase the incidence of one particular type of lung cancer.

The studies of the ASARCO cohort (Enterline and Marsh 1982; Enterline et al. 1987a, 1995) noted a supralinear exposure-response relationship (i.e., steeper at lower doses) between arsenic exposure and lung cancer mortality. Hertz-Picciotto and Smith (1993) extended this observation to several other occupationally exposed cohorts with quantitative exposure information. The authors suggest that neither toxicokinetic mechanisms nor confounding from age, smoking, or other workplace carcinogens that differ by exposure level are likely explanations for the curvilinearity. Plausible explanations offered include: (1) synergism (with smoking), which varies in magnitude according to the level of arsenic exposure, (2) long-term survivorship at higher exposures among the healthier, less susceptible individuals, and (3) exposure estimate errors that were more prominent at higher-exposure levels as a result of past industrial hygiene sampling or worker protection practices.

Quantitative risk estimates for inhaled inorganic arsenic have been derived using the exposure-response data. EPA derived a unit risk estimate (the excess risk of lung cancer associated with lifetime exposure to

1 μg/m^3) of 4.3x10^{-3} per (μg/m^3) based on the dose-response relationships between arsenic exposure and excess lung cancer mortality in workers at the Anaconda smelter in Montana (Brown and Chu 1982, 1983a, 1983b; Lee-Feldstein 1983; and an unpublished paper by Higgins and associates) and the ASARCO smelter in Tacoma, Washington (Enterline and Marsh 1982; EPA 1984a; IRIS 2007). In some cases, calculations of exposure, as well as the procedures for generating quantitative risk estimates, are quite complex and the interested reader is referred to the EPA documents (EPA 1981c, 1984a, 1987e, 1996b; IRIS 2007) for a detailed description. Viren and Silvers (1994) re-evaluated the unit risk estimate using the same methods as EPA, but incorporating updated results from the ASARCO smelter (Enterline et al. 1987a; Mazumdar et al. 1989) and the findings from the Swedish smelter (Järup et al. 1989). Their analysis yielded a revised unit risk of 1.28x10^{-3} per (μg/m^3) that, when pooled with the earlier estimate from the Montana smelter cohort, yielded a composite unit risk of 1.43x10^{-3} per (μg/m^3). This unit risk estimate is a factor of 3 smaller than the EPA's current estimate of 4.3x10^{-3} per (μg/m^3). Figure 3-1 shows the air concentrations that correspond to excess lifetime cancer risks of 10^{-4}–10^{-7} based on the EPA unit risk estimate.

There have been occasional reports of other types of cancer (i.e., nonrespiratory cancer) potentially associated with inhalation exposure to inorganic arsenic, but there is no strong evidence for any of them. For example, Enterline et al. (1995) found significantly increased mortality due to cancer of the large intestine and bone cancer in the ASARCO cohort. However, neither cancer showed any relation to cumulative arsenic exposure, and the purported increase in bone cancer risk was based on a very small number of observations. Pesch et al. (2002) reported an increase in nonmelanoma skin cancers resulting from exposure from a Slovakian coal-burning power plant, but exposure levels associated with the lesions were not presented. Bencko et al. (2005) also reported an increase in the incidence of nonmelanoma skin cancer among workers of a power plant burning coal of a high arsenic content and in the population living in the vicinity of the power plant. Bulbulyan et al. (1996) reported an increase in risk of stomach cancer among workers exposed to the highest average arsenic concentrations at a Russian fertilizer plant, but this finding, which was based on a small number of observations and was only marginally statistically significant, was confounded by exposure to nitrogen oxides, which were more convincingly associated with stomach cancer in this study. Wingren and Axelson (1993) reported an association between arsenic exposure and stomach and colon cancer in Swedish glass workers, but this result was confounded by concomitant exposure to other metals. Lee-Feldstein (1983) observed a small, marginally significant increase in digestive tract cancer (SMR=125) in one study of the Anaconda cohort, but this was not found in other studies of this cohort (Lee and Fraumeni 1969; Lee-Feldstein 1986; Welch et al. 1982). Wulff et al. (1996) observed an apparent increase in the risk of childhood cancer (all types combined) in the

population living within 20 km of the Ronnskar smelter, but the apparent increase was based on a small number of cases (13 observed vs. 6.7 expected) and was not statistically significant, and exposure to arsenic was confounded by exposure to lead, copper, cadmium, sulfur dioxide, and possibly other emissions such as nickel and selenium. A retrospective study of deaths due to unspecified types of malignancies among workers of power plants found no significant differences in death rate between two groups whose exposure levels to arsenic had a difference of one order of magnitude (Bencko et al. 1980). However, the mean age of those deceased due to cancer in the high-exposure group was 55.9 years compared to 61.2 years in the low-exposure group, and this difference was statistically significant ($p<0.05$). Also, when the workers were stratified by exposure-duration, there was a significantly higher frequency of tumors in the high-exposure group after shorter employment periods (<5 or 6–10 years) than after a longer employment period (≥11 years). No information was provided regarding specific types of cancer. Various case reports have implicated occupational arsenic exposure as a potential contributing factor in workers who developed sinonasal cancer (Battista et al. 1996), hepatic angiosarcoma (Tsai et al. 1998a), and skin cancer (Cöl et al. 1999; Tsuruta et al. 1998), but provide no proof that inhaled arsenic was involved in the etiology of the observed tumors. Wong et al. (1992) found no evidence that environmental exposure to airborne arsenic produced skin cancer in residents living near the Anaconda smelter or an open pit copper mine.

No studies were located regarding cancer in animals after inhalation exposure to inorganic arsenicals, although several intratracheal instillation studies in hamsters have provided evidence that both arsenite and arsenate can increase the incidence of lung adenomas and/or carcinomas (Ishinishi et al. 1983; Pershagen and Björklund 1985; Pershagen et al. 1984; Yamamoto et al. 1987). These data support the conclusion that inhalation of arsenic may lead to lung cancer in humans.

Organic Arsenicals. No studies were located regarding cancer effects in humans or animals after inhalation exposure to organic arsenicals.

3.2.2 Oral Exposure

There are a large number of studies in humans and animals on the toxic effects of ingested arsenic. In humans, most cases of toxicity have resulted from accidental, suicidal, homicidal, or medicinal ingestion of arsenic-containing powders or solutions or by consumption of contaminated food or drinking water. In some cases, the chemical form is known (e.g., the most common arsenic medicinal was Fowler's solution, which contained 1% potassium arsenite or arsenic trioxide), but in many cases (e.g., exposures through

drinking water), the chemical form is not known. In these cases, it is presumed that the most likely forms are either inorganic arsenate [As(+5)], inorganic arsenite [As(+3)], or a mixture. Table 3-3 and Figure 3-3 summarize a number of studies that provide reliable quantitative data on health effects in humans and animals exposed to inorganic arsenicals by the oral route. Similar data for MMA, DMA, and roxarsone are listed in Tables 3-4, 3-5, and 3-6, and shown in Figures 3-4, 3-5, and 3-6, respectively. All exposure data are expressed as milligrams of arsenic (as the element) per kilogram body weight per day (mg As/kg/day). These studies and others that provide useful qualitative information are summarized below.

3.2.2.1 Death

Inorganic Arsenicals. There are many case reports of death in humans due to ingestion of high doses of arsenic. In nearly all cases, the most immediate effects are vomiting, diarrhea, and gastrointestinal hemorrhage, and death may ensue from fluid loss and circulatory collapse (Levin-Scherz et al. 1987; Saady et al. 1989; Uede and Furukawa 2003). In other cases, death may be delayed and result from the multiple tissue injuries produced by arsenic (Campbell and Alvarez 1989). Some accounts of fatal arsenic poisoning describe both gastrointestinal effects soon after ingestion and extensive damage to multiple organ systems prior to death (Quatrehomme et al. 1992). A precise estimate of the ingested dose is usually not available in acute poisonings, so quantitative information on lethal dose in humans is sparse. The lethal doses ranged from 22 to 121 mg As/kg in four cases where known amounts were ingested as a single bolus (Civantos et al. 1995; Hantson et al. 1996; Levin-Scherz et al. 1987; Quatrehomme et al. 1992). Two people in a family of eight died from ingestion of water containing about 110 ppm of arsenic for a week (Armstrong et al. 1984). This corresponded to a dose of about 2 mg As/kg/day. Based on a review of clinical reports in the older literature, Holland (1904) estimated the minimum lethal dose to be about 130 mg (also about 2 mg/kg). A similar estimate of 70–180 mg (about 1–3 mg/kg) was provided by Vallee et al. (1960). Death due to chronic arsenic exposure has been reported at lower concentrations. Five children between the ages of 2 and 7 years died from late sequelae of chronic arsenic poisoning after drinking contaminated water throughout their lives at estimated average doses of 0.05–0.1 mg As/kg/day (Zaldívar and Guillier 1977). A 22-year-old man with chronic arsenical dermatosis died from arsenic-related effects after lifetime exposure to an estimated average dose of 0.014 mg As/kg/day in the drinking water (Zaldívar et al. 1981). Systematic studies of lethality from chronic exposure attributable to increased risk of cardiovascular disease or cancer are discussed below in Sections 3.2.2.2 and 3.2.2.7, respectively.

Table 3-3 Levels of Significant Exposure to Inorganic Arsenic - Oral

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference Chemical Form	Comments
ACUTE EXPOSURE								
Death								
1	Human	1 wk (W)				2 (death)	Armstrong et al. 1984 NS	
2	Human	once (IN)				121 M (death)	Civantos et al. 1995 As(+5)	
3	Human	once (IN)				108 M (death)	Hantson et al. 1996 As(+3)	
4	Human	once (IN)				22 M (death)	Levin-Scherz et al. 1987 As(+3)	
5	Human	once (IN)				93 M (death)	Quatrehomme et al. 1992 As(+3)	
6	Rat (wild Norway)	once (G)				104 (LD50)	Dieke and Richter 1946 As(+3)	
7	Rat (Sherman)	once (G)				112 F (LD50)	Gaines 1960 As(+5) calcium arsenate	
8	Rat (Sherman)	once (G)				44 F (LD50)	Gaines 1960 As(+3)	
9	Rat (Sherman)	once (G)				175 F (LD50)	Gaines 1960 As(+5) lead arsenate	
10	Rat (Sprague-Dawley)	once (GW)				15 M (LD50)	Harrisson et al. 1958 As(+3)	

Table 3-3 Levels of Significant Exposure to Inorganic Arsenic - Oral (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference Chemical Form	Comments
11	Rat (Sprague-Dawley)	once (F)				145 M (LD50)	Harrisson et al. 1958 As(+3)	
12	Rat (CD)	once Gd 9 (GW)				23 F (7/25 dams died)	Stump et al. 1999 As(+3)	
13	Mouse (Swiss-Webster)	once (GW)				39 M (LD50)	Harrisson et al. 1958 As(+3)	
14	Mouse (C57H46)	once (GW)				26 M (LD50)	Harrisson et al. 1958 As(+3)	
15	Mouse (Dba2)	once (GW)				32 M (LD50)	Harrisson et al. 1958 As(+3)	
16	Mouse (C3H)	once (GW)				26 M (LD50)	Harrisson et al. 1958 As(+3)	
17	Mouse (ddY)	once (GW)				26 M (LD50)	Kaise et al. 1985 As(+3)	
18	Rabbit (New Zealand)	Gd 6-18 1 x/d (GW)				1.49 F (7/20 dams died)	Nemec et al. 1998 As(+5)	

Table 3-3 Levels of Significant Exposure to Inorganic Arsenic - Oral (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference Chemical Form	Comments
Systemic								
19	Human	1 wk (W)	Gastro		0.2 (vomiting, diarrhea, abdominal pain)	2 M (diffuse inflammation of the GI tract)	Armstrong et al. 1984 NS	
			Hemato			0.2 (pancytopenia, leukopenia)		
			Hepatic			0.4 (hepatitis)		
			Renal			0.2 (nephropathy)		
			Ocular		0.2 (periorbital swelling)			
20	Human	once (IN)	Resp			121 M (respiratory distress, lung hemmorhage and edema)	Civantos et al. 1995 As(+5)	
			Cardio			121 M (hypotension, ventricular fibrillation, cardiac arrest)		
			Gastro			121 M (ulceration of upper gastrointestinal tract)		

Table 3-3 Levels of Significant Exposure to Inorganic Arsenic - Oral (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference Chemical Form	Comments
21	Human	once (IN)	Cardio			19 F (tachycardia)	Cullen et al. 1995 As (+5)	
			Gastro			19 F (profuse vomiting and diarrhea)		
			Hemato	19 F				
			Hepatic	19 F				
			Renal	19 F				
22	Human	once (NS)	Resp			8 M (hemorrhagic bronchitis, pulmonary edema)	Fincher and Koerker 1987 As(+3)	
			Cardio			8 M (hypotension, tachycardia, massive cardiomegaly)		
			Gastro			8 M (gastrointestinal bleeding)		
			Hemato			8 M (hemolysis)		
			Musc/skel			8 M (marked atrophy of distal muscle groups)		
			Renal			8 M (acute renal failure)		
			Dermal		8 M (truncal macular rash)			
23	Human	1 or 2 x (W)	Gastro		0.05 (occasional nausea, diarrhea, and abdominal cramps)		Franzblau and Lilis 1989 As(+3) As(+5)	

Table 3-3 Levels of Significant Exposure to Inorganic Arsenic - Oral (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference Chemical Form	Comments
24	Human	once (W)	Gastro			120 M (vomiting and diarrhea)	Goebel et al. 1990 NS	
			Renal			120 M (anuria)		
			Dermal		120 M (hyperkeratosis)			
25	Human	once (IN)	Gastro		2 F (vomiting)		Hantson et al. 1996 As(+3)	
			Hepatic		2 F (slight increase in serum bilirubin)			
			Renal		2 F (altered renal function tests)			
26	Human	once (IN)	Gastro			13 M (frequent vomiting, diarrhea)	Kamijo et al. 1998 As(+3)	
			Hepatic			13 M (large increase in serum bilirubin, ALT, AST, LDH)		
			Dermal		13 M (erythematous eruption)			
			Ocular		13 M (constricted vision)			

Table 3-3 Levels of Significant Exposure to Inorganic Arsenic - Oral (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference Chemical Form	Comments
27	Human	once (IN)	Resp			22 M (tachypnea, respiratory failure)	Levin-Scherz et al. 1987 As(+3)	
			Cardio			22 M (cyanosis, hypotension, tachycardia, ventricular fibrillation)		
			Gastro			22 M (abdominal pain, nausea, diarrhea, massive vomiting, dysphagia, hemorrhage)		
			Hepatic			22 M (large increase in serum AST and LDH)		
			Renal			22 M (large increase in serum creatinine and BUN indicating acute renal failure)		
28	Human	once pregnancy wk 30 (IN)	Cardio			6 F (hypotension, rapid pulse)	Lugo et al. 1969 As(+3)	
			Gastro			6 F (abdominal pain, vomiting)		
			Hemato		6 F (high leukocyte count, low hematocrit)			
			Renal			6 F (acute renal failure)		

Table 3-3 Levels of Significant Exposure to Inorganic Arsenic - Oral (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference Chemical Form	Comments
29	Human	2-3 wk (F)	Resp		0.05 (sore throat, rhinorrhea, cough, sputum)		Mizuta et al. 1956 As(+5)	
			Cardio			0.05 (abnormal electrocardiogram)		
			Gastro			0.05[b] (nausea, vomiting, diarrhea, occult blood in feces and gastric and duodenal juice)		
			Hemato		0.05 (mild anemia, leukopenia)			
			Musc/skel		0.05 (tender calf muscle)			
			Hepatic		0.05 (mild hepatomegaly, impaired liver function, degenerative lesions)			
			Renal	0.05				
			Dermal		0.05 (pigmentation, itching, desquamation, exanthema)			
			Ocular		0.05 (edema of eyelids, conjunctivitis, central scotoma, neuro-retinitis)			

Table 3-3 Levels of Significant Exposure to Inorganic Arsenic - Oral (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference Chemical Form	Comments
30	Human	once (IN)	Resp	11 M		43 M (shortness of breath, decreased oxygen saturation)	Moore et al. 1994a As(+3)	
			Cardio	11 M		43 M (hypotension, asystolic cardiac arrest)		
			Gastro			11 M (profuse diarrhea and vomiting, severe abdominal pain)		
			Hemato	43 M				
			Renal		11 M (increased serum creatinine)	43 M (acute renal failure)		
31	Human	once (IN)	Resp			93 M (pulmonary edema)	Quatrehomme et al. 1992 As(+3)	
			Gastro			93 M (ulcero-necrotic hemorrhagic gastritis)		
			Hepatic			93 M (hepatomegaly, diffuse fatty degeneration)		
			Renal			93 M (glomerular congestion)		
			Dermal			93 M (dermoepidermic separation)		

Table 3-3 Levels of Significant Exposure to Inorganic Arsenic - Oral (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference Chemical Form	Comments
32	Monkey (Rhesus)	13 d 1 x/d (IN)	Gastro	3		6 (vomiting, unformed stool, "loss of condition")	Heywood and Sortwell 1979 As(+5)	
			Hepatic	3	6 (decreased liver glycogen, vacuolation of hepatocytes)			
			Renal	3	6 (dilation of proximal tubules)			
33	Rat (Wistar-Barby)	4-14 d 5 d/wk 1 x/d (G)	Cardio	2 F	11 F (decreased vasoreactivity)		Bekemeier and Hirschelmann 1989 As(+3)	
			Gastro	2 F		11 F (diarrhea, bloody stools)		
34	Rat (Sprague-Dawley)	2 x (GW)	Resp	14 F			Brown and Kitchin 1996 As(+3)	
			Hepatic		0.9 F (slight increased ornithine decarboxylase and heme oxygenase activity in liver)			
			Dermal	14 F				
35	Rat (Sprague-Dawley)	2 x (GW)	Hepatic	8 F	24 F (increased heme oxygenase activity in liver)		Brown et al. 1997c As(+5)	

Table 3-3 Levels of Significant Exposure to Inorganic Arsenic - Oral (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference Chemical Form	Comments
36	Rat (Sprague-Dawley)	1 x/d 15 d (G)	Bd Wt	10 M	20 M (20-25% decreased body weight)		Rodriguez et al. 2001 As(+3)	
37	Rat (CD)	once Gd 9 (GW)	Bd Wt	15 F	23 F (decreased body weight gain)		Stump et al. 1999 As(+3)	
38	Mouse (CD-1)	Gd 6-15 1 x/d (GW)	Bd Wt	12 F	24 F (decreased body weight gain during gestation)		Nemec et al. 1998 As(+5)	
39	Mouse (B6C3F1)	1 or 4 d 1 x/d (GW)	Hemato	3 M	6 M (decreased polychromatic erythrocytes in bone marrow)		Tice et al. 1997 As(+3)	
40	Gn Pig	1 x/d 8 d (G)	Cardio		3.8 M (prolongation of QT interval)		Chiang et al. 2002 As2O3	
41	Rabbit (New Zealand)	Gd 6-18 1 x/d (GW)	Bd Wt	0.37 F	1.49 F (loss of body weight during treatment during gestation)		Nemec et al. 1998 As(+5)	
Neurological								
42	Human	1 wk (W)				2 (encephalopathy, peripheral neuropathy)	Armstrong et al. 1984 NS	

Table 3-3 Levels of Significant Exposure to Inorganic Arsenic - Oral (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference Chemical Form	Comments
43	Human	once (IN)				121 M (confusion, brain edema)	Civantos et al. 1995 As(+5)	
44	Human	once (IN)				19 F (lethargy)	Cullen et al. 1995 As (+5)	
45	Human	once (NS)				8 M (severe, persistent encephalopathy and peripheral neuropathy)	Fincher and Koerker 1987 As(+3)	
46	Human	once (W)				120 M (severe polyneuropathy)	Goebel et al. 1990 NS	
47	Human	once (IN)				216 M (peripheral neuropathy)	Hantson et al. 1996 As(+3)	
48	Human	once (IN)				13 M (peripheral neuropathy)	Kamijo et al. 1998 As(+3)	
49	Human	once (IN)				22 M (agitation, disorientation, paranoia, violent reactions)	Levin-Scherz et al. 1987 As(+3)	
50	Human	2-3 wk (F)				0.05 (hypesthesia in legs, abnormal patellar reflex)	Mizuta et al. 1956 As(+5)	
51	Human	once (IN)		43 M			Moore et al. 1994a As(+3)	

Table 3-3 Levels of Significant Exposure to Inorganic Arsenic - Oral (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference Chemical Form	Comments
52	Human	once (IN)				93 M (encephalopathy)	Quatrehomme et al. 1992 As(+3)	
53	Monkey (Rhesus)	13 d 1 x/d (IN)		3		6 (marked salivation, uncontrolled head shaking)	Heywood and Sortwell 1979 As(+5)	
54	Rat (Sprague-Dawley)	1 x/d 15 d (G)		10 M	20 M (altered spontaneous locomotor activity)		Rodriguez et al. 2001 As(+3)	
55	Rabbit (New Zealand)	Gd 6-18 1 x/d (GW)		0.37 F		1.49 F (prostration, ataxia)	Nemec et al. 1998 As(+5)	
Developmental								
56	Human	once pregnancy wk 30 (IN)				6 (severe pulmonary hemorrhage that may have contributed to death in premature neonate)	Lugo et al. 1969 As(+3)	
57	Rat (CD)	once Gd 9 (GW)		15		23 (increased post-implantation loss and decreased viable fetuses)	Stump et al. 1999 As(+3)	

Table 3-3 Levels of Significant Exposure to Inorganic Arsenic - Oral (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)		Reference Chemical Form	Comments
58	Mouse (CD-1)	once Gd 8-15 (GW)		11		23	(increased fetal mortality, exencephaly)	Baxley et al. 1981 As(+3)	
59	Mouse (CD-1)	once Gd 7-15 (GW)				48	(increased fetal death, decreased fetal weight, gross and skeletal malformations)	Hood et al. 1978 As(+5)	
60	Mouse (CD-1)	Gd 6-15 1 x/d (GW)		12		24	(increased resorptions per litter, decreased live fetuses per litter, decreased mean fetal weight)	Nemec et al. 1998 As(+5)	
61	Hamster (Lak:LVG [SYR])	once Gd 8-12 (GW)		11		14	(increased fetal mortality, decreased fetal weight)	Hood and Harrison 1982 As(+3)	
62	Rabbit (New Zealand)	Gd 6-18 1 x/d (GW)		0.37		1.49	(increased resorptions per litter, decreased live fetuses per litter)	Nemec et al. 1998 As(+5)	

Table 3-3 Levels of Significant Exposure to Inorganic Arsenic - Oral (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference Chemical Form	Comments
Cancer								
63	Mouse C3H	10 d (W)				9.55 M (CEL: liver and adrenal tumors)	Waalkes et al. 2003 As(+3)	
						19.13 F (CEL: ovarian and lung tumors)		
INTERMEDIATE EXPOSURE								
Systemic								
64	Human	3 mo (W)	Gastro			0.1 (severe nausea, diarrhea, pain, cramps, vomiting, traces of blood in stool)	Franzblau and Lilis 1989 As(+3) As(+5)	
			Hemato			0.1 (anemia, leukopenia)		
			Hepatic			0.1 (large increased AST and ALT)		
			Dermal		0.1 (diffuse erythematous and scaly rash)			
			Ocular		0.1 (swelling and irritation of the eyes, impaired peripheral vision)			
65	Human	0.5-14 yr (W)	Dermal			0.05 (hyperpigmentation with keratosis, possibly pre-cancerous)	Huang et al. 1985 NS	

Table 3-3 Levels of Significant Exposure to Inorganic Arsenic - Oral (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference Chemical Form	Comments
66	Human	4 mo (W)	Gastro			0.06 F (nausea, vomiting, diarrhea)	Wagner et al. 1979 NS	
			Hemato			0.06 F (anemia, leukopenia, erythroid hyperplasia of bone marrow)		
			Dermal			0.06 F (persistent extensive hyperkeratosis of palms and soles)		
			Bd Wt			0.06 F (40 lb weight loss)		
67	Rat (Wistar-Barby)	4 wk 5 d/wk 1 x/d (GW)	Cardio		11 F (decreased vasoreactivity)		Bekemeier and Hirschelmann 1989 As(+3)	
68	Rat (Sprague-Dawley)	6 wk (W)	Renal		4.7 M (increased relative kidney weight, impaired renal mitochondrial respiration, ultrastructural changes in proximal tubule)		Brown et al. 1976 As(+5)	
			Bd Wt	9.4 M	10.9 M (decreased body weight gain)			

Table 3-3 Levels of Significant Exposure to Inorganic Arsenic - Oral (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference Chemical Form	Comments
69	Rat (Wistar)	1 x/d 28 d (G)	Bd Wt	0.14 F			Chattopadhyay et al. 2001 As(+3)	
70	Rat (CD)	6 wk (W)	Hepatic	3 M	6 M (ultrastructural changes in hepatocytes, impaired liver mitochondrial respiration)		Fowler et al. 1977 As(+5)	
			Bd Wt	6 M		12 M (final body weight 28% lower than controls)		
71	Rat (CD)	14 pmd- Gd 19 7 d/wk 6 hr/d (GW)	Gastro	4 F		8 F (stomach adhesions, eroded luminal epithelium in the stomach)	Holson et al. 2000 As(+3)	
			Hepatic	2 F	4 F (increased liver weight)			
			Renal	4 F	8 F (increased kidney weight)			
			Bd Wt	4 F	8 F (decreased body weight gain)			
72	Rat (NS)	16 wk (W)	Hemato		0.92 M (decreased erythrocyte and leukocyte numbers)		Kannan et al. 2001 As(+3)	
			Hepatic	2.3 M				

Table 3-3 Levels of Significant Exposure to Inorganic Arsenic - Oral (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference Chemical Form	Comments
73	Rat (Sprague-Dawley)	4 wk (W)	Hemato	0.12	0.3 (increased platelet aggregation)		Lee et al. 2002 As(+3)	
74	Rat (NS)	1 x/d 30 d (G)	Endocr		2.3 M (decreased islet cells in pancreas, increased pancreatic SOD and catalase)		Mukherjee et al. 2003 As2O3	
75	Rat (Wistar)	1 x/d 5 d/wk 12 wk (G)	Resp	19 M			Schulz et al. 2002 As(+3)	
			Renal	19 M				
			Bd Wt	9.5 M	19 M (~17% decreased body weight gain)			
76	Mouse (C57BL)	6 wk (W)	Hepatic	5 M	10 M (ultrastructural changes in hepatocytes, impaired liver mitochondrial respiration)		Fowler and Woods 1979 As(+5)	
			Bd Wt	5 M	10 M (decreased body weight gain)			

Table 3-3 Levels of Significant Exposure to Inorganic Arsenic - Oral (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference Chemical Form	Comments
77	Mouse (C57BL/6 B6)	14 wk (W)	Hepatic	25 M			Kerkvliet et al. 1980 As(+5)	
			Renal	25 M				
78	Gn Pig (NS)	16 wk (W)	Hemato		0.69 M (decreased erythrocyte number and leukocyte number, decreased ALAD levels)		Kannan et al. 2001 As(+3)	
			Hepatic		0.69 M (increased ALAS activity)			
79	Dog (Beagle)	26 wk ad lib (F)	Hemato	1.9 F			Neiger and Osweiler 1989 As(+3)	
			Hepatic		0.8 F (mild increased serum ALT/AST)			
			Renal	1.9 F				
			Bd Wt	0.8 F	1.5 F (decreased body weight gain)	1.9 F (25% decrease in body weight)		
Immuno/ Lymphoret								
80	Mouse (C57BL/6 B6)	14 wk (W)		25 M			Kerkvliet et al. 1980 As(+5)	
Neurological								
81	Human	3 mo (W)				0.1 (paresthesia of hands and feet; confusion, disorientation and mental sluggishness)	Franzblau and Lilis 1989 As(+3) As(+5)	

Table 3-3 Levels of Significant Exposure to Inorganic Arsenic - Oral (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference Chemical Form	Comments
82	Human	4 mo (W)				0.06 F (weakness, paresthesia)	Wagner et al. 1979 NS	
83	Rat (NS)	16 wk (W)		0.92 M	2.3 M (decreased brain neurotransmitter levels)		Kannan et al. 2001 As(+3)	
84	Rat (Wistar)	1 x/d 5 d/wk 12 wk (G)		19 M			Schulz et al. 2002 As(+3)	
85	Gn Pig (NS)	16 wk (W)		0.69 M	1.7 M (changes in brain neurotransmitter levels)		Kannan et al. 2001 As(+3)	
Reproductive								
86	Rat (Wistar)	1 x/d 28 d (G)			0.14 F (changes in uterine and ovarian weights, decreased estradiol)		Chattopadhyay et al. 2001 As(+3)	
87	Rat (CD)	14 pmd- Gd 19 7 d/wk 6 hr/d (GW)		8 F			Holson et al. 2000 As(+3)	
88	Mouse (CD)	3 gen (W)				1 (decreased litter size)	Schroeder and Mitchener 1971 As(+3)	

Table 3-3 Levels of Significant Exposure to Inorganic Arsenic - Oral (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference Chemical Form	Comments
Developmental								
89	Rat (CD)	14 pmd- Gd 19 7 d/wk 6 hr/d (GW)		4	8 (decreased fetal body weight, increased skeletal variations)		Holson et al. 2000 As(+3)	
90	Rat (Sprague-Dawley)	Gd 15 or pnd 1-4 mo (W)			2.93 M (impaired performance in postnatal neurobehavioral tests)		Rodriguez et al. 2002 As(+3)	
91	Mouse (CD)	3 gen (W)				1 (decreased litter size)	Schroeder and Mitchener 1971 As(+3)	
CHRONIC EXPOSURE								
Death								
92	Human	2-7 yr children (W)				0.05 (death)	Zaldivar and Guillier 1977 NS	
93	Human	22 yr (W)				0.014 M (death)	Zaldivar et al. 1981 NS	Cause of death was liver tumor.
94	Monkey (Rhesus)	1 yr (IN)				3 (2/7 died)	Heywood and Sortwell 1979 As(+5)	
95	Rat (Wistar)	27 mo (F)				30 (increased mortality)	Kroes et al. 1974 As(+5) lead arsenate	
96	Mouse (CD)	2 yr (W)				1 (increased mortality, decreased life span)	Schroeder and Balassa 1967 As(+3)	

Table 3-3 Levels of Significant Exposure to Inorganic Arsenic - Oral (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference Chemical Form	Comments
97	Dog (Beagle)	2 yr (F)				2.4 (6/6 died)	Byron et al. 1967 As(+3)	
98	Dog (Beagle)	2 yr (F)				2.4 (1/6 died)	Byron et al. 1967 As(+5)	
Systemic								
99	Human	NS (W)	Resp	0.032	(cough)		Ahmad et al. 1997 NS	
			Dermal			0.032 (melanosis, keratosis, hyperkeratosis, and depigmentation)		
			Ocular			0.032 (chronic conjunctivitis)		
100	Human	>8 yr (W)	Dermal	0.0012	(increased risk of premalignant skin lesions)		Ahsan et al. 2006 (NS)	
101	Human	4 yr (IN)	Dermal			0.1 F (de-pigmentation with hyperkeratosis, possibly pre-cancerous)	Bickley and Papa 1989 As(+3)	
102	Human	NS (W)	Cardio			0.014 (gangrene of feet)	Biswas et al. 1998 NS	
			Dermal			0.014 (melanosis and keratosis of hand palms and foot soles)		

Table 3-3 Levels of Significant Exposure to Inorganic Arsenic - Oral (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)		LOAEL Serious (mg/kg/day)		Reference Chemical Form	Comments
103	Human	12 yr (W)	Cardio				0.02	(Raynaud's disease, gangrene of toes)	Borgono and Greiber 1972 NS	
			Gastro		0.02	(diarrhea, abdominal pain)				
			Dermal				0.02	(abnormal pigmentation with hyperkaratosis, possibly pre-cancerous)		
104	Human	11-15 yr (W)	Dermal		0.01	(hypo- and hyperpigmentation)			Borgono et al. 1980 NS	
105	Human	NS (W)	Gastro	0.0004	0.022	(gastrointestinal irritation, diarrhea, nausea)			Cebrian et al. 1983 As(+5)	
			Dermal	0.0004			0.022	(pigmentation changes with hyperkeratosis, possibly pre-cancerous)		
106	Human	1-11 yr (W)	Hepatic		0.046	(hepatomegaly)			Chakraborty and Saha 1987 NS	
			Dermal				0.046	(pigmentation changes with keratosis, possibly pre-cancerous)		

Table 3-3 Levels of Significant Exposure to Inorganic Arsenic - Oral (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)		Reference Chemical Form	Comments
107	Human	NS (W)	Cardio			0.064	(Blackfoot disease)	Chen et al. 1988b NS	
108	Human	>10 yr (W)	Cardio	0.0008		0.022	(increased risk of ischemic heart disease mortality)	Chen et al. 1996 NS	
109	Human	NS (W)	Cardio			0.002	(increased prevalence of cerebrovascular disease and cerebral infarction)	Chiou et al. 1997 NS	
110	Human	>5 yr (W)	Hemato	0.006[c] M 0.007 F				EPA 1981b NS	
			Dermal	0.0009[c] M 0.001 F					
111	Human	3-7 yr (W)	Cardio			0.05	(Blackfoot disease)	Foy et al. 1992 NS	
			Dermal			0.05	(melanosis with hyperkeratosis, possibly pre-cancerous)		

Table 3-3 Levels of Significant Exposure to Inorganic Arsenic - Oral (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference Chemical Form	Comments
112	Human	2-6 yr (IN)	Hepatic			0.08 M (cirrhosis, ascites)	Franklin et al. 1950 As(+3)	
			Dermal			0.08 M (pigmentation with hyperkeratosis, possibly pre-cancerous)		
113	Human	1-15 yr (W)	Hepatic		0.16 (portal fibrosis of the liver)		Guha Mazumder 2005 (NS)	
114	Human	NS (W)	Hepatic	0.004		0.014 (hepatomegaly)	Guha Mazumder et al. 1988 NS	
			Dermal	0.004		0.014 (pigmentation changes with hyperkaratosis, possibly pre-cancerous)		
115	Human	1-20 yr (W)	Gastro		0.06 (abdominal pain)		Guha Mazumder et al. 1988 NS	
			Hemato		0.06 (anemia)			
			Hepatic			0.06 (hepatomegaly, fibrosis)		
			Dermal			0.06 (hyperpigmentation with hyperkeratosis, possibly pre-cancerous)		
116	Human	NS (W)	Dermal	0.0016		0.009 (hyperpigmentation with keratosis, possibly pre-cancerous)	Guha Mazumder et al. 1998a NS	

Table 3-3 Levels of Significant Exposure to Inorganic Arsenic - Oral (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference Chemical Form	Comments
117	Human	(W)	Dermal		0.0014 (arsenical dermatosis)		Guo et al. 2001a (NS)	
118	Human	NS (W)	Dermal		0.0043 (hyperkeratosis, hyperpigmentation)		Haque et al. 2003 (NS)	dose listed is that associated with lowest known peak As concentration ingested by a case with complete water history
119	Human	10 yr (W)	Gastro	0.0046			Harrington et al. 1978 NS	
			Hemato	0.0046				
			Dermal	0.0046				
120	Human	NS (W)	Hepatic	0.0008	0.006 (increased serum alkaline phosphatase and bilirubin)		Hernandez-Zavala et al. 1998 NS	
121	Human	lifetime (W)	Hemato		0.002 F (anemia during pregnancy)		Hopenhayn et al. 2006 (NS)	
122	Human	NS (W)	Cardio			0.067 (ischemic heart disease)	Hsueh et al. 1998b NS	

Table 3-3 Levels of Significant Exposure to Inorganic Arsenic - Oral (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)		Reference Chemical Form	Comments
123	Human	0.5-14 yr (W)	Dermal			0.05	(hyperpigmentation with keratosis, possibly pre-cancerous)	Huang et al. 1985 NS	
124	Human	15 yr (IN)	Gastro			0.03 M	(hematemesis, hemoperitoneum, melena)	Lander et al. 1975 As(+3)	
			Dermal			0.03 M	(hyperkeratosis - possibly pre-cancerous)		
125	Human	NS (W)	Cardio	0.004		0.005	(cyanosis of extremities, palpitations/chest discomfort)	Lianfang and Jianzhong 1994 NS	
			Dermal	0.004		0.005	(keratosis, hyperpigmentation, depigmentation)		
126	Human	3-22 yr (IN)	Gastro			0.05 M	(gastrointestinal hemorrhages)	Morris et al. 1974 As(+3)	
			Hepatic			0.05 M	(vascular fibrosis, portal hypertension)		
			Dermal			0.05 M	(hyperpigmentation with keratoses, possibly pre-cancerous)		

Table 3-3 Levels of Significant Exposure to Inorganic Arsenic - Oral (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference Chemical Form	Comments
127	Human	15 yr (IN)	Hepatic			0.05 F (central fibrosis)	Piontek et al. 1989 As(+3)	
			Dermal			0.05 F (hyperkeratosis, possibly pre-cancerous)		
128	Human	NS (W)	Endocr			0.11 (diabetes mellitus)	Rahman et al. 1998 NS	
129	Human	NS (W)	Cardio	0.018	0.055 (hypertension)		Rahman et al. 1999 NS	

Table 3-3 Levels of Significant Exposure to Inorganic Arsenic - Oral (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference Chemical Form	Comments
130	Human	28 mo (IN)	Cardio	0.06 F			Silver and Wainman 1952 As(+3)	
			Gastro		0.06 F (intermittent, progressively severe nausea, cramps, and diarrhea)			
			Hemato	0.06 F				
			Hepatic		0.06 F (hepatomegaly, fatty liver)			
			Renal	0.06 F				
			Dermal			0.06 F (melanosis with hyperkeratosis, possibly pre-cancerous)		
			Ocular		0.06 F (conjunctival injection, periocular edema)			
131	Human	55 yr (IN)	Hepatic			0.03 M (portal fibrosis and hypertension, bleeding from esophageal varices)	Szuler et al. 1979 As(+3)	
			Dermal			0.03 M (hyperpigmentation with hyperkeratosis, possibly pre-cancerous)		
132	Human	45 yr (W)	Cardio			0.014 (Blackfoot disease)	Tseng 1977 NS	

Table 3-3 Levels of Significant Exposure to Inorganic Arsenic - Oral (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference Chemical Form	Comments
133	Human	NS (W)	Cardio			0.014 (Blackfoot disease)	Tseng 1989 NS	
134	Human	>45 yr (W)	Dermal	0.0008[d] M	0.014 M (hyperkeratosis and hyperpigmentation)		Tseng et al. 1968 NS	
135	Human	>30 yr (W)	Cardio		0.064 M (deficits in cutaneous microcirculation of the toes)		Tseng et al. 1995 As(+3)	
136	Human	52.6 yr (avg) (W)	Cardio	0.016		0.031 (peripheral vascular disease)	Tseng et al. 1996 NS	
137	Human	16 mo (IN)	Resp	0.1 M			Wade and Frazer 1953 As(+3)	
			Cardio	0.1 M				
			Hemato	0.1 M				
			Hepatic		0.1 M (liver enlargment)			
			Dermal			0.1 M (hyperkeratosis, hyperpigmentation with hyperkeratosis, possibly pre-cancerous)		

Table 3-3 Levels of Significant Exposure to Inorganic Arsenic - Oral (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference Chemical Form	Comments
138	Human	30-33 yr (W)	Dermal			0.015 M (hyperkeratosis of foot, possibly pre-cancerous)	Zaldivar 1974 NS	
139	Human	12 yr (W)	Resp		0.015[c] M (bronchitis, bronchiectasis)		Zaldivar 1974 NS	
					0.018 F (bronchitis, bronchiectasis)			
			Cardio			0.015[c] M (Raynaud's disease, thrombosis)		
						0.018 F		
			Gastro		0.015[c] M (diarrhea)			
					0.018 F (diarrhea)			
			Dermal		0.015[c] M (scaling of skin, hyperkeratosis, leukoderma, melanoderma)			
					0.018 F			
			Bd Wt		0.015[c] M (unspecified decreased body weight)			
					0.018 F (unspecified decreased body weight)			

Table 3-3 Levels of Significant Exposure to Inorganic Arsenic - Oral (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference Chemical Form	Comments
140	Human	NS (W)	Dermal			0.063 (hyperpigmentation with keratoses, possibly pre-cancerous)	Zaldivar 1977 NS	
141	Human	2-7 yr children (W)	Resp			0.08 (inflammation of bronchi and larynx, bronchopneumonia)	Zaldivar and Guillier 1977 NS	
			Cardio			0.05 (vascular spasms, thrombosis, ischemia, hypotension, cardiac failure)		
			Gastro			0.05 (nause, vomiting, diarrhea, intestinal hemorrhage)		
			Hemato			0.05 (anemia)		
			Hepatic			0.08 (cirrhosis)		
			Renal		0.08 (cloudy swelling in kidneys)			
			Dermal			0.05 (hyperkeratosis of palms and soles, melanoderma, leukoderma)		
142	Human	1-39 yr (W)	Cardio			0.06 (arterial thickening, Raynaud's disease)	Zaldivar and Guillier 1977 NS	

Table 3-3 Levels of Significant Exposure to Inorganic Arsenic - Oral (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference Chemical Form	Comments
143	Rat (Osborne-Mendel)	2 yr (F)	Resp	20			Byron et al. 1967 As(+3)	
			Cardio	20				
			Gastro	20				
			Hemato	9	20 (slight transient decrease in Hb and Hct values)			
			Hepatic	4		9 (enlarged bile duct, bile duct proliferation)		
			Renal	9	20 (pigmentation)			
			Bd Wt	2	4 (decreased body weight gain)			
144	Rat (Osborne-Mendel)	2 yr (F)	Resp	30			Byron et al. 1967 As(+5)	
			Cardio	30				
			Gastro	30				
			Hemato	30				
			Hepatic	9	20 (enlarged bile duct)			
			Renal	9	20 (pigmentation, cysts)			
			Bd Wt		2 (decreased body weight gain in females)			

Table 3-3 Levels of Significant Exposure to Inorganic Arsenic - Oral (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference Chemical Form	Comments
145	Rat (Wistar)	27 mo (F)	Resp	7			Kroes et al. 1974 As(+5)	
			Cardio	7				
			Gastro	7				
			Hemato	7				
			Musc/skel	7				
			Hepatic	7				
			Renal	7				
			Endocr	7				
			Bd Wt		7 (decreased body weight gain)			

Table 3-3 Levels of Significant Exposure to Inorganic Arsenic - Oral (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference Chemical Form	Comments
146	Rat (Wistar)	27 mo (F)	Resp	30			Kroes et al. 1974 As(+5) lead arsenate	
			Cardio	30				
			Gastro	30				
			Hemato	7	30 (slight anemia)			
			Musc/skel	30				
			Hepatic	7		30 (enlarged bile duct with extensive dilation and inflammation)		
			Renal	30				
			Endocr	30				
			Bd Wt	7	30 (decreased body weight gain)			
147	Rat (Long-Evans)	3 yr (W)	Resp	0.6			Schroeder et al. 1968 As(+3)	
			Cardio	0.6				
			Hepatic	0.6				
			Renal	0.6				
			Dermal	0.6				
			Bd Wt	0.6				

Table 3-3 Levels of Significant Exposure to Inorganic Arsenic - Oral (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference Chemical Form	Comments
148	Mouse (NS)	48 wk (W)	Hepatic	11.1			Liu et al. 2000 As(+3)	
			Renal		5.6 (histological alterations of the kidney)			
			Bd Wt	11.1				
149	Mouse (NS)	48 wk (W)	Hepatic	18.5			Liu et al. 2000 As (+5)	
			Renal		18.5 (increased relative kidney weight)			
			Bd Wt	18.5				
150	Mouse (BALB/c)	15 mo (W)	Hepatic		0.7 M (increased liver weight, altered liver histopathology, decreased hepatic enzymes in serum)		Santra et al. 2000 (NS)	
			Bd Wt		0.7 M (13-17% decreased body weight)			
151	Mouse (CD)	2 yr (W)	Bd Wt		1 (decreased body weight gain after the first 6 months of the study)		Schroeder and Balassa 1967 As(+3)	

Table 3-3 Levels of Significant Exposure to Inorganic Arsenic - Oral (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)		LOAEL Serious (mg/kg/day)		Reference Chemical Form	Comments
152	Dog (Beagle)	2 yr (F)	Resp	2.4					Byron et al. 1967 As(+3)	
			Cardio	2.4						
			Gastro	1			2.4	(bleeding in the gut)		
			Hemato	1	2.4	(slight to moderate anemia)				
			Hepatic	1	2.4	(hemosiderin deposits in hepatic macrophages)				
			Renal	2.4						
			Bd Wt	1			2.4	(44-61% weight loss)		
153	Dog (Beagle)	2 yr (F)	Resp	2.4					Byron et al. 1967 As(+5)	
			Cardio	2.4						
			Gastro	2.4						
			Hemato	1	2.4	(mild anemia)				
			Hepatic	1	2.4	(pigmentation in hepatic macrophages)				
			Renal	2.4						
			Bd Wt	1			2.4	(marked decreased weight gain)		

Table 3-3 Levels of Significant Exposure to Inorganic Arsenic - Oral (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference Chemical Form	Comments
Neurological								
154	Human	>5 yr (W)		0.006[c] M 0.007 F			EPA 1981b NS	
155	Human	3-7 yr (W)				0.11 F (wrist weakness)	Foy et al. 1992 NS	
156	Human	1-20 yr (W)			0.06 (tingling of hands and feet)		Guha Mazumder et al. 1988 NS	
157	Human	10 yr (W)		0.0046			Harrington et al. 1978 NS	
158	Human	NS (W)		0.0014		0.04 (functional denervation)	Hindmarsh et al. 1977 NS	
159	Human	NS (W)		0.004	0.005 (fatigue, headache, dizziness, insomnia, nighmare, numbness)		Lianfang and Jianzhong 1994 NS	
160	Human	28 mo (IN)				0.06 F (paresthesia)	Silver and Wainman 1952 As(+3)	
161	Human	55 yr (IN)			0.03 M (absent ankle jerk reflex and vibration sense in legs)		Szuler et al. 1979 As(+3)	

Table 3-3 Levels of Significant Exposure to Inorganic Arsenic - Oral (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference Chemical Form	Comments
162	Human	NS (W)			0.0017 (decreased performance in neurobehavioral tests)		Tsai et al. 2003 (NS)	
163	Human	lifetime continuous (W)			0.005 (decreased performance in neurobehavioral tests)		Wasserman et al. 2004 (NS)	
164	Human	lifetime		0.0008	0.003 (decreased score in Performance domain of an intelligence scale)		Wasserman et al. 2007 (NS)	
Reproductive								
165	Human	NS (W)				0.008 F (increased frequencies for spontaneous abortion, stillbirth, and preterm birth rates)	Ahmad et al. 2001 (NS)	98% of the exposed group drank water containing 0.1 mg As/L or more.
166	Human	lifetime (W)				0.006 F (increased incidence of spontaneous abortion)	Milton et al. 2005 (NS)	
167	Human	lifetime (W)				0.02 F (increased risk of stilbirth)	von Ehrenstein et al. 2006 (NS)	

Table 3-3 Levels of Significant Exposure to Inorganic Arsenic - Oral (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference Chemical Form	Comments
Developmental								
168	Human	continuous (W)			0.002 (reduced birth weight)		Hopenhayn et al. 2003a (NS)	
169	Human	>1 yr 1 x/d (W)				0.03 (increased SMR for malignant and non-malignant lung disease)	Smith et al. 2006 (NS)	
170	Human	lifetime (W)		0.008			von Ehrenstein et al. 2006 (NS)	NOAEL is for no increase in risk of neonatal mortality or overall infant mortality.
Cancer								
171	Human	NS (W)				0.022 (CEL: skin cancer)	Cebrian et al. 1983 As(+5)	
172	Human	NS (W)				0.064 (CEL: bladder, lung and liver cancers)	Chen et al. 1986 NS	
173	Human	NS (W)				0.064 (CEL: malignant neoplasms of the bladder, skin, lung and liver)	Chen et al. 1988b NS	
174	Human	NS (W)				0.003 (CEL: bladder cancer)	Chiou et al. 2001 (NS)	

Table 3-3 Levels of Significant Exposure to Inorganic Arsenic - Oral (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)		Reference Chemical Form	Comments
175	Human	2 wk- 12 yr (IN)				3.67	(CEL: bladder cancer risk)	Cuzick et al.1992 As(+3)	
176	Human	NS (W)				0.0011	(CEL: lung cancer)	Ferreccio et al. 1998 NS	
177	Human	NS (W)				0.0017	(CEL: lung cancers)	Ferreccio et al. 2000 (NS)	
178	Human	NS (W)				0.018	(CEL: lung cancer mortality)	Guo 2004 (NS)	
179	Human	NS (W)				0.018	(CEL: bladder cancer)	Guo and Tseng 2000 (NS)	
180	Human	NS (W)				0.052	(CEL: increased incidence of transitional cell carcinomas of the bladder, kidney, ureters, and all urethral cancer)	Guo et al. 1997 NS	
181	Human	NS (W)				0.0049[c] M	(CEL: squamous cell carcinoma of the skin)	Guo et al. 2001b (NS)	
						0.0094 F			

Table 3-3 Levels of Significant Exposure to Inorganic Arsenic - Oral (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)		Reference Chemical Form	Comments
182	Human	>1 yr (W)				0.0075	(CEL: basal or squamous skin carcinoma)	Haupert et al. 1996 NS	
183	Human	16 yr (avg) (IN)				0.04 M	(CEL: basal cell and squamous cell carcinomas of the skin, small cell and squamous cell carcinoma of the lung)	Luchtrath 1983 As(+5)	
184	Human	60 yr (W)				0.038	(CEL: intraepidermal carcinoma)	Tseng 1977 NS	
185	Human	>45 yr (W)				0.014	(CEL: squamous cell carcinoma of the skin)	Tseng et al. 1968 NS	
186	Human	~5 yr (W)				0.033	(CEL: lung, urinary tract cancer)	Tsuda et al. 1995a As(+3)	

Table 3-3 Levels of Significant Exposure to Inorganic Arsenic - Oral (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference Chemical Form	Comments
187	Human	12 yr (W)				0.015[c] M (CEL: squamous cell carcinoma of the skin)	Zaldivar 1974 NS	
						0.018 F (CEL: squamous cell carcinoma of the skin)		
188	Human	22-34 yr (W)				0.014 M (CEL: basal cell and squamous cell carcinomas of the skin, hemangioendothelioma of the liver)	Zaldivar et al. 1981 NS	

a The number corresponds to entries in Figure 3-3.

b Used to derive provisional acute oral minimal risk level (MRL) of 0.005 mg/kg/day; dose divided by an uncertainty factor of 10 (for extrapolation from a LOAEL to a NOAEL).

c Differences in levels of health effects and cancer effects between male and females are not indicated in Figure 3-3. Where such differences exist, only the levels of effect for the most sensitive gender are presented.

d Used to derive chronic oral minimal risk level (MRL) of 0.0003 mg/kg/day; dose divided by an uncertainty factor of 3 (for human variability).

avg = average; ALAD = delta-aminolevulinic acid dehydratase; ALAS = delta-aminolevulinic acid synthetase; ALT = alanine aminotransferase; AST = aspartate aminotransferase; Bd Wt = body weight; BUN = blood urea nitrogen; Cardio = cardiovascular; CEL = cancer effect level; d = day(s); Endocr = endocrine; (F) = feed; F = female; (G) = gavage; Gastro = gastrointestinal; Gd = gestational day; GI = gastrointestinal; (GW) = gavage in water; gen = generation; Gd = gestation day; Gn pig = guinea pig; Hemato = hematological; Hb = hemoglobin; Hct = hematocrit; Hemato = hematological; hr = hour(s); (IN) = ingestion; LD50 = lethal dose, 50% kill; LDH = lactate dehydrogenase; LOAEL = lowest-observable-adverse-effect level; M = male; Metab = metabolic; mo = month(s); Musc/skel = musculoskeletal; NOAEL = no-observable-adverse-effect level; NS = not specified; pmd = pre-mating day; pnd = post-natal day; Resp = respiratory; SMR =standardized mortality ratio; (W) = drinking water; wk = week(s); x = time(s); yr = year(s)

Figure 3-3 Levels of Significant Exposure to Inorganic Arsenic - Oral

Acute (≤14 days)

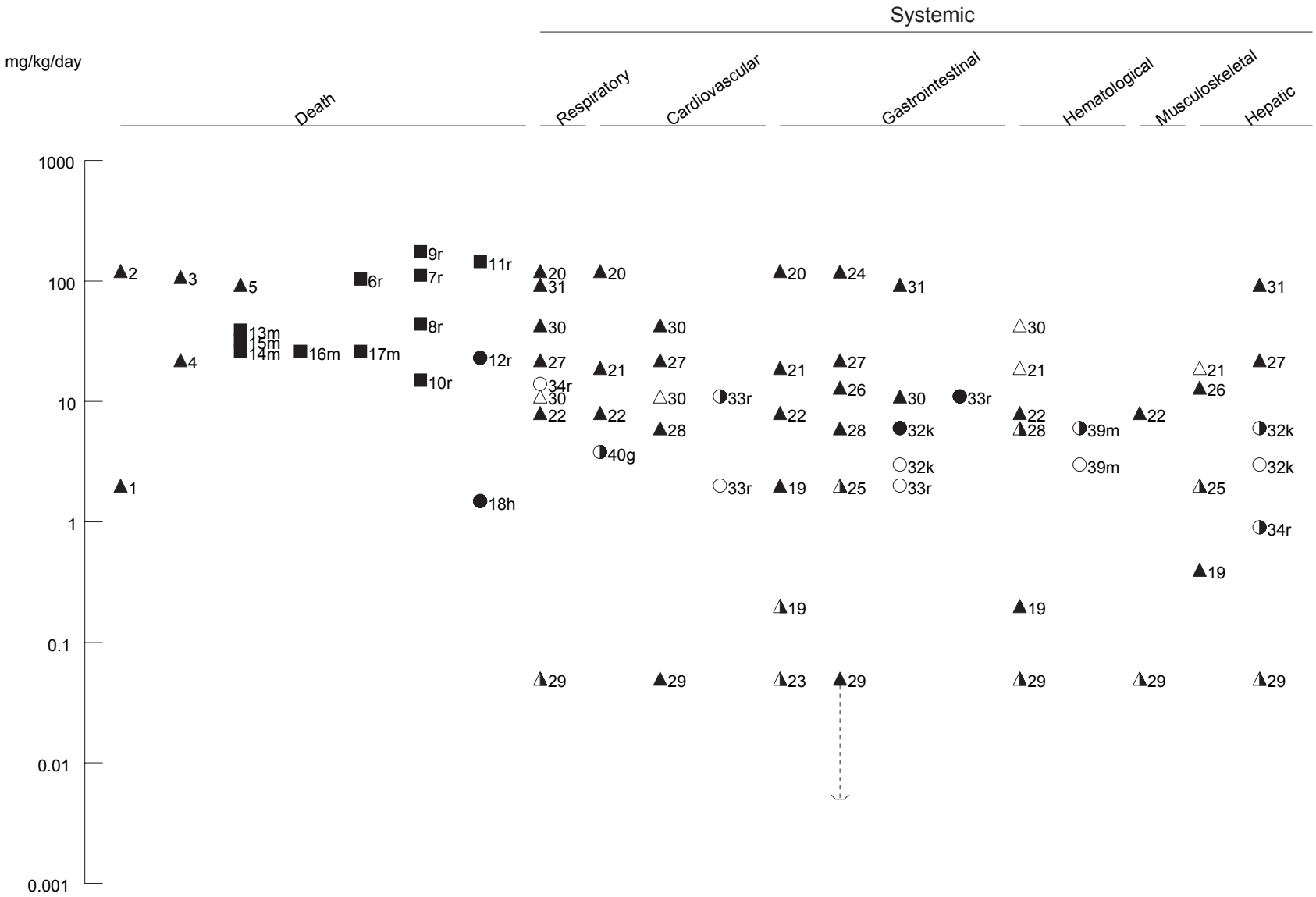


Figure 3-3 Levels of Significant Exposure to Inorganic Arsenic - Oral *(Continued)*

Acute (≤14 days)

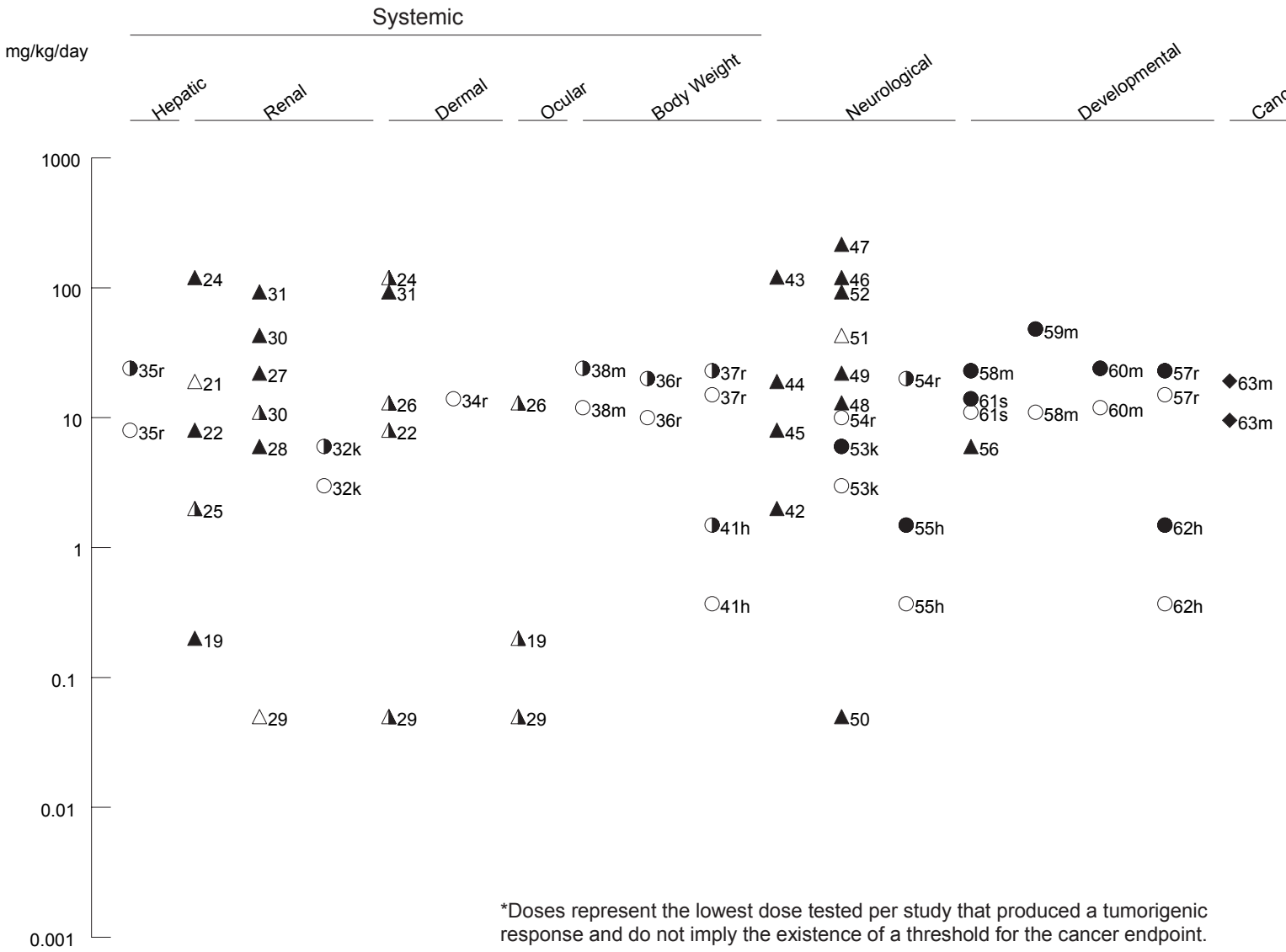


Figure 3-3 Levels of Significant Exposure to Inorganic Arsenic - Oral *(Continued)*

Intermediate (15-364 days)

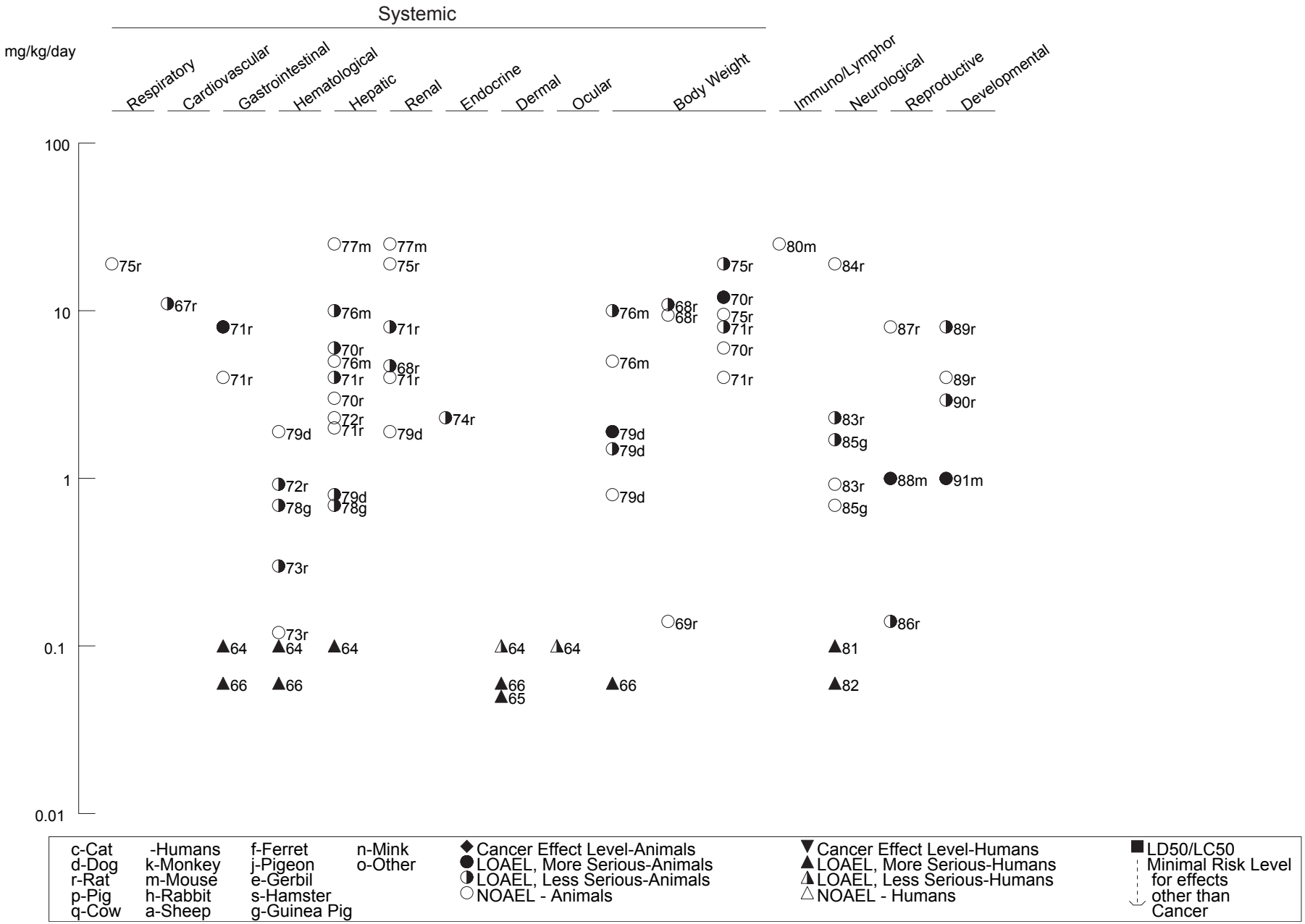


Figure 3-3 Levels of Significant Exposure to Inorganic Arsenic - Oral *(Continued)*

Chronic (≥365 days)

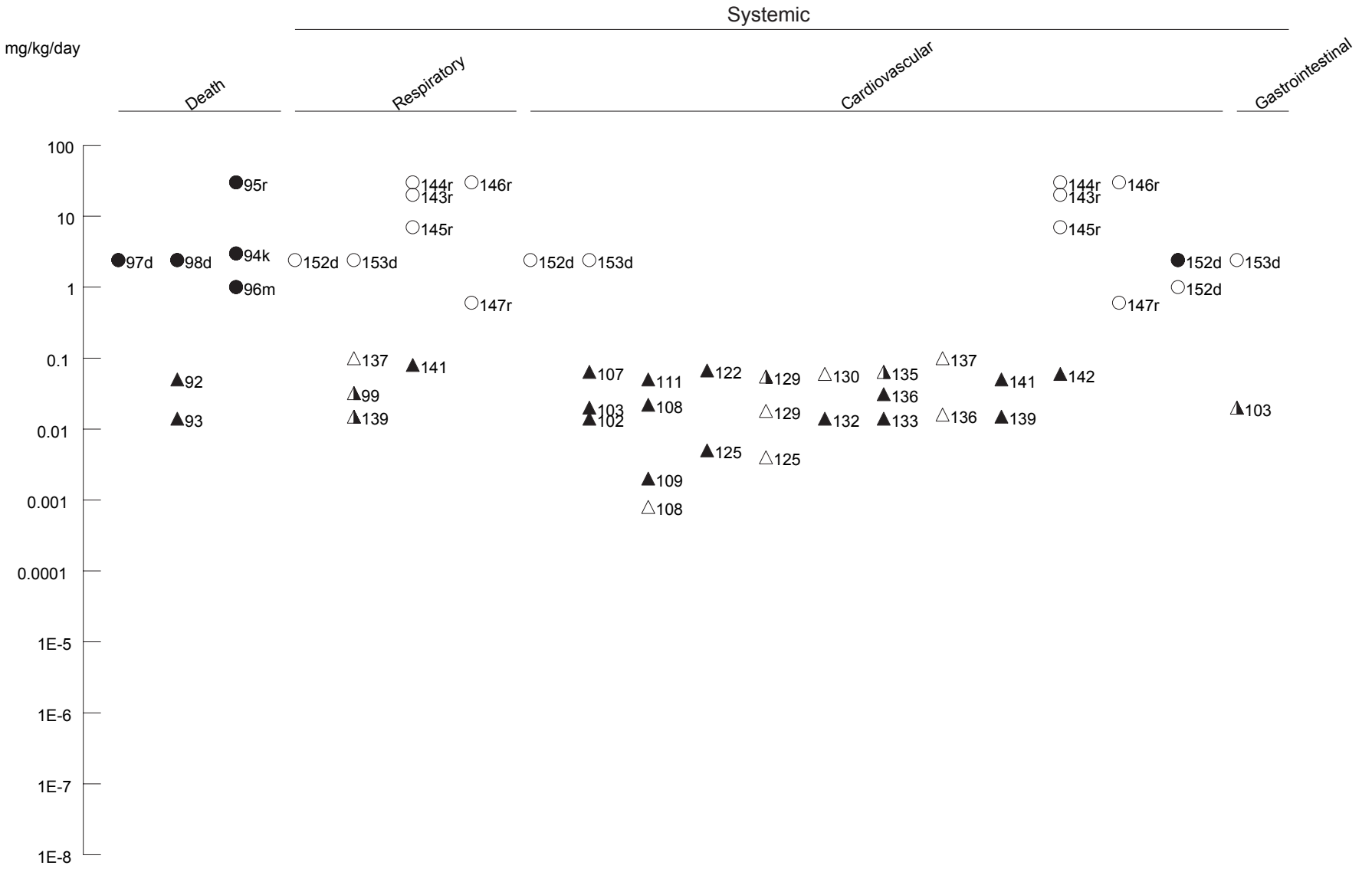


Figure 3-3 Levels of Significant Exposure to Inorganic Arsenic - Oral *(Continued)*

Chronic (≥365 days)

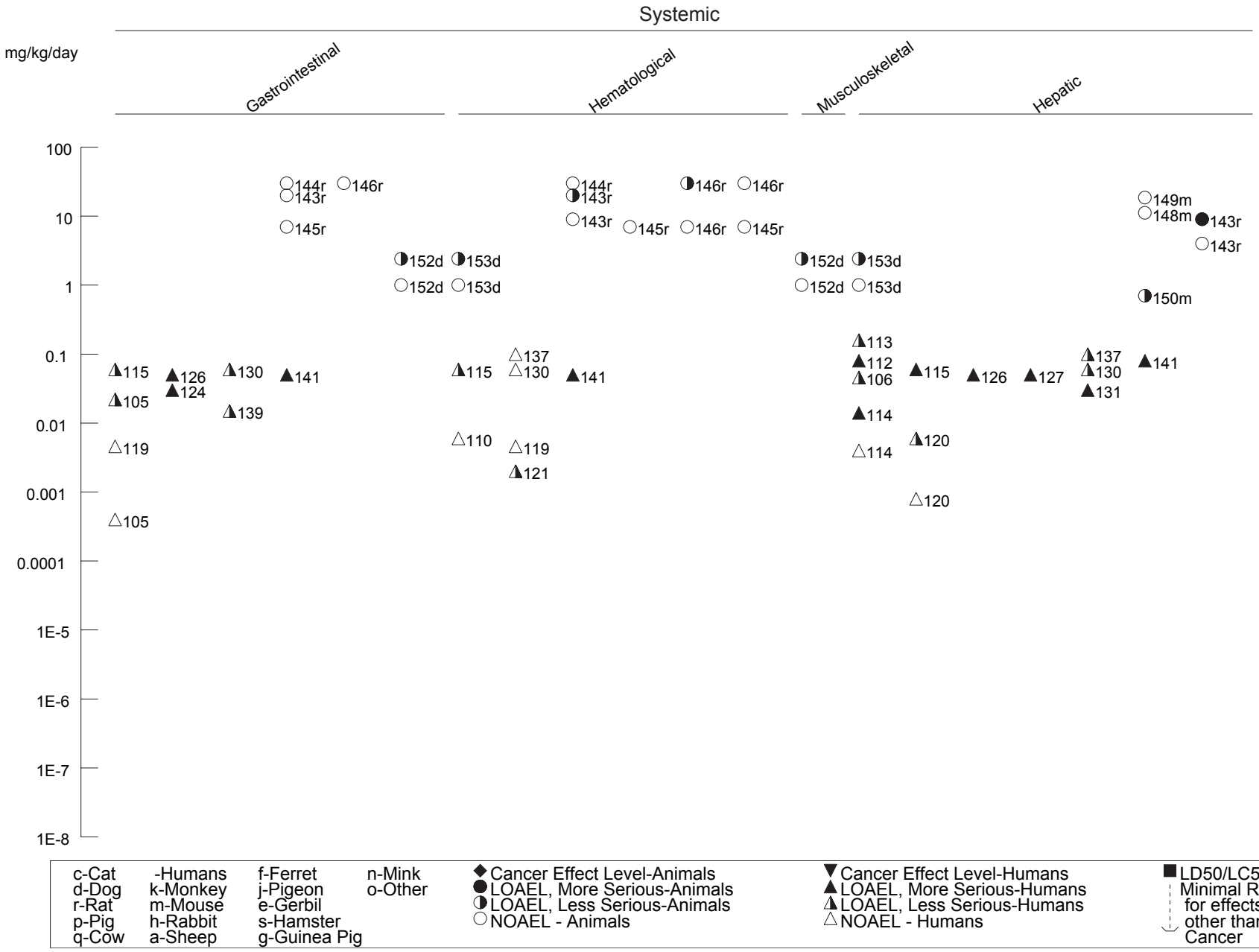
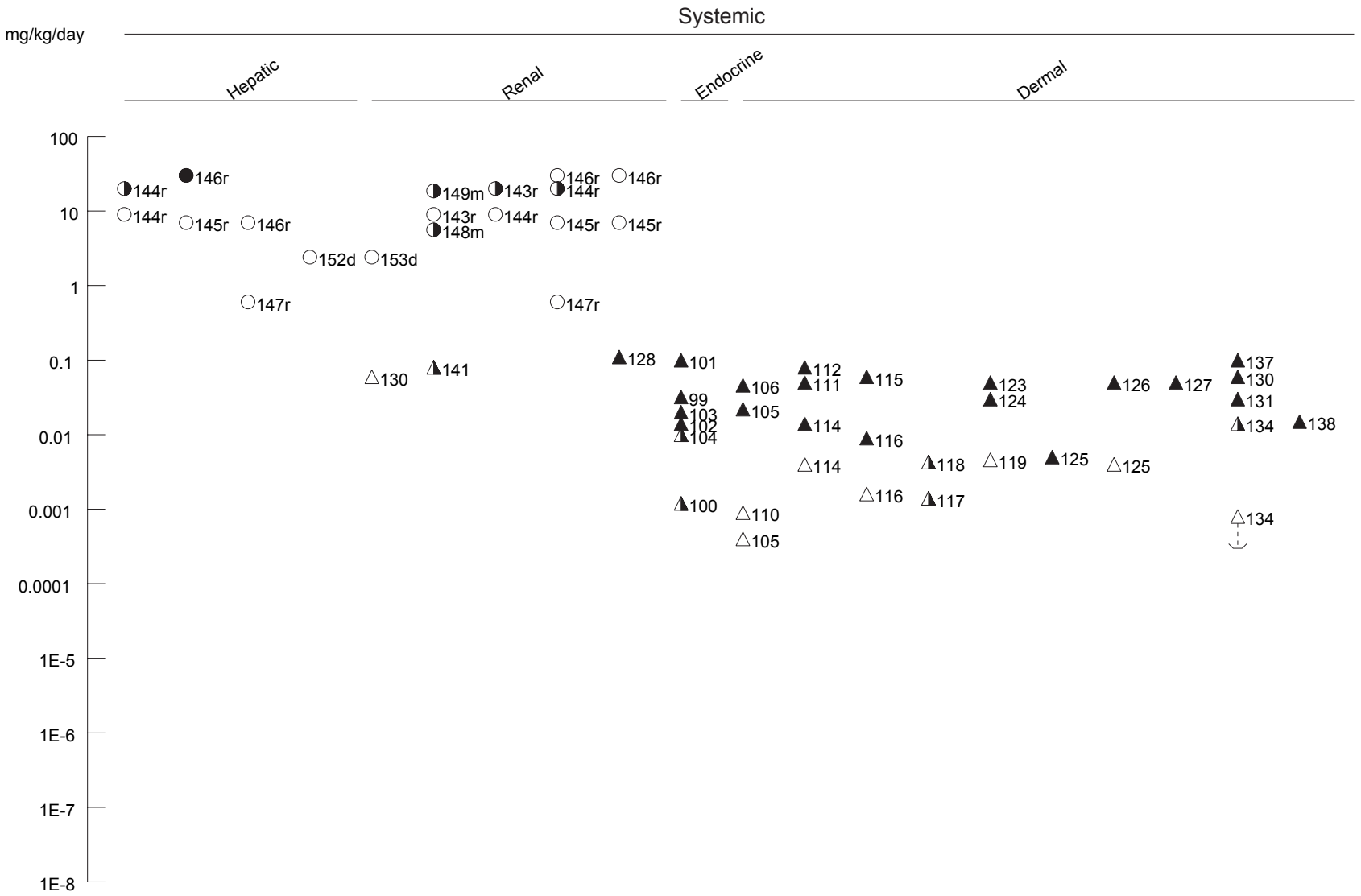


Figure 3-3 Levels of Significant Exposure to Inorganic Arsenic - Oral *(Continued)*

Chronic (≥365 days)



c-Cat	-Humans	f-Ferret	n-Mink
d-Dog	k-Monkey	j-Pigeon	o-Other
r-Rat	m-Mouse	e-Gerbil	
p-Pig	h-Rabbit	s-Hamster	
q-Cow	a-Sheep	g-Guinea Pig	

◆ Cancer Effect Level-Animals
● LOAEL, More Serious-Animals
◑ LOAEL, Less Serious-Animals
○ NOAEL - Animals

▼ Cancer Effect Level-Humans
▲ LOAEL, More Serious-Humans
◭ LOAEL, Less Serious-Humans
△ NOAEL - Humans

■ LD50/LC50
Minimal Risk Level for effects other than Cancer

Figure 3-3 Levels of Significant Exposure to Inorganic Arsenic - Oral *(Continued)*

Chronic (≥365 days)

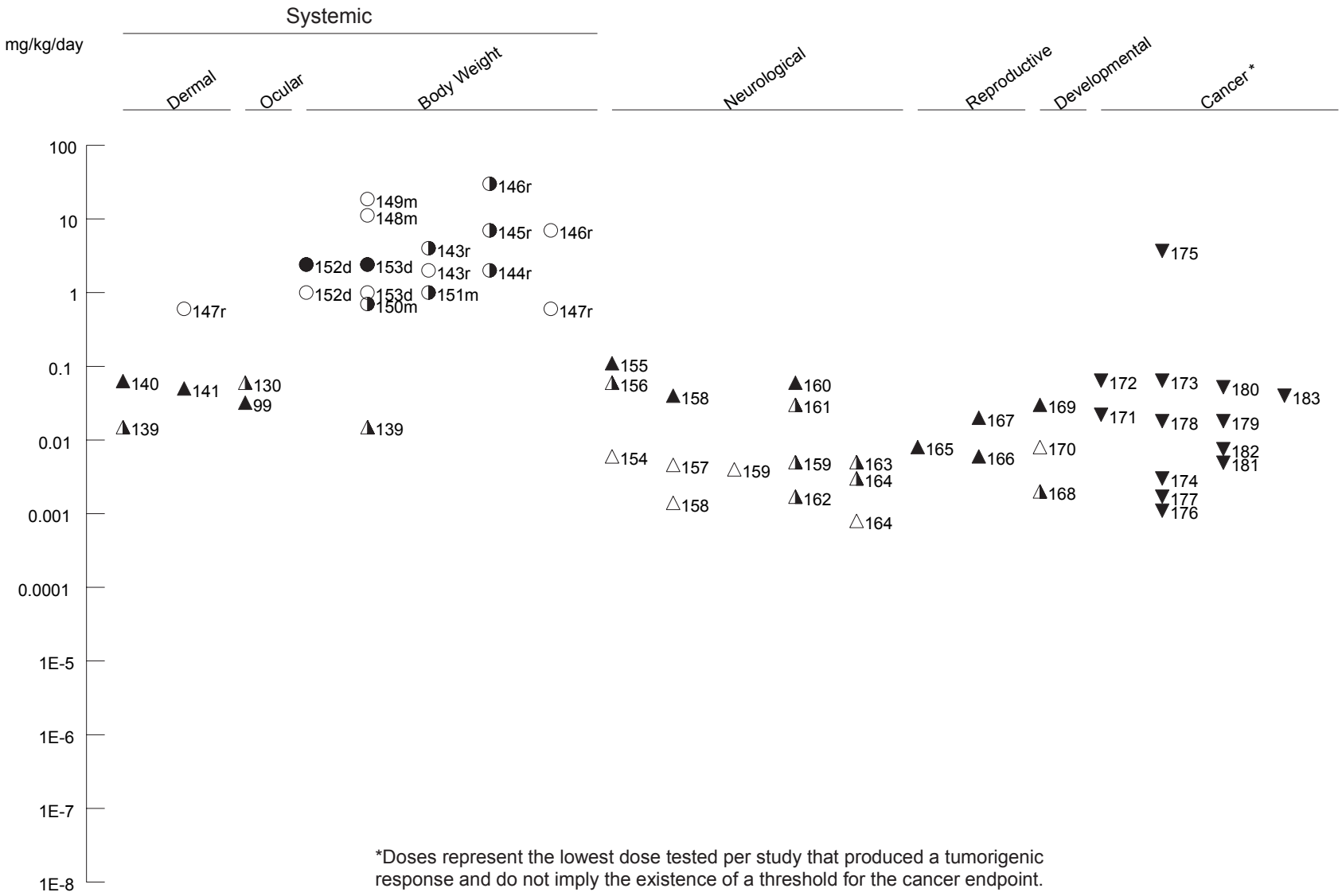


Figure 3-3 Levels of Significant Exposure to Inorganic Arsenic - Oral *(Continued)*

Chronic (≥365 days)

*Doses represent the lowest dose tested per study that produced a tumorigenic response and do not imply the existence of a threshold for the cancer endpoint.

c-Cat	-Humans	f-Ferret	n-Mink	◆ Cancer Effect Level-Animals	▼ Cancer Effect Level-Humans	■ LD50/LC50
d-Dog	k-Monkey	j-Pigeon	o-Other	● LOAEL, More Serious-Animals	▲ LOAEL, More Serious-Humans	Minimal Risk Level for effects other than Cancer
r-Rat	m-Mouse	e-Gerbil		◑ LOAEL, Less Serious-Animals	◭ LOAEL, Less Serious-Humans	
p-Pig	h-Rabbit	s-Hamster		○ NOAEL - Animals	△ NOAEL - Humans	
q-Cow	a-Sheep	g-Guinea Pig				

Table 3-4 Levels of Significant Exposure to Monomethylarsonic Acid - Oral

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference Chemical Form	Comments
ACUTE EXPOSURE								
Death								
1	Rat (Sprague-Dawley)	once (GW)				3184 M (LD50) 2449[b] F (LD50)	Gur and Nyska 1990 MSMA	
2	Mouse (ddY)	once (GW)				1800 M (LD50)	Kaise et al. 1989 MMA	
3	Rabbit (New Zealand)	once (GW)				102 M (LD50)	Jaghabir et al. 1988 MSMA	
Systemic								
4	Rat (Sprague-Dawley)	once (GW)	Gastro		2030 (mucoid feces and diarrhea)		Gur and Nyska 1990 MSMA	
			Bd Wt	2030				
5	Rat (Sprague-Dawley)	Gd 6-15 (GW)	Bd Wt	10 F	100 F (17% decrease in maternal body weight gain)	500 F (40% decrease in maternal body weight gain)	Irvine et al. 2006 MMA	
6	Mouse (ddY)	once (GW)	Resp			1800 M (respiratory arrest)	Kaise et al. 1989 MMA	
			Gastro		2200 M (diarrhea, slight congestion of the small intestine)			

Table 3-4 Levels of Significant Exposure to Monomethylarsonic Acid - Oral (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference Chemical Form	Comments
7	Rabbit (New Zealand)	Gd 7-19 (GW)	Gastro	7 F	12 F (loose feces/diarrhea in 7/14 pregnant rabbits)		Irvine et al. 2006 MMA	
			Bd Wt	7 F		12 F (67% decrease in maternal body weight gain)		
8	Rabbit (New Zealand)	once (GW)	Gastro		60 M (diarrhea)		Jaghabir et al. 1988 MSMA	
Developmental								
9	Rat (Sprague-Dawley)	Gd 6-15 (GW)		100	500 (decreased fetal weight and increased fetal incidence of imcomplete ossification of thoracic vertebrae)		Irvine et al. 2006 MMA	
10	Rabbit (New Zealand)	Gd 7-19 (GW)		7	12 (supernumerary thoracic ribs and eight lumbar vertebrae)		Irvine et al. 2006 MMA	
INTERMEDIATE EXPOSURE								
Death								
11	Rat (Fischer- 344)	52 wk (F)				106.9 M (increased mortality)	Arnold et al. 2003 MMA	

Table 3-4 Levels of Significant Exposure to Monomethylarsonic Acid - Oral (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference Chemical Form	Comments
Systemic								
12	Rat (Fischer- 344)	52 wk (F)	Gastro	3.5[c] M	30.2 M (diarrhea)		Arnold et al. 2003 MMA	
			Bd Wt	30.2 M	106.9 M (14% decrease in body weight)			
13	Rat (Sprague-Dawley)	146-171 d pre-mating, mating, gestation, and lactation (F)	Bd Wt	76			Schroeder 1994 MMA	
14	Dog (Beagle)	52 wk (C)	Gastro		2 M (diarrhea)		Waner and Nyska 1988 MMA	
			Bd Wt	2 F	8 F (decrease in body weight)			
Reproductive								
15	Rat (Sprague-Dawley)	146-171 d pre-mating, mating, gestation, and lactation (F)		22	76 (decreased pregnancy rate and male fertility index in F0 and F1)		Schroeder 1994 MMA	
16	Mouse (Swiss)	19 d 3 d/wk (GW)			119 M (reduced fertility)		Prukop and Savage 1986 MSMA	

Table 3-4 Levels of Significant Exposure to Monomethylarsonic Acid - Oral (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference Chemical Form	Comments
Developmental								
17	Rat (Sprague-Dawley)	146-171 d pre-mating, mating, gestation, and lactation (F)		22	76 (decreased pup survival F1 and F2)		Schroeder 1994 MMA	
CHRONIC EXPOSURE								
Death								
18	Rat (Fischer- 344)	104 wk (F)				72.4 M (increased mortality)	Arnold et al. 2003 MMA	

Table 3-4 Levels of Significant Exposure to Monomethylarsonic Acid - Oral (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference Chemical Form	Comments
Systemic								
19	Rat (Fischer- 344)	104 wk (F)	Gastro	3 M	25.7 M (diarrhea)	72.4 M (necrosis, ulceration, perforation in large intestine)	Arnold et al. 2003 MMA	
			Hemato	72.4 M				
			Musc/skel	72.4 M				
			Hepatic	72.4 M				
			Renal	3.9 F	33.9 F (increased absolute kidney weight and progressive glomerulonephropathy)			
			Endocr	3.9 F	33.9 F (hypertrophy of thyroid follicular epithelium, decreased absolute thyroid weight)			
			Dermal	72.4 M				
			Ocular	72.4 M				
			Bd Wt	3 M	25.7 M (15% decrease in body weight)	33.9 F (30% decrease in body weight)		

Table 3-4 Levels of Significant Exposure to Monomethylarsonic Acid - Oral (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference Chemical Form	Comments
20	Rat (Fischer- 344)	104 wk (W)	Hemato	8.4 M			Shen et al. 2003 MMA	
			Hepatic	2.1 M	8.4 M (increased GST-P-positive foci)			
			Renal		2.1 M (hyperplasia of the bladder)			
			Bd Wt	8.4 M				
21	Mouse (B6C3F1)	104 wk (F)	Cardio	67.1 M			Arnold et al. 2003 MMA	
			Gastro	24.9 M	67.1 M (loose and mucoid feces, metaplasia of the cecum and colon)			
			Musc/skel	67.1 M				
			Hepatic	67.1 M				
			Renal	1.2[d] M	6 M (increased incidence of progressive glomeruloephropathy)			
			Dermal	67.1 M				
			Ocular	67.1 M				
			Bd Wt	24.9 M	67.1 M (17% decrease in body weight)			

Table 3-4 Levels of Significant Exposure to Monomethylarsonic Acid - Oral (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference Chemical Form	Comments
22	Dog (Beagle)	52 wk (C)	Resp	35			Waner and Nyska 1988 MMA	
			Gastro		2 M (diarrhea)			
			Hemato	35				
			Hepatic	35				
			Renal		8 (increased urine specific gravity; increased kidney weight)			
			Ocular	35				
			Bd Wt	2 F		8 F (42% decrease in body weight)		
Neurological								
23	Dog (Beagle)	52 wk (C)		35			Waner and Nyska 1988 MMA	
Reproductive								
24	Dog (Beagle)	52 wk (C)		35 M 8 F	35 F (decrease in estrus)		Waner and Nyska 1988 MMA	Histological examination of reproductive tissues.

Table 3-4 Levels of Significant Exposure to Monomethylarsonic Acid - Oral (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference Chemical Form	Comments
Cancer								
25	Rat (Fischer- 344)	104 wk (W)		8.4 M			Shen et al. 2003 MMA	

a The number corresponds to entries in Figure 3-4.

b Differences in levels of health effects and cancer effects between male and females are not indicated in Figure 3-4. Where such differences exist, only the levels of effect for the most sensitive gender are presented.

c The intermediate-duration oral MRL of 0.1 mg MMA/kg/day was calculated using a benchmark dose analysis. The BMDL10 of 12.38 mg MMA/kg/day was divided by an uncertainty factor of 100 (10 to account for extrapolation from animals to humans and 10 for human variability)

d The chronic-duration oral MRL of 0.01 mg MMA/kg/day was calculated using a benchmark dose analysis. The BMDL10 of 1.09 mg MMA/kg/day was divided by an uncertainty factor of 100 (10 to account for extrapolation from animals to humans and 10 for human variability)

Bd Wt = body weight; (C) = capsule; Cardio = cardiovascular; d = day(s); Endocr = endocrine; (F) = feed; F = female; Gastro = gastrointestinal; (GO) = gavage in oil; (GW) = gavage in water; Gd = gestation day; GST-P = glutathione S-transferase placental form; Hemato = hematological; IN = ingestion; LD50 = lethal dose, 50% kill; LOAEL = lowest-observable-adverse-effect level; M = male; MMA = monomethylarsonic acid; MSMA = monosodium methane arsonate; Musc/skel = musculoskeletal; NOAEL = no-observable-adverse-effect level; Resp = respiratory; (W) = drinking water; wk = week(s); x = time(s)

Figure 3-4 Levels of Significant Exposure to Monomethylarsonic Acid - Oral

Acute (≤14 days)

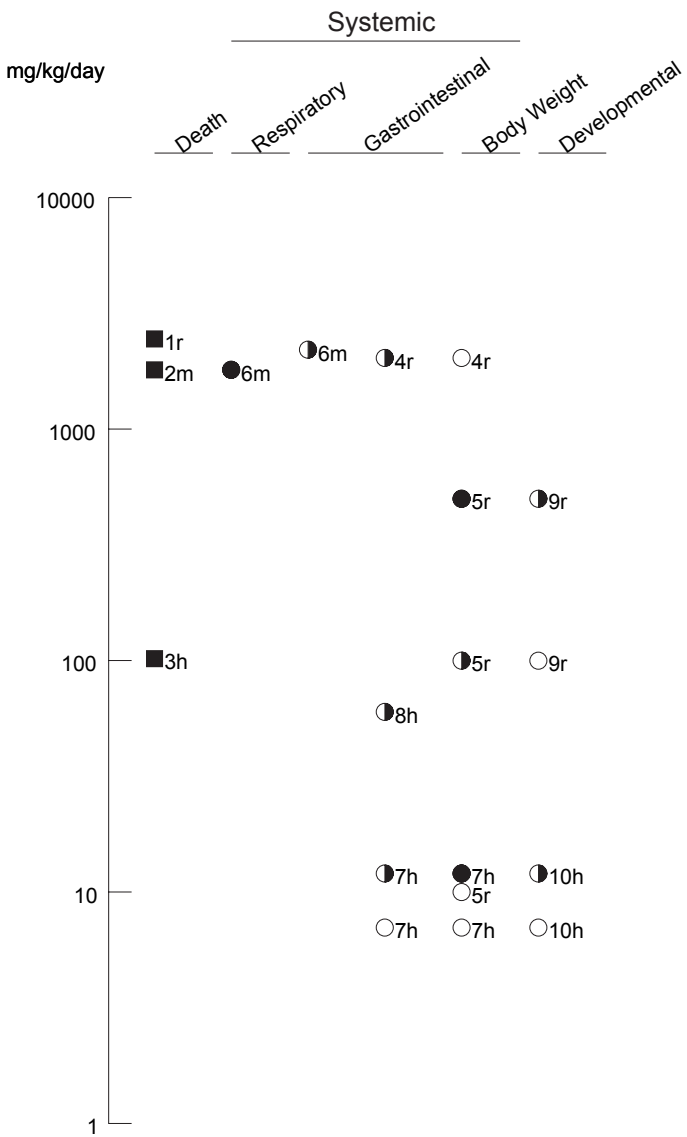


Figure 3-4 Levels of Significant Exposure to Monomethylarsonic Acid - Oral *(Continued)*

Intermediate (15-364 days)

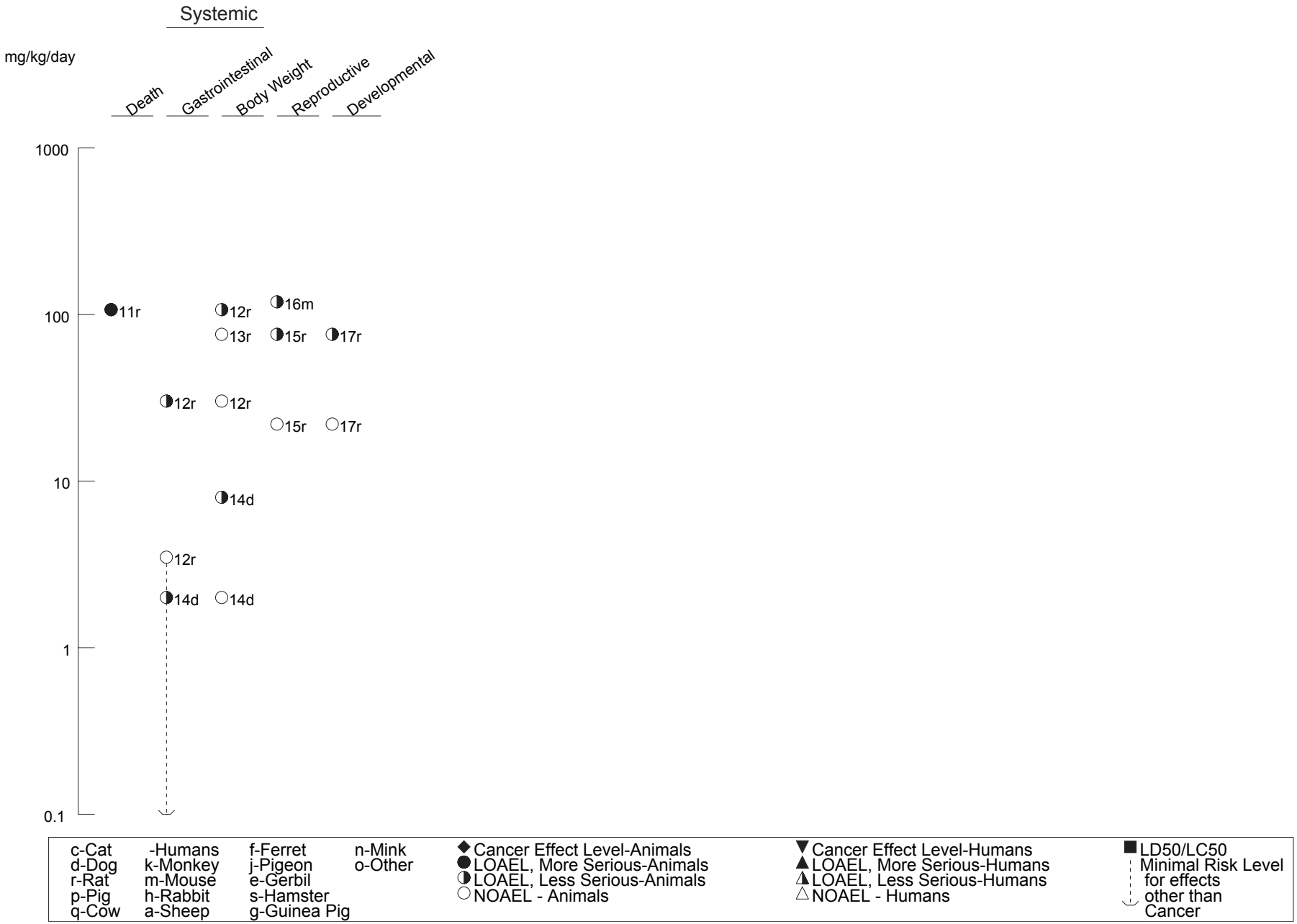


Figure 3-4 Levels of Significant Exposure to Monomethylarsonic Acid - Oral *(Continued)*

Chronic (≥365 days)

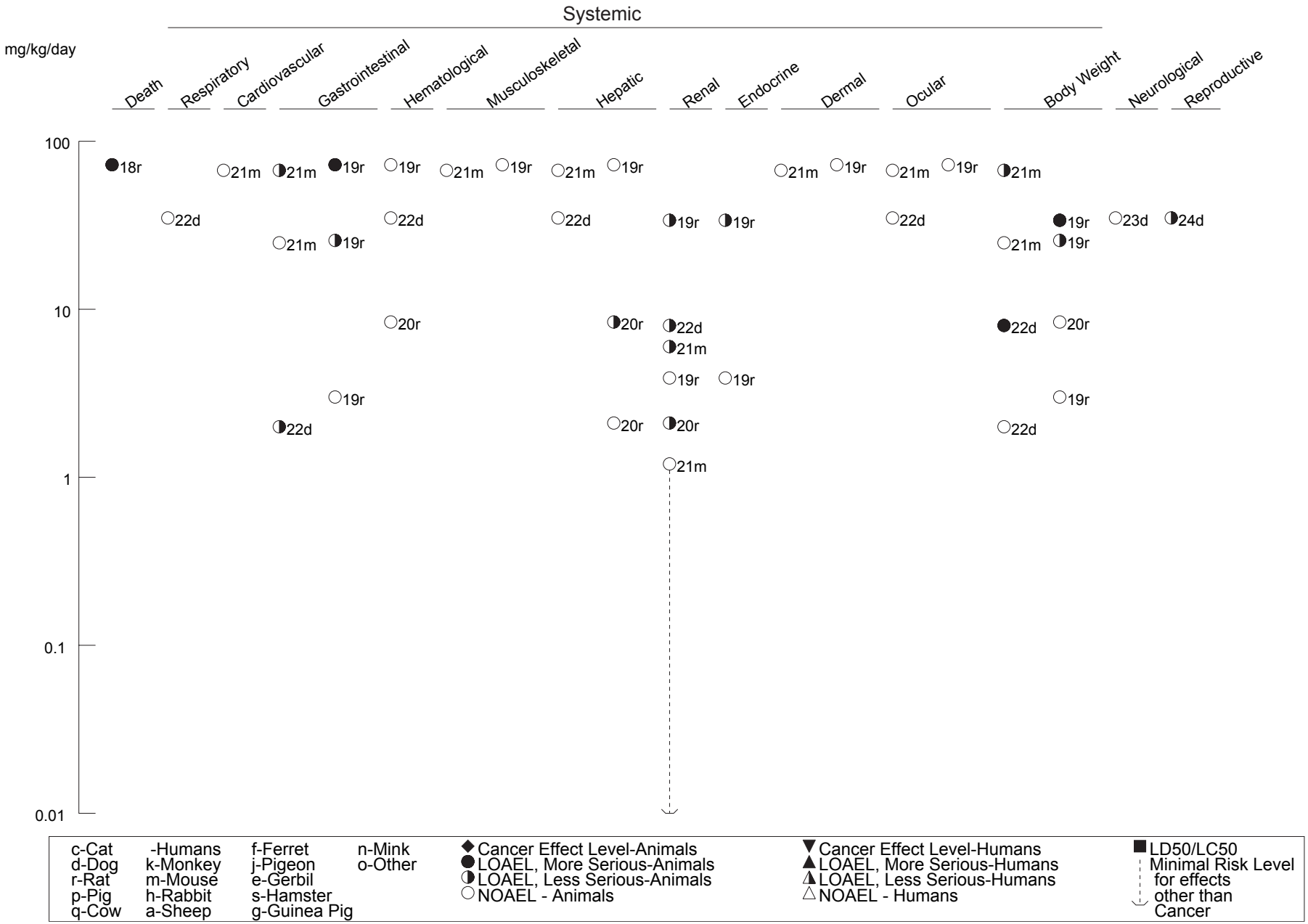


Figure 3-4 Levels of Significant Exposure to Monomethylarsonic Acid - Oral *(Continued)*

Chronic (≥365 days)

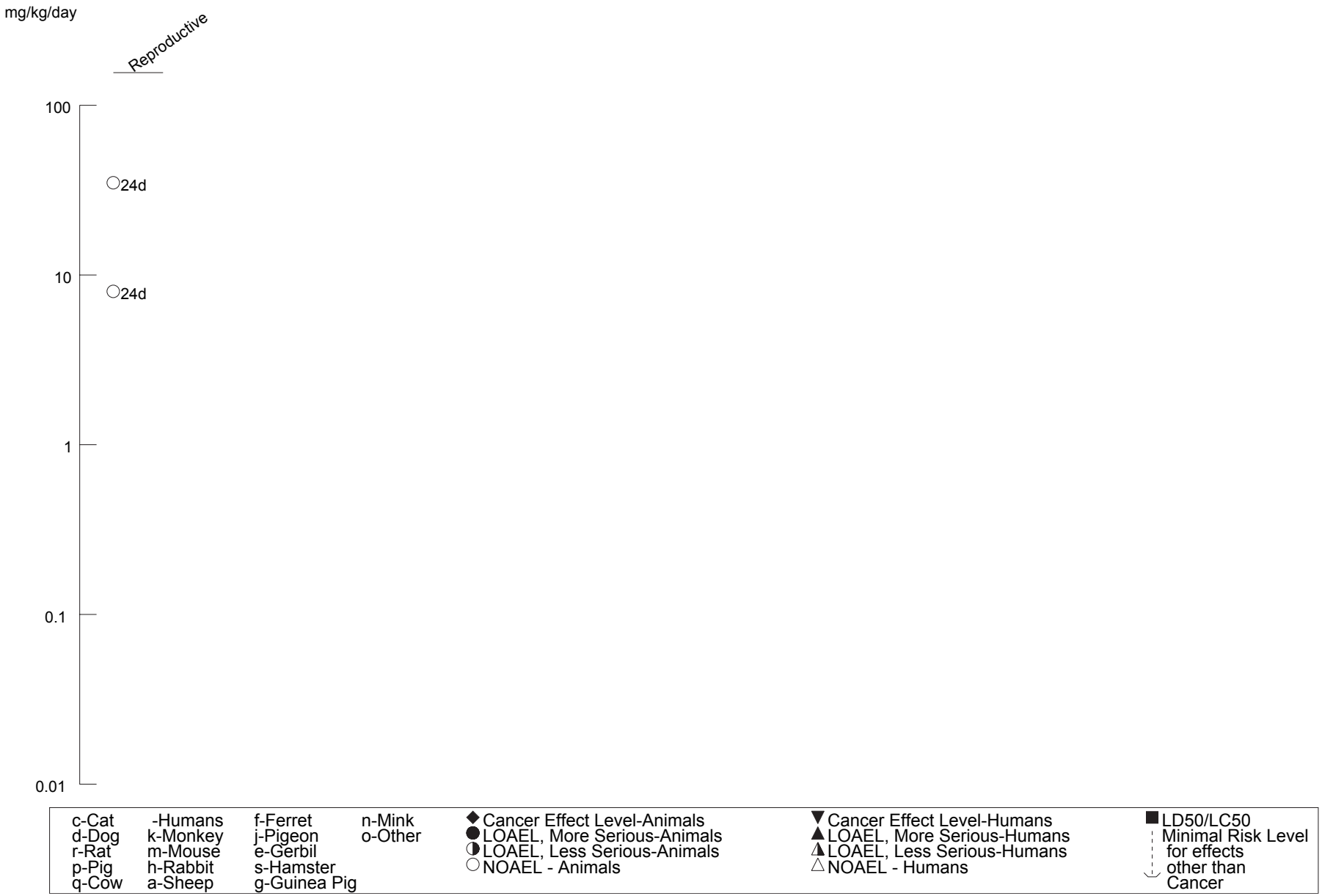


Table 3-5 Levels of Significant Exposure to Dimethylarsinic Acid - Oral

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference Chemical Form	Comments
ACUTE EXPOSURE								
Death								
1	Rat (Fischer- 344)	13 wk (F)				475 M (100% mortality during first 2 weeks of study)	Crown et al. 1987 DMA	
2	Rat (CD)	10 d Gd 7-16 1 x/d (GW)				60 F (67% mortality)	Rogers et al. 1981 DMA	
3	Mouse (ddY)	once (GW)				1200 M (LD50)	Kaise et al. 1989 DMA	
4	Mouse (CD-1)	10 d Gd 7-16 1 x/d (GW)				600 F (59% mortality)	Rogers et al. 1981 DMA	
Systemic								
5	Rat (Fischer- 344)	2 wk (F)	Renal		11 F (altered bladder cell surface characteristics)		Cohen et al. 2001 DMA	
			Bd Wt	11 F				
6	Rat (Fischer- 344)	13 wk (F)	Gastro		475 M (diarrhea and congestion and hemorrhagic contents in gastrointestinal tract in rats dying during first 2 weeks)		Crown et al. 1987 DMA	

Table 3-5 Levels of Significant Exposure to Dimethylarsinic Acid - Oral (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference Chemical Form	Comments
7	Rat (Sprague-Dawley)	Gd 6-15 (GW)	Bd Wt	12 F	36 F (decreased maternal body weight gain)		Irvine et al. 2006 DMA	
8	Rat (CD)	10 d Gd 7-16 1 x/d (GW)	Bd Wt			40 F (27% decreased maternal weight gain)	Rogers et al. 1981 DMA	
9	Mouse (B6C3F1)	24 hr 1 or 2 x (GW)	Resp		720 F (decreased lung ODC)		Ahmad et al. 1999a DMA	
			Hepatic		720 F (decreased liver GSH, GSSG, CYP-450 and ODC; increased serum ALT)			
10	Mouse (ddY)	once (GW)	Resp			900 M (respiratory arrest)	Kaise et al. 1989 DMA	
			Gastro		1757 M (diarrhea, slight congestion of the intestion)			
11	Mouse (CD-1)	10 d Gd 7-16 1 x/d (GW)	Bd Wt			200 F (26% decreased maternal weight gain)	Rogers et al. 1981 DMA	
12	Dog (Beagle)	52 wk (C)	Gastro	6.5	16 (vomiting and diarrhea)		Zomber et al. 1989 DMA	

Table 3-5 Levels of Significant Exposure to Dimethylarsinic Acid - Oral (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference Chemical Form	Comments
13	Rabbit (New Zealand)	Gd 7-19 (GW)	Gastro	12 F	48 F (fluid gastrointestinal tract contents)		Irvine et al. 2006 DMA	
			Bd Wt	12 F		48 F (maternal weight loss)		
Neurological								
14	Mouse (ddY)	once (GW)				1757 M (increased startle reflex; ataxia)	Kaise et al. 1989 DMA	
Developmental								
15	Rat (Sprague-Dawley)	GD6-15 (GW)			40 F (decreased fetal body weight)		Chernoff et al. 1990 DMA	
16	Rat (Sprague-Dawley)	Gd 6-15 (GW)		12	36 (decreases in number of live fetuses and fetal weight; increases in fetuses with diaphragmatic hernia; delayed ossification)		Irvine et al. 2006 DMA	
17	Rat (CD)	10 d Gd 7-16 1 x/d (GW)		15		30 (malformed palates in 15%)	Rogers et al. 1981 DMA	

Table 3-5 Levels of Significant Exposure to Dimethylarsinic Acid - Oral (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference Chemical Form	Comments
18	Mouse (CD-1)	Gd 8 (GW)				1600 F (fetal deaths, decreased fetal weight, delayed ossification, skeletal malformations)	Kavlock et al. 1985 DMA	
19	Mouse (CD-1)	10 d Gd 7-16 1 x/d (GW)		200		400 (18% decrease in fetal weight, delayed ossification, cleft palate in 12/28; irregular palatine rugae in 4.8%)	Rogers et al. 1981 DMA	
20	Rabbit (New Zealand)	Gd 7-19 (GW)		12			Irvine et al. 2006 DMA	
INTERMEDIATE EXPOSURE								
Death								
21	Rat (Fischer- 344)	13 wk (F)				190 M (100% mortality during first 4 weeks of study)	Crown et al. 1987 DMA	
22	Rat (Fischer- 344)	4 wk 5 d/wk 1 x/d (G)				57 (50% survival in males; 20% survival in females)	Murai et al. 1993 DMA	

Table 3-5 Levels of Significant Exposure to Dimethylarsinic Acid - Oral (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference Chemical Form	Comments
23	Rat (Fischer- 344)	8 wk (W)				17 M (10/10 died)	Wanibuchi et al. 1996 DMA	
Systemic								
24	Rat (Fischer- 344)	10 or 20 wk (F)	Renal	1 F	10 M (necrosis in bladder epithelium)		Arnold et al. 1999 DMA	
					5 F[b] (increased kidney weight, calcification at corticomedullary junction; increased bladder weight and increased BrdU labelling in bladder epithelium)			
			Bd Wt	10				
25	Rat (Fischer- 344)	10 wk (F)	Renal		11 F (increased bladder and kidney weights, hyperplasia and necrosis of bladder epithelium)		Cohen et al. 2001 DMA	

Table 3-5 Levels of Significant Exposure to Dimethylarsinic Acid - Oral (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference Chemical Form	Comments
26	Rat (Fischer- 344)	13 wk (F)	Resp	43.2 M			Crown et al. 1987 DMA	
			Cardio	43.2 M				
			Gastro	43.2 M	190 M (diarrhea and congestion and hemorrhagic contents in gastrointestinal tissues)			
			Hemato	0.44 F	4.5 F (decreased hemoglobin and erythrocyte levels)			
			Hepatic	23.5 M				
			Renal	0.4 M	4 M (increased urine volume and decreased specific gravity)			
			Endocr	0.4 M	4 M (hypertrophy of thyroid follicle epithelium)			
			Bd Wt	43.2 M				
27	Rat (Fischer- 344)	4 wk 5 d/wk 1 x/d (G)	Renal			57 (papillary necrosis and hyperplasia; cortical degeneration and necrosis)	Murai et al. 1993 DMA	
			Bd Wt		57 (decreased body weight)			

Table 3-5 Levels of Significant Exposure to Dimethylarsinic Acid - Oral (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference Chemical Form	Comments
28	Rat (Sprague-Dawley)	10 wk pre-mating, gestation and lactation periods (F)	Resp	16.5			Rubin et al. 1989 DMA	
			Cardio	16.5				
			Hemato	0.34 M	2.3 M (decreased mean corpscular hemoglobin concentration)			
			Hepatic	16.5				
			Renal	16.5				
			Endocr	2.3 F	16.5 F (hypertrophy of thyroid follicle epithelium)			
			Bd Wt	16.5				
29	Rat (Sprague-Dawley)	42 d (F)	Hemato	3.7 M			Siewicki 1981 DMA	
			Hepatic	3.7 M				
			Renal	3.7 M				
			Bd Wt	3.7 M				

Table 3-5 Levels of Significant Exposure to Dimethylarsinic Acid - Oral (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference Chemical Form	Comments
30	Dog (Beagle)	6 d/wk 52 wk (C)	Gastro	6.5	16 (vomiting and diarrhea)		Zomber et al. 1989 DMA	
			Hemato	16 M	40 M (decreased erythrocyte and increased leukocyte levels)			
Reproductive								
31	Rat (Sprague-Dawley)	10 wk pre-mating, gestation and lactation periods (F)		16.5			Rubin et al. 1989 DMA	
Cancer								
32	Mouse A/J	50 wk ad lib (W)				10.4 M (CEL: lung tumors)	Hayashi et al. 1998 DMA	

Table 3-5 Levels of Significant Exposure to Dimethylarsinic Acid - Oral (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference Chemical Form	Comments
CHRONIC EXPOSURE								
Systemic								
33	Rat (Fischer- 344)	daily 2 yr (F)	Resp	7.8			Arnold et al. 2006 DMA	
			Cardio	7.8				
			Gastro	7.8				
			Hemato	7.8				
			Musc/skel	7.8				
			Hepatic	7.8				
			Renal	0.77 M	3.1 M (nephrocalcinosis)			
				0.77 F	3.1 F (urothelial vacular degeneration and hyperplasia of urothelial cells in urinary bladder)			
			Endocr	3.1	7.8 (hypertrophy of thyroid follicle epithelium)			
			Dermal	7.8				
			Ocular	7.8				
			Bd Wt	7.8				
34	Rat (Fischer- 344)	104 wk (W)	Renal	0.75 M	3.4 M (nodular or papillary hyperplasia in urinary bladder)		Wei et al. 1999, 2002 DMA	

Table 3-5 Levels of Significant Exposure to Dimethylarsinic Acid - Oral (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference Chemical Form	Comments
35	Mouse (B6C3F1)	daily 2 yr (F)	Resp	94			Arnold et al. 2006 DMA	
			Cardio	94				
			Gastro	94				
			Hemato	94 F	94 F (decreased lymphocytes and increased monocytes)			
			Musc/skel	94				
			Hepatic	94				
			Renal	1.3[c] F	37 M (progressive glomerulonephropathy)			
					7.8[b] F (vacuolization of superficial cells of urotheliumin urinary bladder)			
			Dermal	94				
			Ocular	94				
			Bd Wt	94				

Table 3-5 Levels of Significant Exposure to Dimethylarsinic Acid - Oral (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference Chemical Form	Comments
36	Dog (Beagle)	6 d/wk 52 wk (C)	Resp	40			Zomber et al. 1989 DMA	
			Cardio	40				
			Gastro	6.5	16 (vomiting and diarrhea)			
			Hepatic	40				
			Renal	40				
Reproductive								
37	Rat (Fischer- 344)	daily 2 yr (F)		7.8			Arnold et al. 2006 DMA	Histological examination of reproductive tissues.
38	Mouse (B6C3F1)	daily 2 yr (F)		94			Arnold et al. 2006 DMA	Histological examination of reproductive tissues.
Cancer								
39	Rat (Fischer- 344)	daily 2 yr (F)				7.8 (CEL: urothelial cell papillomas and carcinomas in urinary bladder)	Arnold et al. 2006 DMA	
40	Rat (Fischer- 344)	104 wk (W)				3.4 M (CEL: urinary bladder tumors)	Wei et al. 1999, 2002 DMA	

Table 3-5 Levels of Significant Exposure to Dimethylarsinic Acid - Oral (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference Chemical Form	Comments
41	Mouse knockout	continuous 18 mo (W)				11.8 M (CEL)	Salim et al. 2003 DMA	

a The number corresponds to entries in Figure 3-5.

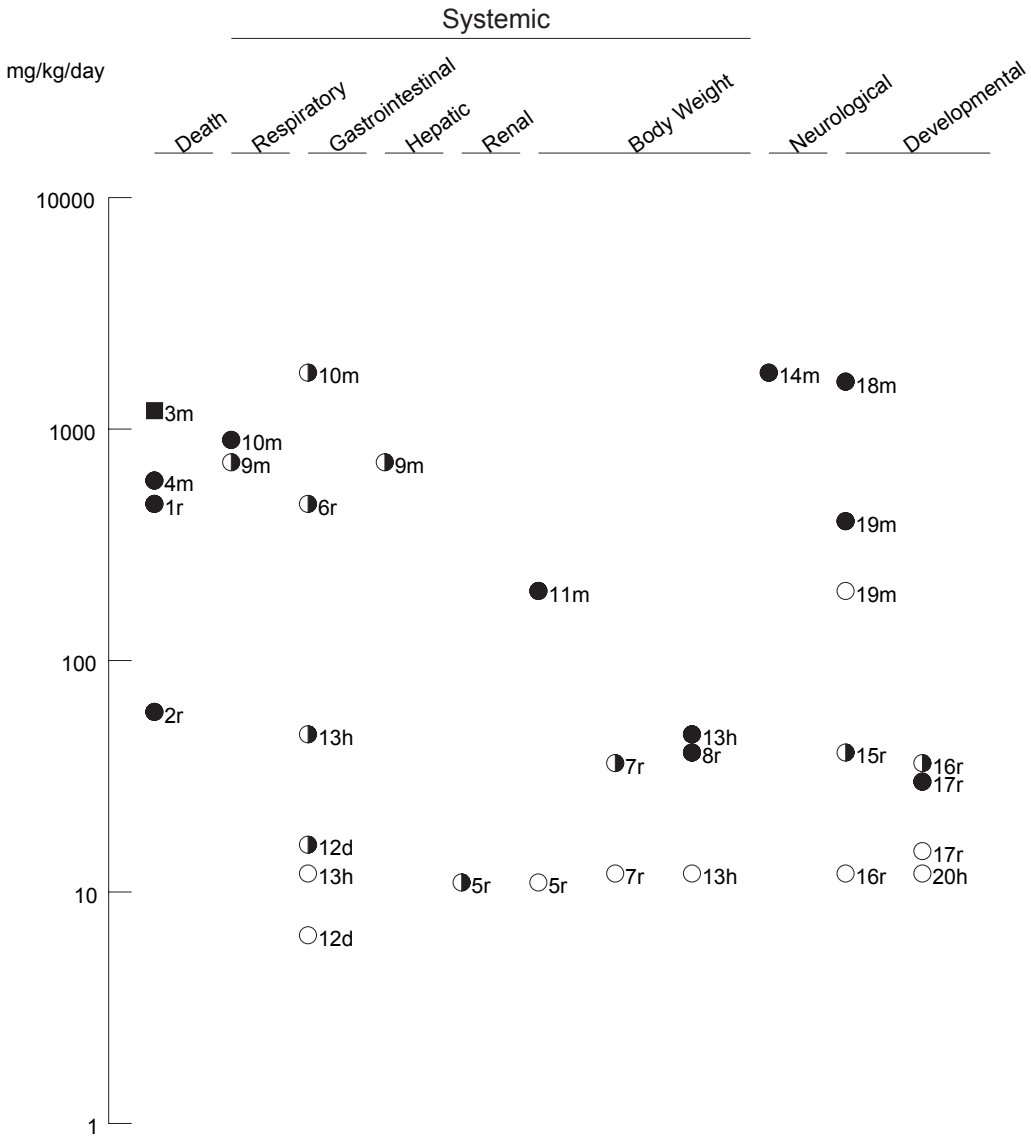
b Differences in levels of health effects and cancer effects between male and females are not indicated in Figure 3-5. Where such differences exist, only the levels of effect for the most sensitive gender are presented.

c The chronic-duration oral MRL of 0.02 mg DMA/kg/day was calculated using a benchmark dose analysis. The BMDL10 of 1.80 mg DMA/kg/day was divided by an uncertainty factor of 100 (10 to account for extrapolation from animals to humans and 10 for human variability)

ad lib = ad libitum; ALT = alanine aminotransferase; Bd Wt = body weight; BrdU = bromodeoxyuridine; (C) = capsule; Cardio = cardiovascular; CEL = cancer effect level; CYP = cytochrome p; d= day(s); DMA = dimethylarsinic acid; Endocr = endocrine; (F) = feed; F = female; (G) = gavage; Gastro = gastrointestinal; Gd = gestation day; GSH = reduced glutathione; GSSG = oxidized glutathione; (GW) = gavage in water; Hemato = hematological; hr = hour(s); LD50 = lethal dose, 50% kill; ODC = ornithine decarboxylase; LOAEL = lowest-observable-adverse-effect level; M = male; mo = month; Musc/skel = musculoskeletal; NOAEL = no-observable-adverse-effect level; Resp = respiratory; (W) = drinking water; wk = week(s); x = time(s); yr = year(s)

Figure 3-5 Levels of Significant Exposure to Dimethylarsinic Acid - Oral

Acute (≤14 days)



c-Cat, d-Dog, r-Rat, p-Pig, q-Cow
-Humans, k-Monkey, m-Mouse, h-Rabbit, a-Sheep
f-Ferret, j-Pigeon, e-Gerbil, s-Hamster, g-Guinea Pig
n-Mink, o-Other

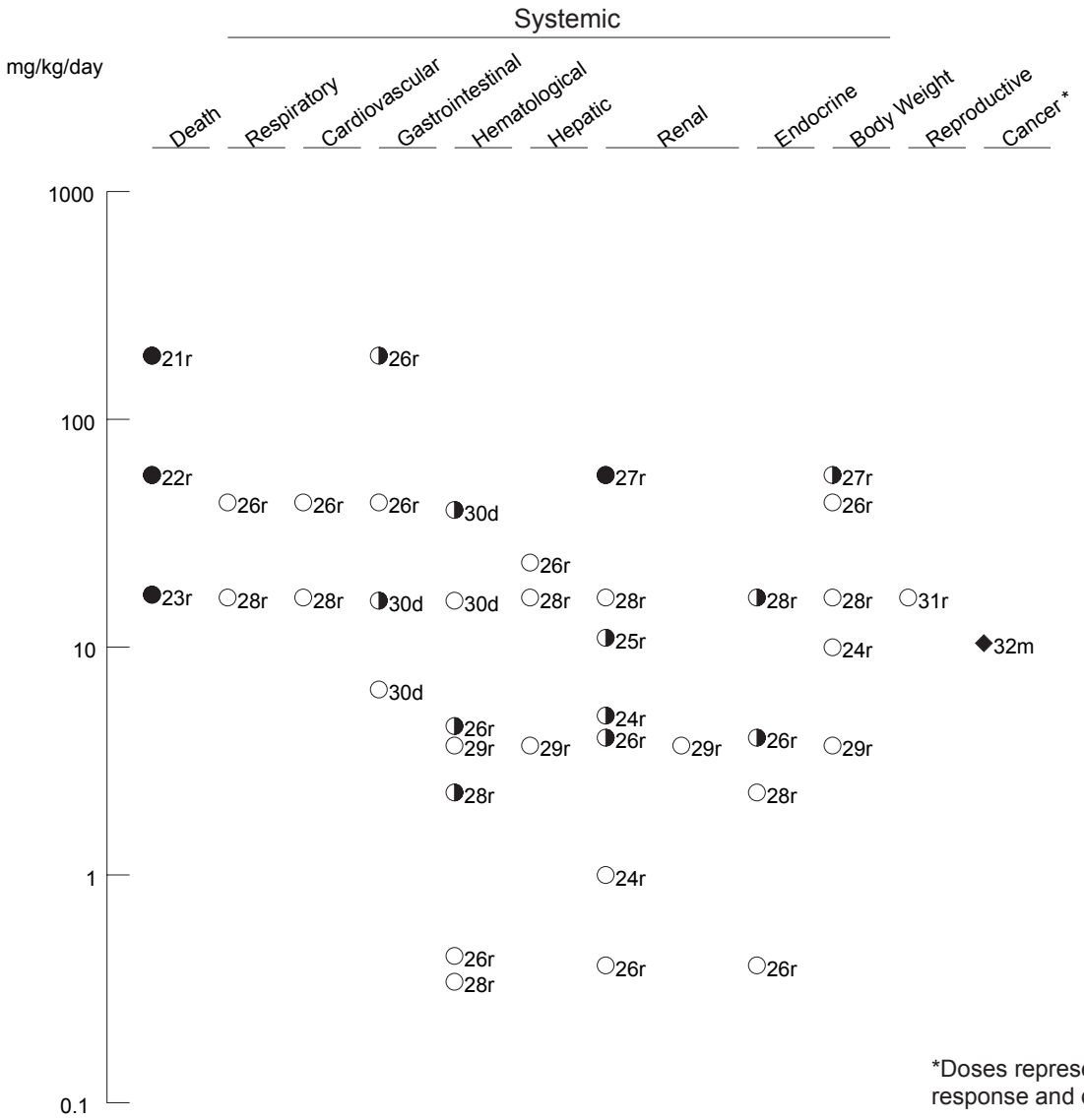
◆ Cancer Effect Level-Animals
● LOAEL, More Serious-Animals
◑ LOAEL, Less Serious-Animals
○ NOAEL - Animals

▼ Cancer Effect Level-Humans
▲ LOAEL, More Serious-Humans
◭ LOAEL, Less Serious-Humans
△ NOAEL - Humans

■ LD50/LC50
Minimal Risk Level for effects other than Cancer

Figure 3-5 Levels of Significant Exposure to Dimethylarsinic Acid - Oral *(Continued)*

Intermediate (15-364 days)

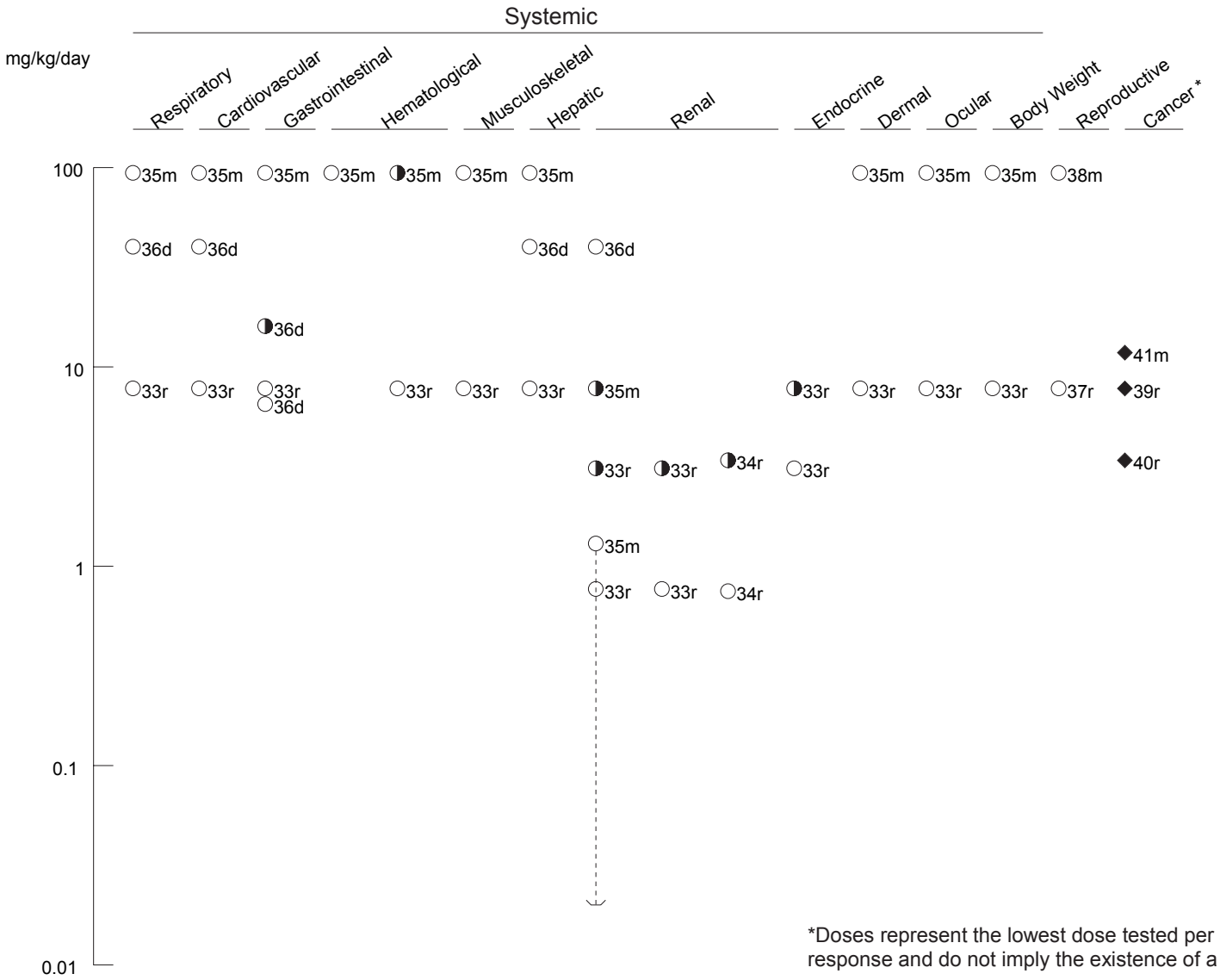


*Doses represent the lowest dose tested per study that produced a tumorigenic response and do not imply the existence of a threshold for the cancer endpoint.

c-Cat	-Humans	f-Ferret	n-Mink	◆ Cancer Effect Level-Animals	▼ Cancer Effect Level-Humans	■ LD50/LC50
d-Dog	k-Monkey	j-Pigeon	o-Other	● LOAEL, More Serious-Animals	▲ LOAEL, More Serious-Humans	Minimal Risk Level for effects other than Cancer
r-Rat	m-Mouse	e-Gerbil		◑ LOAEL, Less Serious-Animals	◭ LOAEL, Less Serious-Humans	
p-Pig	h-Rabbit	s-Hamster		○ NOAEL - Animals	△ NOAEL - Humans	
q-Cow	a-Sheep	g-Guinea Pig				

Figure 3-5 Levels of Significant Exposure to Dimethylarsinic Acid - Oral *(Continued)*

Chronic (≥365 days)



*Doses represent the lowest dose tested per study that produced a tumorigenic response and do not imply the existence of a threshold for the cancer endpoint.

c-Cat	-Humans	f-Ferret	n-Mink
d-Dog	k-Monkey	j-Pigeon	o-Other
r-Rat	m-Mouse	e-Gerbil	
p-Pig	h-Rabbit	s-Hamster	
q-Cow	a-Sheep	g-Guinea Pig	

◆ Cancer Effect Level-Animals
● LOAEL, More Serious-Animals
◑ LOAEL, Less Serious-Animals
○ NOAEL - Animals

▼ Cancer Effect Level-Humans
▲ LOAEL, More Serious-Humans
◭ LOAEL, Less Serious-Humans
△ NOAEL - Humans

■ LD50/LC50
Minimal Risk Level for effects other than Cancer

Table 3-6 Levels of Significant Exposure to Roxarsone - Oral

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference Chemical Form	Comments
ACUTE EXPOSURE								
Death								
1	Rat (Holtzman)	once (GW)				155 (LD50)	Kerr et al. 1963 ROX	
2	Rat (Fischer- 344)	once (GO)				150 M (5/5 died) 81[b] F (LD50)	NTP 1989b ROX	
3	Rat (Fischer- 344)	14 d (F)				128[b] M (3/5 died) 144 F (5/5 died)	NTP 1989b ROX	
4	Mouse (B6C3F1)	once (GO)				300 M (5/5 died) 244[b] F (LD50)	NTP 1989b ROX	
5	Mouse (B6C3F1)	14 d (F)				168 F (5/5 died)	NTP 1989b ROX	
Systemic								
6	Rat (Fischer- 344)	14 d (F)	Bd Wt	16 M	32 M (22% reduced body weight)		NTP 1989b ROX	
7	Mouse (B6C3F1)	14 d (F)	Bd Wt	84		168 (34% decrease in body weight)	NTP 1989b ROX	
Neurological								
8	Rat (Fischer- 344)	14 d (F)		16 M	32 M (slight inactivity)		NTP 1989b ROX	

Table 3-6 Levels of Significant Exposure to Roxarsone - Oral (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference Chemical Form	Comments
9	Mouse (B6C3F1)	14 d (F)		20	42 (slight inactivity; ruffled fur)		NTP 1989b ROX	
10	Pig (Landrace)	30 d ad lib (F)				6.3 (muscle tremors and clonic convulsions)	Rice et al. 1985; Kennedy et al. 1986 ROX	
INTERMEDIATE EXPOSURE								
Death								
11	Rat (Holtzman)	13 wk (F)				20 (10/12 died)	Kerr et al. 1963 ROX	
12	Rat (Fischer- 344)	13 wk ad lib (F)				64 M (3/10 died)	NTP 1989b ROX	
13	Mouse (B6C3F1)	13 wk ad lib (F)				136 (6/10 males and 8/10 females died)	NTP 1989b ROX	

Table 3-6 Levels of Significant Exposure to Roxarsone - Oral (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference Chemical Form	Comments
Systemic								
14	Rat (Fischer- 344)	31 or 90 d ad lib (F)	Hemato	32 M			NTP 1989b ROX	
			Hepatic	9 F	36 F (decreased absolute and relative liver weight)			
			Renal	8 M	32 M (increased kidney weight; minimal tubular degeneration)			
			Bd Wt	8 M		32 M (27% decrease in body weight)		

Table 3-6 Levels of Significant Exposure to Roxarsone - Oral (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference Chemical Form	Comments
15	Rat (Fischer- 344)	13 wk ad lib (F)	Resp	64 M			NTP 1989b ROX	
			Cardio	64 M				
			Gastro	64 M				
			Musc/skel	64 M				
			Hepatic	4 M	8 M (increased relative liver weight)			
			Renal	16 M	32 M (interstitial inflammation, focal regenerative hyperplasia of tubular cell epithelium and mineralization)			
			Endocr	64 M				
			Dermal	64 M				
			Bd Wt	8 M	16 M (14% decreased body weight)	32 M (26% decreased body weight)		

Table 3-6 Levels of Significant Exposure to Roxarsone - Oral (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference Chemical Form	Comments
16	Mouse (B6C3F1)	13 wk ad lib (F)	Cardio	136			NTP 1989b ROX	
			Gastro	136				
			Musc/skel	136				
			Hepatic	136				
			Renal	136				
			Endocr	136				
			Dermal	136				
			Bd Wt		136 (18% decreased body weight in males; 11% decreased body weight in females)			
17	Mouse (B6C3F1)	29 or 91 d ad lib (F)	Hemato	68			NTP 1989b ROX	
			Hepatic	68				
			Renal	68				
Neurological								
18	Rat (Fischer- 344)	13 wk ad lib (F)		32 M		64 M (trembling, ataxia, hyperexcitability, slight inactivity, ruffled fur)	NTP 1989b ROX	

Table 3-6 Levels of Significant Exposure to Roxarsone - Oral (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)		Reference Chemical Form	Comments
19	Pig	28 d (F)				10	(muscle tremors)	Edmonds and Baker 1986 ROX	
20	Pig (Landrace)	30 d ad lib (F)				6.3	(paraplegia, myelin degeneration in spinal cord, peripheral nerves, optic nerve)	Rice et al. 1985; Kennedy et al. 1986 ROX	
CHRONIC EXPOSURE									
Systemic									
21	Rat (Fischer- 344)	103 wk ad lib (F)	Resp	4				NTP 1989b ROX	
			Cardio	4					
			Gastro	4					
			Musc/skel	4					
			Hepatic	4					
			Renal	4					
			Endocr	4					
			Dermal	4					
			Ocular	4					
			Bd Wt	4					
22	Mouse (Fischer- 344)	103 wk ad lib (F)	Resp	43 M				NTP 1989b ROX	
			Cardio	43 M					
			Gastro	43 M					
			Musc/skel	43 M					
			Hepatic	43 M					

Table 3-6 Levels of Significant Exposure to Roxarsone - Oral (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference Chemical Form	Comments
			Renal	43 M				
			Endocr	43 M				
			Dermal	43 M				
			Ocular	43 M				
			Bd Wt	43 F				

a The number corresponds to entries in Figure 3-6.

b Differences in levels of health effects and cancer effects between male and females are not indicated in Figure 3-6. Where such differences exist, only the levels of effect for the most sensitive gender are presented.

ad lib = ad libitum; Bd Wt = body weight; Cardio = cardiovascular; d = day(s); Endocr = endocrine; (F) = feed; F = Female; Gastro = gastrointestinal; (GO) = gavage in oil; (GW) = gavage in water; Hemato = hematological; LC50 = lethal concentration, 50% kill, LD50 = lethal dose, 50% kill; LOAEL = lowest-observed-adverse-effect level; M = male; Musc/skel = musculoskeletal; NOAEL = no-observed-adverse-effect level; occup = occupational; Resp = respiratory; wk = week(s)

Figure 3-6 Levels of Significant Exposure to Roxarsone - Oral

Acute (≤14 days)

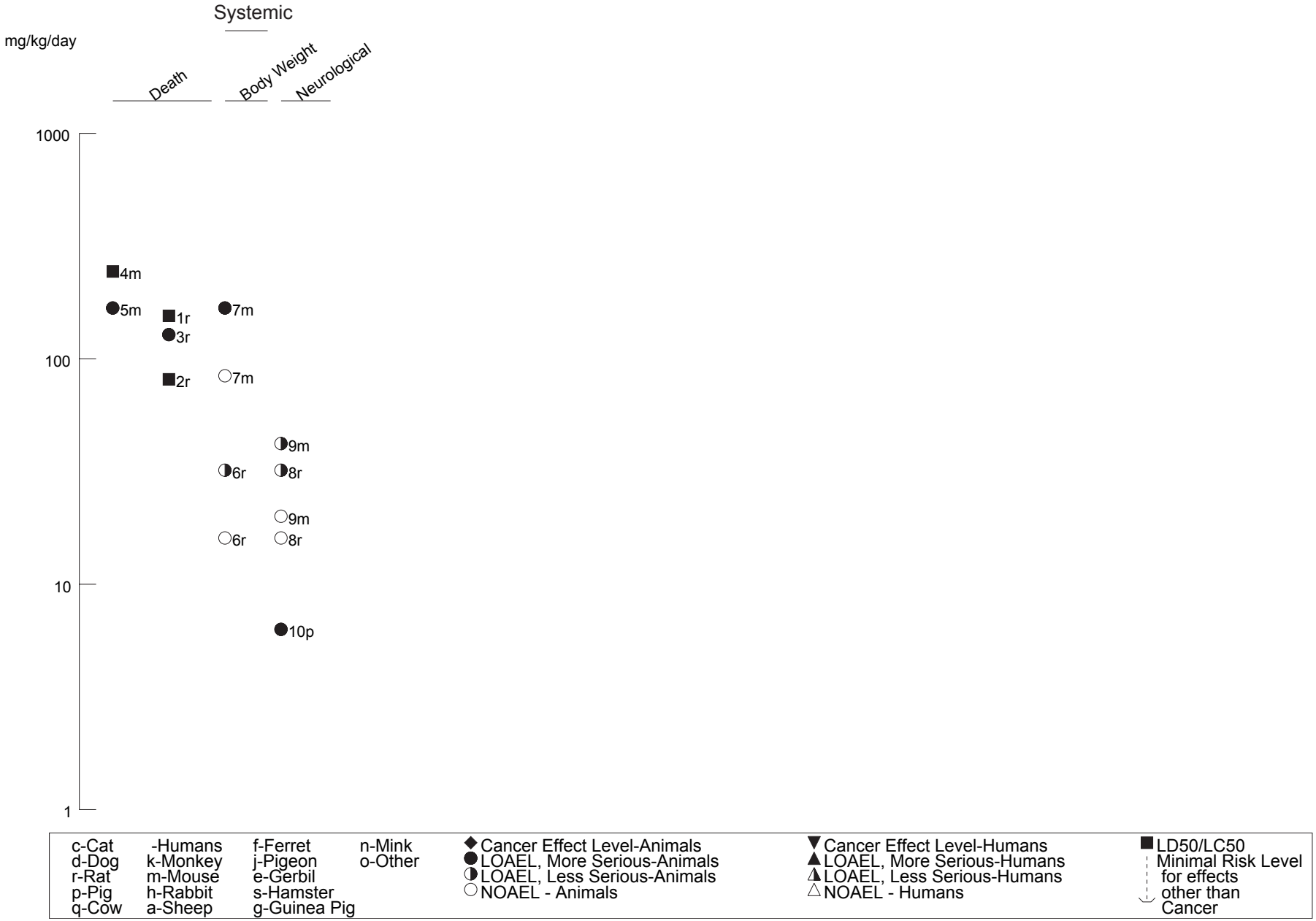
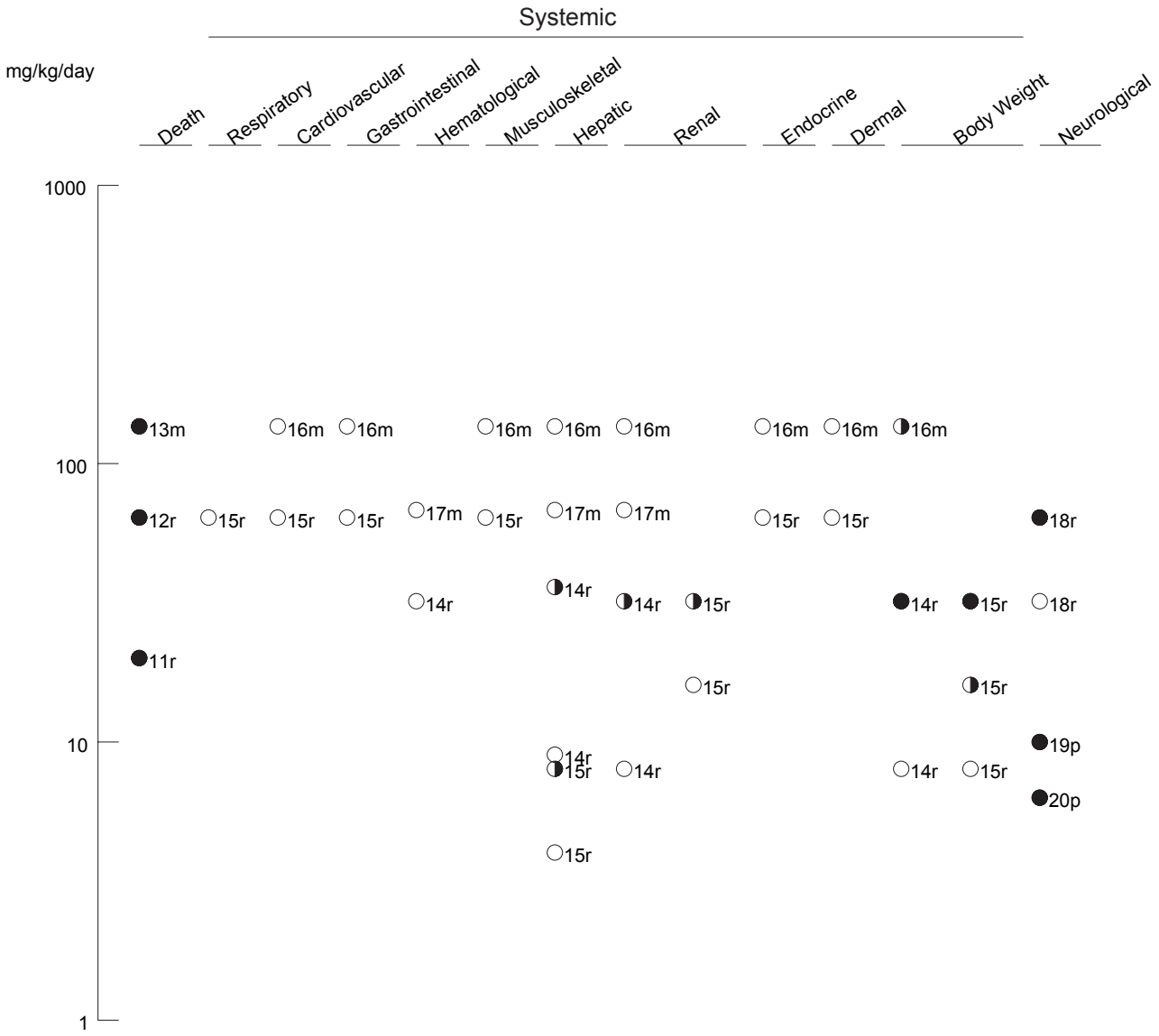


Figure 3-6 Levels of Significant Exposure to Roxarsone - Oral *(Continued)*

Intermediate (15-364 days)



c-Cat d-Dog r-Rat p-Pig q-Cow
-Humans k-Monkey m-Mouse h-Rabbit a-Sheep
f-Ferret j-Pigeon e-Gerbil s-Hamster g-Guinea Pig
n-Mink o-Other

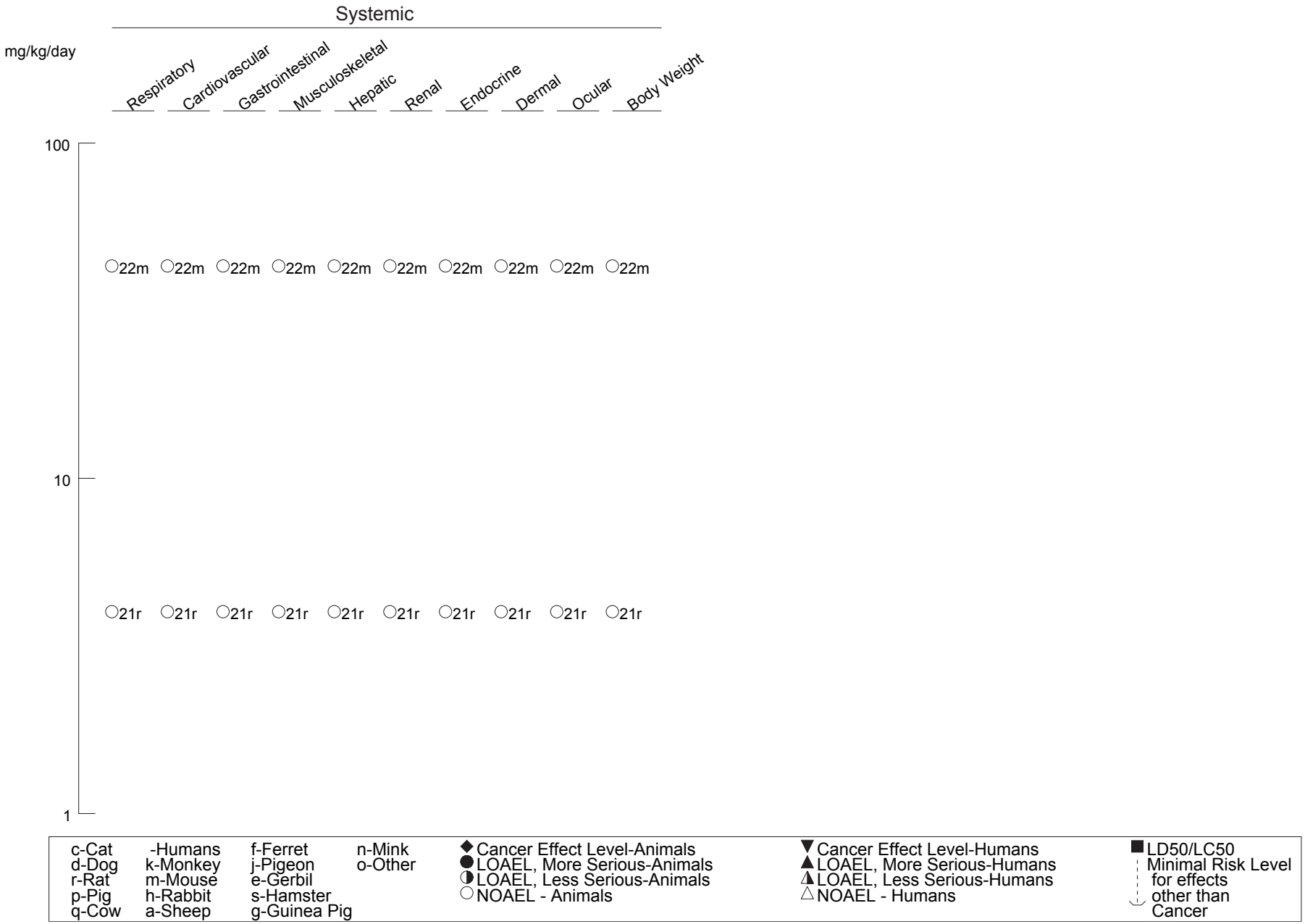
◆ Cancer Effect Level-Animals
● LOAEL, More Serious-Animals
◑ LOAEL, Less Serious-Animals
○ NOAEL - Animals

▼ Cancer Effect Level-Humans
▲ LOAEL, More Serious-Humans
◭ LOAEL, Less Serious-Humans
△ NOAEL - Humans

■ LD50/LC50
Minimal Risk Level for effects other than Cancer

Figure 3-6 Levels of Significant Exposure to Roxarsone - Oral *(Continued)*

Chronic (≥365 days)



Available LD_{50} values for arsenate and arsenite in rats and mice range from 15 to 175 mg As/kg (Dieke and Richter 1946; Gaines 1960; Harrisson et al. 1958; Kaise et al. 1985). The variability can be attributed to differences based on species, strain, specific route of exposure (feed vs. gavage), specific compound tested, and testing laboratory. Most deaths occurred within 1 day of exposure, but details regarding cause of death were not generally reported. Seven of 25 pregnant rats given a single gavage dose of 23 mg As/kg as arsenic trioxide on day 9 of gestation died soon after dosing, while no deaths occurred at doses of 4–15 mg As/kg (Stump et al. 1999). Data on lethality from repeated exposure studies in animals are relatively sparse. Seven of 20 pregnant rabbits died from repeated gavage doses of 1.5 mg As/kg/day as arsenic acid during gestation, while none died at 0.1–0.4 mg As/kg/day (Nemec et al. 1998). Chronic studies observed treatment-related mortality in monkeys exposed to 3 mg As/kg/day as arsenate (Heywood and Sortwell 1979), dogs exposed to 2.4 mg As/kg/day as arsenite or arsenate (Byron et al. 1967), mice exposed to 1 mg As/kg/day as arsenite (Schroeder and Balassa 1967), and rats exposed to 30 mg As/kg/day as lead arsenate (Kroes et al. 1974).

Reliable LOAEL and LD_{50} values for lethality from oral exposure to inorganic arsenicals in each species and duration category are recorded in Table 3-3 and plotted in Figure 3-3.

Organic Arsenicals. No studies were located regarding death in humans after oral exposure to organic arsenicals, but the acute lethality of MMA, DMA, and roxarsone have been investigated in several animal studies. The LD_{50} values for MMA (including MSMA), DMA, and roxarsone are 102–3,184 mg/kg MMA or MSMA (Gur and Nyska 1990; Jaghabir et al. 1988; Kaise et al. 1989), 1,200 mg DMA/kg/day (Kaise et al. 1989), and 14.2–69.5 mg DMA/kg/day (Kerr et al. 1963; NTP 1989b), respectively. The cause of death was not investigated in any of these studies. Intermediate-duration exposure to MMA, DMA, or roxarsone resulted in increased mortality in laboratory animals exposed to 106.9 mg MMA/kg/day (Arnold et al. 2003), 17–190 mg DMA/kg/day (Crown et al. 1987; Murai et al. 1993; Wanibuchi et al. 1996) or 20–64 mg/kg/day roxarsone (Edmonds and Baker 1986; Kerr et al. 1963; NTP 1989b), respectively. Increased mortality was also observed in rats chronically exposed to 72.4 mg MMA/kg/day (Arnold et al. 2003).

3.2.2.2 Systemic Effects

The highest NOAEL values and all reliable LOAEL values for systemic effects from oral exposure in each species and duration category are recorded in Table 3-3 and plotted in Figure 3-3. Similar data for

oral exposure to MMA, DMA, and roxarsone are shown in Tables 3-4, 3-5, and 3-6, and shown in Figures 3-4, 3-5, and 3-6, respectively.

Respiratory Effects.

Inorganic Arsenicals. Serious respiratory effects, including respiratory distress, hemorrhagic bronchitis, and pulmonary edema, have been reported in some cases of acute oral arsenic poisoning at doses of 8 mg As/kg and above (e.g., Civantos et al. 1995; Fincher and Koerker 1987; Levin-Scherz et al. 1987; Moore et al. 1994b; Quatrehomme et al. 1992). These effects may be secondary to injury to the pulmonary vasculature (see Cardiovascular Effects, below). In addition, bronchitis and sequelae (bronchiectasis, bronchopneumonia) have been observed in patients and at autopsy in some chronic poisoning cases (Guha Mazumder et al. 2005; Milton and Rahman 2002; Rosenberg 1974; Tsai et al. 1999; Zaldívar 1974; Zaldívar and Guillier 1977). Bronchopneumonia secondary to arsenic-induced bronchitis was considered to be the cause of death in one young child who died after several years of exposure to an average dose of 0.08 mg As/kg/day (Zaldívar and Guillier 1977). Decrements in lung function, measured as decreased FEV_1, FVC, and $FEF_{25–75}$ have also been reported in subjects exposed to 0.1–0.5 mg As/L in the drinking water and exhibiting skin lesions (von Ehrenstein et al. 2005). In general, however, respiratory effects have not been widely associated with repeated oral ingestion of low arsenic doses. Nevertheless, a few studies have reported minor respiratory symptoms, such as cough, sputum, rhinorrhea, and sore throat, in people with repeated oral exposure to 0.03–0.05 mg As/kg/day (Ahmad et al. 1997; Mizuta et al. 1956).

There are few data regarding respiratory effects in animals following acute oral exposure to inorganic arsenic. An infant Rhesus monkey that died after 7 days of oral exposure to a complex arsenate salt at a dose of 3 mg As/kg/day exhibited bronchopneumonia with extensive pulmonary hemorrhage, edema, and necrosis (Heywood and Sortwell 1979). Two other monkeys in this treatment group survived a 1-year exposure period and had no gross or microscopic pulmonary lesions at sacrifice. Increased relative lung weights were seen in rats exposed to 6.66 mg As/kg/day as sodium arsenite 5 days/week for 12 weeks (Schulz et al. 2002). Chronic oral studies in dogs and rats treated with arsenate or arsenite failed to find respiratory lesions (Byron et al. 1967; Kroes et al. 1974; Schroeder et al. 1968).

One study utilizing gallium arsenide included limited investigation of respiratory function. Respiration rate was significantly decreased in rats following ingestion of a single dose of gallium arsenide at 1,040 mg As/kg, but was unaffected at a dose of 520 mg As/kg (Flora et al. 1997a). Respiration rate was measured 1, 7, and 15 days after dosing, but the decrease was most noticeable after 15 days.

Organic Arsenicals. No respiratory effects were noted after acute human ingestion of 1,714 mg MSMA/kg (Shum et al. 1995). Mice exhibited respiratory arrest after a single oral dose of 1,800 mg MMA/kg (Kaise et al. 1989) or 900 mg DMA/kg (Kaise et al. 1989) and lung ornithine decarboxylase activity was reduced after ingestion of one or two doses of 720 mg DMA/kg (Ahmad et al. 1999a). Localized lung hemorrhage was observed in dogs after a single oral dose of 14.2 mg/kg roxarsone in a capsule (Kerr et al. 1963). No respiratory effects were seen after intermediate or chronic exposure of rats, mice, or dogs exposed to 35 mg MMA/kg/day (Waner and Nyska 1988), 7.8–94 mg DMA/kg/day (Arnold et al. 2006; Crown et al. 1987; Rubin et al. 1989; Zomber et al. 1989), or 4–136 mg/kg/day roxarsone (NTP 1989b).

Cardiovascular Effects.

Inorganic Arsenicals. A number of studies in humans indicate that arsenic ingestion may lead to serious effects on the cardiovascular system. Characteristic effects on the heart from both acute and long-term exposure include altered myocardial depolarization (prolonged QT interval, nonspecific ST segment changes) and cardiac arrhythmias (Cullen et al. 1995; Glazener et al. 1968; Goldsmith and From 1986; Heyman et al. 1956; Little et al. 1990; Mizuta et al. 1956; Moore et al. 1994b; Mumford et al. 2007). A significant dose-related increase in the prevalence of cardiac electrophysiologic abnormalites was observed in residents of Inner Mongolia, China; the incidences of QT prolongation were observed in 3.9, 11.1, and 20.6% of the residents with drinking water levels of <21, 110–300, and 430–690 μg/L, respectively (Mumford et al. 2007). Hypertrophy of the ventricular wall was observed at autopsy after acute exposure to 93 mg of arsenic (Quatrehomme et al. 1992). Long-term, low-level exposures may also lead to damage to the vascular system. The most dramatic example of this is "Blackfoot Disease," a condition that is endemic in an area of Taiwan where average drinking water levels of arsenic range from 0.17 to 0.80 ppm (Tseng 1977), corresponding to doses of about 0.014–0.065 mg As/kg/day (IRIS 2007). The disease is characterized by a progressive loss of circulation in the hands and feet, leading ultimately to necrosis and gangrene (Chen et al. 1988b; Ch'i and Blackwell 1968; Tseng 1977, 1989; Tseng et al. 1968, 1995, 1996). Several researchers have presented evidence that other factors besides arsenic (e.g., other water contaminants, dietary deficits) may play a role in the etiology of this disease (Ko 1986; Lu et al. 1990; Yu et al. 1984). While this may be true, the clear association between the occurrence of Blackfoot Disease and the intake of elevated arsenic levels indicates that arsenic is at least a contributing factor. The results of a recent study suggested that individuals with a lower capacity to methylate

inorganic arsenic to DMA have a higher risk of developing peripheral vascular disease in the Blackfoot Disease-hyperendemic area in Taiwan (Tseng et al. 2005).

Arsenic exposure in Taiwan has also been associated with an increased incidence of cerebrovascular and microvascular diseases (Chiou et al. 1997; Wang et al. 2002, 2003) and ischemic heart disease (Chang et al. 2004; Chen et al. 1996; Hsueh et al. 1998b; Tsai et al. 1999; Tseng et al. 2003). Moreover, effects of arsenic on the vascular system have also been reported in a number of other populations. For example, hypertension, defined as a systolic blood pressure of ≥140 mm Hg in combination with a diastolic blood pressure of ≥90 mm Hg, was associated with estimated lifetime doses of approximately 0.055 mg As/kg/day (0.25 mg/L in water) in a study of people in Bangladesh (Rahman et al. 1999); no significant association was found with estimated doses of 0.018 mg As/kg/day (0.75 mg/L in water). Wang et al. (2003) found an increased incidence of microvascular and macrovascular disease among subjects in Taiwan living in an arseniasis-endemic area in which the water of artesian wells had arsenic concentrations >0.35 mg/L (estimated doses of >0.03 mg As/kg/day). An additional study of Taiwanese subjects reported a significant increase in incidence of hypertension associated with concentrations of arsenic in the water >0.7 mg/L (estimated doses of >0.06 mg As/kg/day) (Chen et al. 1995). Studies in Chile indicate that ingestion of 0.6–0.8 ppm arsenic in drinking water (corresponding to doses of 0.02–0.06 mg As/kg/day, depending on age) increases the incidence of Raynaud's disease and of cyanosis of fingers and toes (Borgoño and Greiber 1972; Zaldívar 1974, 1977; Zaldívar and Guillier 1977). Autopsy of five children from this region who died of apparent arsenic toxicity showed a marked thickening of small and medium sized arteries in tissues throughout the body, especially the heart (Rosenberg 1974). In addition, cardiac failure, arterial hypotension, myocardial necrosis, and thrombosis have been observed in children who died from chronic arsenic ingestion (Zaldívar 1974), as well as adults chronically exposed to arsenic (Dueñas et al. 1998). Likewise, thickening and vascular occlusion of blood vessels were noted in German vintners exposed to arsenical pesticides in wine and in adults who drank arsenic-contaminated drinking water (Roth 1957; Zaldívar and Guillier 1977). A survey of Wisconsin residents using private wells for their drinking water found that residents exposed for at least 20 years to water concentrations of >10 µg As/L had increased incidences of cardiac bypass surgery, high blood pressure, and circulatory problems as compared with residents exposed to lower arsenic concentrations (Zierold et al. 2004). Similarly, Lewis et al. (1999) reported increased mortality from hypertensive heart disease in both men and women among a cohort exposed to arsenic in their drinking water in Utah, as compared with the general population of Utah. Limitations in the study included lack of evaluation of smoking as a confounder and of other dietary sources of arsenic, and the lack of a dose-response for hypertensive heart disease. Another ecological study (Engel and Smith 1994) found significant increases in deaths from

arteriosclerosis, aortic aneurysm, and all other diseases of the arteries, arterioles, and capillaries among U.S. residents with arsenic drinking waters of >20 µg/L; the increase in deaths from congenital anomalies of the heart and other anomalies of the circulatory system also observed in this subpopulation limits the interpretation of the findings.

Similar alterations in vascular reactivity have been noted in rats given repeated oral doses of arsenic trioxide (11 mg As/kg/day) for several weeks (Bekemeier and Hirschelmann 1989), although no histological effects could be detected in the hearts of rats or dogs exposed to up to 30 mg As/kg/day as arsenate or arsenite for 2 years (Byron et al. 1967; Kroes et al. 1974; Schroeder et al. 1968). Acute exposure of rats to gallium arsenide at a dose of 1,040 mg As/kg resulted in an increase in blood pressure and heart rate, while 520 mg As/kg had no effect (Flora et al. 1997a). Guinea pigs exposed to arsenic trioxide for 1 day (0, 7.6, 22.7, or 37.9 mg As/kg) or 8 days (0 or 3.8 mg As/kg/day) showed prolongation of the cardiac QT interval and action potential duration (Chiang et al. 2002).

Organic Arsenicals. No adverse cardiovascular effects were noted after acute human ingestion of 1,714 mg MSMA/kg (Shum et al. 1995). However, sinus tachycardia was noted after acute ingestion of 73 mg DMA/kg (as dimethyl arsenic acid and dimethyl arsenate) (Lee et al. 1995). No cardiovascular effects were seen after intermediate or chronic exposure of laboratory animals to 35–67.1 mg MMA/kg/day (Arnold et al. 2003; Waner and Nyska 1988), 7.8–94 mg DMA/kg/day (Arnold et al. 2006; Crown et al. 1987; Rubin et al. 1989; Zomber et al. 1989), or 4–136 mg/kg/day roxarsone (NTP 1989b).

Gastrointestinal Effects.

Inorganic Arsenicals. Clinical signs of gastrointestinal irritation, including nausea, vomiting, diarrhea, and abdominal pain, are observed in essentially all cases of short-term high-dose exposures to inorganic arsenic (e.g., Armstrong et al. 1984; Bartolome et al. 1999; Campbell and Alvarez 1989; Chakraborti et al. 2003a; Cullen et al. 1995; Fincher and Koerker 1987; Goebel et al. 1990; Kingston et al. 1993; Levin-Scherz et al. 1987; Lugo et al. 1969; Moore et al. 1994b; Muzi et al. 2001; Uede and Furukawa 2003; Vantroyen et al. 2004). Similar signs are also frequently observed in groups or individuals with longer-term, lower-dose exposures (e.g., Borgoño and Greiber 1972; Cebrián et al. 1983; Franzblau and Lilis 1989; Guha Mazumder et al. 1988, 1998a; Haupert et al. 1996; Holland 1904; Huang et al. 1985; Mizuta et al. 1956; Nagai et al. 1956; Silver and Wainman 1952; Wagner et al. 1979; Zaldívar 1974), but effects are usually not detectable at exposure levels below about 0.01 mg As/kg/day (Harrington et al. 1978; Valentine et al. 1985). These symptoms generally decline within a short time after exposure ceases.

Gastrointestinal irritation symptoms form the basis (in part) for the acute oral MRL of 0.005 mg/kg/day for inorganic arsenic, as described in footnote b in Table 3-3. More severe symptoms (hematemesis, hemoperitoneum, gastrointestinal hemorrhage, and necrosis) have been reported in some cases with acute exposure to 8 mg As/kg or more (Civantos et al. 1995; Fincher and Koerker 1987; Levin-Scherz et al. 1987; Quatrehomme et al. 1992), and also in some people with long-term ingestion of 0.03–0.05 mg As/kg/day as a medicinal preparation (Lander et al. 1975; Morris et al. 1974).

Clinical signs of gastrointestinal irritation were observed in monkeys and rats given repeated oral doses of arsenic (6 and 11 mg As/kg/day, respectively) for 2 weeks (Bekemeier and Hirschelmann 1989; Heywood and Sortwell 1979). Hemorrhagic gastrointestinal lesions have also been reported in animal studies. A monkey that died after repeated oral treatment with 6 mg As/kg/day for approximately 1 month was found to have acute inflammation and hemorrhage of the small intestine upon necropsy (Heywood and Sortwell 1979). This lesion was not found in other monkeys that died in this study, or in the survivors. Two pregnant mice that died after repeated gavage treatment with 24 mg As/kg/day as arsenic acid had hemorrhagic lesions in the stomach (Nemec et al. 1998). Gross gastrointestinal lesions (stomach adhesions, eroded luminal epithelium in the stomach) were seen frequently in rats treated by gavage with 8 mg As/kg/day as arsenic trioxide starting before mating and continuing through the end of gestation (Holson et al. 2000). The lesions were not found in rats treated with 4 mg As/kg/day in this study. No histological evidence of gastrointestinal injury was detected in rats exposed to arsenate or arsenite in the feed for 2 years at doses up to 30 mg As/kg/day, but dogs fed a diet containing 2.4 mg As/kg/day as arsenite for 2 years had some bleeding in the gut (Byron et al. 1967; Kroes et al. 1974).

Organic Arsenicals. Vomiting was noted after ingestion of 793 mg/kg arsenic (as monosodium methanearsenate) in a suicide attempt (Shum et al. 1995). Ingestion of 78 mg DMA/kg (as dimethyl arsenic acid and dimethyl arsenate) induced vomiting, abdominal pain, hyperactive bowel, and diarrhea (Lee et al. 1995).

The gastrointestinal tract appears to be the critical target of toxicity following oral exposure to MMA. Diarrhea/loose feces has been reported in mice and rabbits following a single gavage dose of 2,200 mg MMA/kg or 60 mg MSMA/kg, respectively (Jaghabir et al. 1988; Kaise et al. 1989), pregnant rabbits administered 12 mg MMA/kg/day via gavage (Irvine et al. 2006), rats exposed to 30.2 mg MMA/kg/day in the diet during the first year of a 2-year study (Arnold et al. 2003), dogs administered 2 mg MMA/kg/day via capsule for 52 weeks (Waner and Nyska 1988), rats fed diets containing 25.7 mg MMA/kg/day for 2 years (Arnold et al. 2003), and mice exposed to 67.1 mg MMA/kg/day in the diet for

2 years (Arnold et al. 2003). However, the increased incidence of diarrhea is not always accompanied by macroscopic or histological alterations in the gastrointestinal tissues. For example, in the 2-year rat study (Arnold et al. 2003; incidence data reported in Crown et al. 1990), an increased incidence of diarrhea was observed at 25.7 mg MMA/kg/day; macroscopic or histological alterations were observed in some animals, but the incidence was similar to controls. At the next highest dose level (72.4 mg MMA/kg/day), thickened wall and edema and hemorrhagic, necrotic, ulcerated, or perforated mucosa were observed in the large intestine and significant increases in the incidence of squamous metaplasia of the epithelial columnar absorptive cells were found in the cecum, colon, and rectum. Squamous metaplasia was also observed in the cecum and colon of mice chronically exposed to 67.1 mg MMA/kg/day (Arnold et al. 2003; incidence data reported in Gur et al. 1991).

There are some reports of gastrointestinal effects in rats and dogs exposed to DMA; however, the LOAELs for these effects are higher than the LOAELs for MMA and most rodent studies do not report effects at nonlethal doses. Diarrhea with congestion and hemorrhagic gastrointestinal contents were observed in rats exposed to a lethal dose of 190 mg DMA/kg/day in the diet for 4 weeks (Crown et al. 1987) and diarrhea and vomiting were reported in dogs administered 16 mg DMA/kg/day via capsule 6 days/week (Zomber et al. 1989). No gastrointestinal effects were observed in rats or mice chronically exposed to 7.8 or 94 mg DMA/kg/day, respectively (Arnold et al. 2006).

Vomiting and gastrointestinal hemorrhage were observed in dogs after a single capsulized dose of 50 mg/kg roxarsone (Kerr et al. 1963), although slightly higher doses administered for 13 weeks to rats and mice had no effect (NTP 1989b). No gastrointestinal effects were seen after chronic exposure of rats (4 mg/kg/day) or mice (43 mg/kg/day) to roxarsone (NTP 1989b).

Hematological Effects.

Inorganic Arsenicals. Anemia and leukopenia are common effects of arsenic poisoning in humans, and have been reported following acute (Armstrong et al. 1984; Goldsmith and From 1986; Mizuta et al. 1956; Muzi et al. 2001; Westhoff et al. 1975), intermediate (Franzblau and Lilis 1989; Heyman et al. 1956; Nagai et al. 1956; Wagner et al. 1979), and chronic oral exposures (Chakraborti et al. 2003a; Glazener et al. 1968; Guha Mazumder et al. 1988; Hopenhayn et al. 2006; Kyle and Pease 1965; Tay and Seah 1975) at doses of 0.002 mg As/kg/day or more. These effects may be due to both a direct cytotoxic or hemolytic effect on the blood cells (Armstrong et al. 1984; Fincher and Koerker 1987; Goldsmith and From 1986; Kyle and Pease 1965; Lerman et al. 1980) and a suppression of erythropoiesis (Kyle and

Pease 1965; Lerman et al. 1980). However, hematological effects are not observed in all cases of arsenic exposure (EPA 1981b; Harrington et al. 1978; Huang et al. 1985; Silver and Wainman 1952) or even all acute poisoning cases (Cullen et al. 1995; Moore et al. 1994b).

In an acute animal study, Tice et al. (1997) found that there was a decrease in polychromatic erythrocytes in the bone marrow of mice treated with 6 mg As/kg/day for 1 or 4 days. There was no effect at 3 mg As/kg/day. Long-term studies found mild anemia in dogs fed arsenite or arsenate for 2 years at 2.4 mg As/kg/day, but no hematological effect in dogs fed 1 mg As/kg/day for 2 years or 1.9 mg As/kg/day for 26 weeks (Byron et al. 1967; Neiger and Osweiler 1989). Chronic rat studies found little or no evidence of anemia at doses up to 30 mg As/kg/day, even with co-exposure to lead (Byron et al. 1967; Kroes et al. 1974). No hematological effects were found in monkeys exposed to arsenic doses of 3–6 mg As/kg/day for 1 year (Heywood and Sortwell 1979).

Rats exposed to arsenate for 6 weeks had decreased activities of several enzymes involved in heme synthesis, but data were not provided on whether this resulted in anemia (Woods and Fowler 1977, 1978). Exposure of rats to ≥5 ppm of arsenic (0.30 mg As/kg/day as sodium arsenite) in the drinking water for 4 weeks resulted in increased platelet aggregation, while 10 or 25 ppm (0.60 or 1.5 mg As/kg/day) was associated with increased P-selectin-positive cells and decreased occlusion time (Lee et al. 2002), representing a change in platelet function. Similarly, exposure of rats or guinea pigs to 10 or 25 ppm of arsenic as arsenite (approximate doses of 0, 0.92, or 2.3 mg As/kg/day for rats and 0, 0.69, or 1.7 mg As/kg/day for guinea pigs) in the drinking water for 16 weeks (Kannan et al. 2001) resulted in decreases in erythrocyte and leukocyte numbers (rats and guinea pigs), increased blood mean corpuscular volume and corpuscular hemoglobin mass (guinea pigs only), and decreased mean corpuscular hemoglobin concentration (rats only). Gallium arsenide also disrupts heme synthesis in rats, although the evidence suggests that this effect is due primarily to the gallium moiety (Flora et al. 1997a).

Organic Arsenicals. No adverse hematological effects were noted in a man who ingested 78 mg/kg as dimethyl arsenic acid and dimethyl arsenate (Lee et al. 1995). No hematological effects were observed in rats exposed to 8.4 or 72.4 mg MMA/kg/day for 2 years (Arnold et al. 2003; Shen et al. 2003) or dogs administered 35 mg MMA/kg/day for 52 weeks (Waner and Nyska 1988); additionally, no alterations in total or differential leukocyte levels were observed in mice exposed to 67.1 mg MMA/kg/day for 2 years (Arnold et al. 2003). Although some studies have reported hematological alterations following oral exposure to DMA, this is not a consistent finding. Observed alterations include decreased mean corpuscular hemoglobin concentration in rats exposed to 2.3 mg DMA/kg/day for 10 weeks (Rubin et al.

1989), decreased hemoglobin and erythrocyte levels in rats exposed to 4.5 mg DMA/kg/day for 13 weeks (Crown et al. 1987), decreased erythrocyte levels and increased leukocyte levels in dogs administered capsules containing 40 mg DMA/kg/day for 52 weeks (Zomber et al. 1989), and decreased lymphocyte and increased monocyte levels were observed in mice chronically exposed to 94 mg DMA/kg/day (Arnold et al. 2006). No hematological alterations have been observed in rats exposed to 7.8 mg DMA/kg/day for 2 years (Arnold et al. 2006). Similarly, no hematological effects were observed in rats (Kerr et al. 1963; NTP 1989b), mice (NTP 1989b), or dogs (Prier et al. 1963) exposed to 20–32, 68, or 5 mg/kg/day roxarsone, respectively, for intermediate or chronic durations

Musculoskeletal Effects.

Inorganic Arsenicals. No studies were located regarding musculoskeletal effects in humans or animals after oral exposure to inorganic arsenicals.

Organic Arsenicals. No studies were located regarding musculoskeletal effects in humans after oral exposure to organic arsenicals. No musculoskeletal effects were seen after intermediate or chronic exposure of rats and mice to MMA (Arnold et al. 2003), DMA (Arnold et al. 2006), or roxarsone (NTP 1989b).

Hepatic Effects.

Inorganic Arsenicals. A number of studies in humans exposed to inorganic arsenic by the oral route have noted signs or symptoms of hepatic injury. Clinical examination often reveals that the liver is swollen and tender (Chakraborty and Saha 1987; Franklin et al. 1950; Guha Mazumder et al. 1988, 1998a; Liu et al. 2002; Mizuta et al. 1956; Silver and Wainman 1952; Wade and Frazer 1953; Zaldívar 1974), and analysis of blood sometimes shows elevated levels of hepatic enzymes (Armstrong et al. 1984; Franzblau and Lilis 1989; Guha Mazumder 2005; Hernández-Zavala et al. 1998). These effects are most often observed after repeated exposure to doses of 0.01–0.1 mg As/kg/day (Chakraborty and Saha 1987; Franklin et al. 1950; Franzblau and Lilis 1989; Guha Mazumder et al. 1988; Mizuta et al. 1956; Silver and Wainman 1952; Wade and Frazer 1953), although doses as low as 0.006 mg As/kg/day have been reported to have an effect following chronic exposure (Hernández-Zavala et al. 1998). Hepatic effects have also been reported in acute bolus poisoning cases at doses of 2 mg As/kg/day or more (Hantson et al. 1996; Kamijo et al. 1998; Levin-Scherz et al. 1987; Quatrehomme et al. 1992; Vantroyen et al. 2004), although acute exposure to 19 mg As/kg did not cause hepatic effects in an infant (Cullen et al. 1995).

Histological examination of the livers of persons chronically exposed to similar doses has revealed a consistent finding of portal tract fibrosis (Guha Mazumder 2005; Guha Mazumder et al. 1988; Morris et al. 1974; Piontek et al. 1989; Szuler et al. 1979), leading in some cases to portal hypertension and bleeding from esophageal varices (Szuler et al. 1979); cirrhosis has also been reported at an increased frequency in arsenic-exposed individuals (Tsai et al. 1999). Several researchers consider that these hepatic effects are secondary to damage to the hepatic blood vessels (Morris et al. 1974; Rosenberg 1974), but this is not directly established.

Acute exposure of monkeys to 6 mg As/kg/day resulted in vacuolization of the hepatocytes (Heywood and Sortwell 1979). Studies in dogs or mice have not detected clinically significant hepatic injury following exposure to either arsenite or arsenate (Byron et al. 1967; Fowler and Woods 1979; Kerkvliet et al. 1980; Neiger and Osweiler 1989; Schroeder and Balassa 1967), although enlargement of the common bile duct was noted in rats fed either arsenate or arsenite in the diet for 2 years (Byron et al. 1967; Kroes et al. 1974) and lipid vacuolation and fibrosis were seen in the livers of rats exposed to 12 mg As/kg/day as arsenate in the drinking water for 6 weeks (Fowler et al. 1977). Similarly, fatty changes and inflammatory cell infiltration were seen in the livers of both normal and metallothionein-null mice exposed to 5.6 mg arsenic/kg/day in the drinking water for 48 weeks (Liu et al. 2000). Increases in liver zinc and copper concentrations were noted in rats receiving a single oral dose of 10 mg As/kg as sodium arsenite (Flora and Tripathi 1998) and hepatic levels of malondialdehyde were increased and glutathione levels were decreased in livers of rats receiving 200 mg As/kg as GaAs (Flora et al. 1998). An increase in indices of peroxidation was reported in rats dosed with approximately 0.02 mg As/kg/day for 60 days from drinking water containing 2.5 mg sodium arsenite/L (Bashir et al. 2006); absolute liver weight was also increased at this dose level. Elevated levels of serum aspartate aminotransferase (AST) were observed in rats administered a single oral dose of 100 mg As/kg as GaAs (Flora et al. 1998). Exposure of guinea pigs to 0.69 or 1.7 mg As/kg/day in the drinking water for 16 weeks, but not in rats exposed to 0.92 or 2.3 mg As/kg/day, resulted in increases in delta-aminolevulinic acid synthetase (ALAS) levels (Kannan et al. 2001). Exposure of BALB/C mice to 0.7 mg arsenic/kg/day in the drinking water for 15 months resulted in increased liver weights, changes in liver enzymes (glutathione S-transferase, glutathione reductase, catalase, glucose-6-phosphate dehydrogenase, glutathione peroxidase), fatty liver, and fibrosis (Santra et al. 2000).

Organic Arsenicals. No adverse hepatic effects were noted after ingestion of 1,714 mg/kg MSMA or 78 mg DMA/kg (as dimethyl arsenic acid and dimethyl arsenate) in a suicide attempt (Lee et al. 1995; Shum et al. 1995). No other studies of the hepatic effects of organic arsenicals in humans were located.

Histological examination of livers from rabbits given repeated oral doses of MMA showed diffuse inflammation and hepatocellular degeneration (Jaghabir et al. 1989), but the lesions were not severe. Male rats exposed to a time-weighted average (TWA) dose of 72.4 mg MMA/kg/day for 104 weeks showed a decrease in absolute liver weight, while females exposed to 98.5 mg MMA/kg/day showed histiocytic proliferation of the liver (Arnold et al. 2003); however, these effects were probably due to a decrease in body weight and secondary complications of perforation and ulceration of the gastrointestinal effect, respectively. Shen et al. (2003) reported increases in and the number of GST-P-positive foci in the livers of rats exposed to average concentrations of 8.4 mg MMA/kg/day in the diet for 104 weeks. No effects were observed in rats exposed to DMA (Siewicki 1981), but mice exposed to one or two oral doses of 720 mg DMA/kg had decreased liver glutathione and cytochrome P-450 content and serum ornithine decarboxylase activity (Ahmad et al. 1999a). Generalized icterus was reported in dogs after acute exposure to roxarsone (Kerr et al. 1963). Some small fluctuations in liver weight have been noted in rats and mice after intermediate oral exposure to roxarsone, but the toxicological significance of this is not clear and is not observed after chronic exposure of rats and mice to lower doses (NTP 1989b).

Renal Effects.

Inorganic Arsenicals. Most case studies of acute and chronic arsenic toxicity do not report clinical signs of significant renal injury, even when other systems are severely impaired (e.g., Cullen et al. 1995; Franzblau and Lilis 1989; Jenkins 1966; Kersjes et al. 1987; Mizuta et al. 1956; Silver and Wainman 1952). In some cases, elevated serum levels of creatinine or bilirubin have been noted (Armstrong et al. 1984; Levin-Scherz et al. 1987; Moore et al. 1994b), and mild proteinuria may occur (Armstrong et al. 1984; Glazener et al. 1968; Tay and Seah 1975). Acute renal failure in some bolus poisoning episodes (e.g., Fincher and Koerker 1987; Goebel et al. 1990; Levin-Scherz et al. 1987; Lugo et al. 1969; Moore et al. 1994b) is probably a result of fluid imbalances or vascular injury (Rosenberg 1974; Zaldívar 1974). Glomerular congestion has been observed after an acute exposure to high doses (Quatrehomme et al. 1992).

Studies in animals also indicate that the kidney is not a major target organ for inorganic arsenic (Byron et al. 1967; Schroeder and Balassa 1967; Woods and Southern 1989), although some effects have been reported at high exposure levels. Mild histological changes in the renal tubules of monkeys exposed to arsenate for 2 weeks were noted by Heywood and Sortwell (1979), and some mild alterations in renal mitochondria in rats exposed to arsenate for 6 weeks were noted by Brown et al. (1976). Mild proteinuria

(Flora et al. 1998) and an increase in kidney zinc concentration (Flora and Tripathi 1998) have also been noted in rats exposed orally to a single dose of 100 mg As/kg as GaAs or 10 mg As/kg as sodium arsenite, respectively. These data suggest that the kidney is relatively less sensitive to arsenic than most other organ systems, and renal effects are unlikely to be of concern except secondary to fluid imbalances or cardiovascular injury.

Organic Arsenicals. No adverse renal effects were noted after ingestion of 1,714 mg MSMA/kg in a suicide attempt (Shum et al. 1995). Animal studies have reported renal and urinary bladder effects following oral exposure to organic arsenicals; the available data suggest that the urinary system is a more sensitive target for DMA, than for MMA or roxarsone. A decrease in urine volume was observed in rabbits following a single gavage dose of 30 mg MSMA/kg/day (Jaghabir et al. 1988) and a decrease in urine volume (35 mg MMA/kg/day) and an increase in urine specific gravity (8 mg MMA/kg/day) were observed in dogs administered MMA via capsule for 52 weeks (Waner and Nyska 1988). However, these effects may be indicative of dehydration due to diarrhea rather than a direct effect on the kidney. In a 2-year study in rats (Arnold et al. 2003), an increase in the severity of progressive glomerulonephropathy was observed in females at 33.9 mg MMA/kg/day. Hydronephrosis, pyelonephritis, cystitis, and decreases in urine volume and pH were also observed 72.4 mg MMA/kg/day; however, the investigators noted that these lesions probably resulted from urinary tract obstruction, which was secondary to peritonitis caused by gastrointestinal tract ulcerations. An increased incidence of progressive glomerulonephropathy was also observed in male mice exposed to ≥6.0 mg MMA/kg/day in the diet for 2 years (Arnold et al. 2003; incidence data reported in Gur et al. 1991); the investigators (Gur et al. 1991) noted that the kidney lesions were consistent with the normal spectrum of spontaneous lesions and that there were no differences in character or severity of the lesions between the different groups.

Exposure to DMA has resulted in kidney effects in rats and mice exposed to at least 3.1 or 37 mg DMA/kg/day, respectively; no renal effects were observed in dogs exposed to doses as high as 40 mg As/kg/day for 52 weeks (Zomber et al. 1989). In rats, the renal damage is characterized by increased urine volume and pH, decreased urine osmolarity and electrolyte (sodium, potassium, chlorine) levels, increased urinary calcium levels, and increased organ weight, nephrocalcinosis, and necrosis in the renal papillae and/or cortex; an increase in water consumption is also typically observed. The LOAELs for these effects are 5–57 mg DMA/kg/day in intermediate-duration studies (Arnold et al. 1999; Crown et al. 1987; Murai et al. 1993) and 3.1 mg DMA/kg/day in a chronic-duration study (Arnold et al. 2006). Another study did not find renal effects in rats exposed to 16.5 mg DMA/kg/day (Rubin et al. 1989). This study involved exposure to Sprague-Dawley rats compared to Fischer 344 rats used in the studies with

positive results; it is not known if this reflects a difference in strain sensitivity. In mice, progressive glomerulonephropathy was observed at 37 mg DMA/kg/day and nephrocalcinosis was observed at 94 mg DMA/kg/day (Arnold et al. 2006).

Increased kidney weights and minimal tubular epithelial cell degeneration, tubular casts, and focal mineralization were observed in rats exposed to 32 mg/kg/day roxarsone for 13 weeks (NTP 1989b). No adverse effects were observed in rats at doses as high as 20 mg/kg/day (Kerr et al. 1963; NTP 1989b) for 13 weeks or 10 mg/kg/day for 2 years (NTP 1989b; Prier et al. 1963). No adverse renal effects have been observed in mice exposed to roxarsone doses as high as 136 mg/kg/day (NTP 1989b) or 43 mg/kg/day (NTP 1989b; Prier et al. 1963) for intermediate or chronic durations, respectively, or in dogs exposed to 5 mg/kg/day for a chronic duration (Prier et al. 1963).

Damage to the urinary bladder has been observed in several studies in which rats were exposed to DMA. The observed effects include altered bladder cell surface characteristics in rats exposed to 11 mg DMA/kg/day in the diet for 2 weeks (Cohen et al. 2001), increased bladder weight and regenerative proliferation (measured as an increase in BrdU labeling) in bladder epithelium at 5 mg DMA/kg/day for 10 weeks (Arnold et al. 1999), necrosis of bladder epithelium at 10 mg DMA/kg/day for 10 weeks (Arnold et al. 1999), nodular or papillar hyperplasia at 3.4 mg DMA/kg/day for 2 years (Wei et al. 2002), and urothelial vacuolar degeneration and hyperplasia of urothelial cells at 3.1 mg DMA/kg/day for 2 years (Arnold et al. 2006). Vacuolization of the urothelium in the urinary bladder have also been observed in mice exposed to 7.8 mg DMA/kg/day in the diet for 2 years (Arnold et al. 2006). Inconsistent results were found for MMA. Hyperplasia was observed in the bladders of rats exposed to 1 mg As/kg/day as MMA in drinking water for 2 years (Shen et al. 2003), but bladder effects were not observed in another 2-year study (Arnold et al. 2003) in which rats were exposed to doses as high as 34.8 mg As/kg/day as MMA in the diet. No urinary bladder effects were found in rats and mice exposed to 64 or 136 mg/kg/day roxarsone for 13 weeks (NTP 1989b) or 4 or 43 mg/kg/day roxarsone for 2 years (NTP 1989b).

Endocrine Effects.

Inorganic Arsenicals. Very little has been written about the effects of oral exposure to arsenic on endocrine glands. In a report of the autopsies of five children who died in Chile after chronic exposure to arsenic in the drinking water, arterial thickening in the pancreas was noted (Rosenberg 1974). An association has been demonstrated between exposure to arsenic in drinking water and an increased

incidence of diabetes mellitus (Lai et al. 1994; Rahman et al. 1998; Tsai et al. 1999; Tseng et al. 2000; Wang et al. 2003), although dose-response relationships are not available.

Exposure of rats to 2.3 mg As/kg/day as arsenic trioxide for 30 days resulted in reductions in the number of islet cells in the pancreas, as well as significant reductions in pancreatic superoxide dismutase (SOD) and catalase enzyme levels and increases in the production of nitric oxide and malondialdehyde (Mukherjee et al. 2004).

Organic Arsenicals. No studies of effects of organic arsenic compounds on endocrine glands in humans were found. Hypertrophy of thyroid epithelium was observed in rats exposed to 33.9 mg MMA/kg/day in the diet for 2 years (Arnold et al. 2003), 4.0 mg DMA/kg/day in the diet for 13 weeks (Crown et al. 1987), 16.5 mg DMA/kg/day in the diet for at least 10 weeks (Rubin et al. 1989), and 7.8 mg DMA/kg/day in the diet for 2 years (Arnold et al. 2006). No other biologically significant effects were observed in other endocrine tissues following exposure to MMA or DMA. No adverse effects were seen in the adrenal or pituitary glands, thyroid, or pancreas after intermediate or chronic exposure of rats (20–64 or 4 mg/kg/day, respectively) and mice (136 or 43 mg/kg/day, respectively) to roxarsone (NTP 1989b).

Dermal Effects.

Inorganic Arsenicals. One of the most common and characteristic effects of arsenic ingestion is a pattern of skin changes that include generalized hyperkeratosis and formation of hyperkeratotic warts or corns on the palms and soles, along with areas of hyperpigmentation interspersed with small areas of hypopigmentation on the face, neck, and back. These and other dermal effects have been noted in a large majority of human studies involving repeated oral exposure (e.g., Ahmad et al. 1997, 1999b; Ahsan et al. 2000; Bickley and Papa 1989; Borgoño and Greiber 1972; Borgoño et al. 1980; Cebrián et al. 1983; Chakraborti et al. 2003a, 2003b; Chakraborty and Saha 1987; Foy et al. 1992; Franklin et al. 1950; Franzblau and Lilis 1989; Guha Mazumder et al. 1988, 1998a, 1998b, 1998c; Guo et al. 2001a; Haupert et al. 1996; Huang et al. 1985; Lander et al. 1975; Liu et al. 2002; Lüchtrath 1983; Milton et al. 2004; Mizuta et al. 1956; Morris et al. 1974; Nagai et al. 1956; Piontek et al. 1989; Rosenberg 1974; Saha and Poddar 1986; Silver and Wainman 1952; Szuler et al. 1979; Tay and Seah 1975; Tseng et al. 1968; Wade and Frazer 1953; Wagner et al. 1979; Wong et al. 1998a, 1998b; Zaldívar 1974, 1977). In cases of low-level chronic exposure (usually from water), these skin lesions appear to be the most sensitive indication of effect, so this end point is considered to be the most appropriate basis for establishing a chronic oral MRL. This is supported by the finding that other effects (hepatic injury, vascular disease, neurological

effects) also appear to have similar thresholds. As shown in Table 3-3 and Figure 3-3, numerous studies in humans have reported dermal effects at chronic dose levels generally ranging from about 0.01 to 0.1 mg As/kg/day (Ahmad et al. 1997; Bickley and Papa 1989; Borgoño and Greiber 1972; Borgoño et al. 1980; Cebrián et al. 1983; Chakraborty and Saha 1987; Foy et al. 1992; Franklin et al. 1950; Guha Mazumder et al. 1988; Huang et al. 1985; Lüchtrath 1983; Piontek et al. 1989; Silver and Wainman 1952; Tseng et al. 1968; Zaldívar 1974, 1977). However, in a study with detailed exposure assessment, all confirmed cases of skin lesions ingested water containing >100 µg/L arsenic (approximately 0.0037 mg As/kg/day) and the lowest known peak arsenic concentration ingested by a case was 0.115 µg/L (approximately 0.0043 mg As/kg/day) (Haque et al. 2003). Another large study reported increased incidence of skin lesions associated with estimated doses of 0.0012 mg As/kg/day (0.023 mg As/L drinking water) (Ahsan et al. 2006). Several epidemiological studies of moderately sized populations (20–200 people) exposed to arsenic through drinking water have detected no dermal or other effects at average chronic doses of 0.0004–0.01 mg As/kg/day (Cebrián et al. 1983; EPA 1981b; Guha Mazumder et al. 1988; Harrington et al. 1978; Valentine et al. 1985), and one very large study detected no effects in any person at an average total daily intake (from water plus food) of 0.0008 mg As/kg/day (Tseng et al. 1968). This value has been used to calculate a chronic oral MRL for inorganic arsenic of 0.0003 mg/kg/day, as described in footnote c in Table 3-3.

Another prominent dermal effect associated with chronic ingestion of inorganic arsenic is skin cancer. As discussed in greater detail in Section 3.2.2.7 (below), some of these skin cancers may evolve from the hyperkeratotic corns or warts, while the areas of altered pigmentation are not considered to be precancerous (EPA 1988d).

Dermal lesions similar to those observed in humans have not been noted in oral exposure studies in monkeys (Heywood and Sortwell 1979), dogs (Byron et al. 1967), or rodents (Schroeder et al. 1968). However, a hyperplastic response to oral arsenic exposure was reported in arsenic-exposed mice (Rossman et al. 2004).

Organic Arsenicals. No studies were located regarding dermal effects in humans after oral exposure to organic arsenicals. No gross or histological skin alterations were observed in rats or mice following intermediate- or chronic-duration exposure to MMA (Arnold et al. 2003; as reported in Crown et al. 1990; Gur et al. 1991), DMA (Arnold et al. 2006; as reported in Gur et al. 1989a, 1989b), or roxarsone (NTP 1989b)

Ocular Effects.

Inorganic Arsenicals. Periorbital swelling was reported in people drinking contaminated well water at an approximate dose of 0.2 mg As/kg for 1 week (Armstrong et al. 1984). Facial edema, generally involving the eyelids, was a prominent feature of arsenic poisoning among 220 cases associated with an episode of arsenic contamination of soy sauce in Japan (Mizuta et al. 1956). Exposure was to an estimated dose of 0.05 mg/kg/day and lasted for up to 2–3 weeks. The edema developed soon after the initial exposure and then subsided. This effect forms the basis (in part) for the acute oral MRL of 0.005 mg/kg/day for inorganic arsenic, as described in footnote b in Table 3-3. Nemec et al. (1998) noted the appearance of dried red material around the eyes of mice receiving daily oral doses of 24 mg As/kg as arsenic acid for 10 days during gestation.

Organic Arsenicals. No studies were located regarding ocular effects in humans or animals after oral exposure to organic arsenicals. No gross or histological alterations in the eye were observed in rats or mice following intermediate- or chronic-duration exposure to MMA (Arnold et al. 2003; as reported in Crown et al. 1990; Gur et al. 1991), DMA (Arnold et al. 2006; as reported in Gur et al. 1989a, 1989b), or roxarsone (NTP 1989b).

Body Weight Effects.

Inorganic Arsenicals. A 41-year old woman exposed to arsenic in the drinking water for 4 months at an approximate dose of 0.06 mg As/kg/day reported losing 40 pounds (18 kg) of body weight before seeking treatment (Wagner et al. 1979). Weight loss was also among the effects observed in a series of 475 chronic arsenism patients hospitalized in Antofagasto, Chile after receiving approximate doses of 0.02 mg As/kg/day in the drinking water for an unspecified number of years (Zaldívar 1974).

Reductions in body weight gain are commonly seen in animal studies of ingested arsenic. In pregnant rats, body weight gain was reduced by gavage treatment with 23 mg As/kg/day as arsenic trioxide on day 9 of gestation (NOAEL=15 mg As/kg/day, Stump et al. 1999), and by repeated gavage treatment with 8 mg As/kg/day as arsenic trioxide from 2 weeks prior to mating through gestation (NOAEL=4 mg As/kg/day, Holson et al. 2000). Exposure of rats by gavage to 26.6 mg As/kg/day as sodium arsenite, but not 13.3 mg As/kg/day or lower, 5 days/week for 4 weeks resulted in a significant decrease in body weight (Schulz et al. 2002). In 6-week rat studies, body weight gain was decreased at 11–12 mg As/kg/day, but not at 6–9 mg As/kg/day (Brown et al. 1976; Fowler et al. 1977). In a 12-week oral

gavage study, rats dosed with 1.5 mg/kg/day sodium arsenite had a median final body weight 18% lower than controls Dhar et al. (2005). A 60-day rat study with sodium arsenite in the drinking water reported a 13% reduction in final body weight in rats dosed with approximately 0.02 mg As/kg/day (Bashir et al. 2006). In chronic rat studies of arsenate and arsenite, body growth decreases were found at doses as low as 2 mg As/kg/day in feeding studies (Byron et al. 1967; Kroes et al. 1974), while rats exposed to lower levels of sodium arsenite in the drinking water (0.6 mg As/kg/day) throughout their lifetimes grew normally (Schroeder et al. 1968). Rats given a single oral dose of 100 mg As/kg as GaAs exhibited a 15% reduction in body weight compared to controls 7 days after exposure (Flora et al. 1998). Body weight gain was decreased in mice at 24 mg As/kg/day in a gestation exposure study (Nemec et al. 1998), 10 mg As/kg/day in a 6-week study (Fowler and Woods 1979), and 1 mg As/kg/day in a 2-year study (Schroeder and Balassa 1967). Growth was unaffected in mice that received 12 mg As/kg/day in the gestation exposure study (Nemec et al. 1998), 5 mg As/kg/day in the 6-week study (Fowler and Woods 1979), or 0.7–0.8 mg As/kg/day in 1–3 month arsenate drinking water studies (Healy et al. 1998). Dogs chronically treated with 2.4 mg As/kg/day as sodium arsenite lost 44–61% of their starting body weight and died, while lower doses had no effect on growth (Byron et al. 1967). Weight depression was also reported in dogs chronically treated with 2.4 mg As/kg/day as sodium arsenate (Byron et al. 1967). Feed consumption and body weight gain were significantly reduced in a dose-related manner in dogs fed 1.5 or 1.9 mg As/kg/day as sodium arsenite in the diet (Neiger and Osweiler 1989). Dogs in the high-dose group lost 25% of their body weight over the 17-week study period. Pair-fed controls lost weight at the same rate as high-dose dogs, showing that the effect on body weight was due to reduced feed consumption, rather than a direct effect of arsenic.

Organic Arsenicals. No studies were located regarding body weight effects in humans after oral exposure to organic arsenicals. In animal studies of organic arsenicals, decreases in body weight gain were observed in rats and mice after acute, intermediate, and chronic duration exposure to MMA (Arnold et al. 2003; Waner and Nyska 1988), DMA (Murai et al. 1993), and roxarsone (NTP 1989b); decreases in body weight gain have also been reported in pregnant rats and rabbits exposed to MMA (Irvine et al. 2006) or DMA (Irvine et al. 2006; Rogers et al. 1981). For MMA, the decreases in body weight gain were observed following intermediate-duration exposure of rats and dogs to 106.9 or 8 mg MMA/kg/day (Arnold et al. 2003; Waner and Nyska 1988), respectively, and following chronic-duration exposure of rats, mice, and dogs to 25.7, 67.1, or 8 mg MMA/kg/day, respectively (Arnold et al. 2003; Waner and Nyska 1988). The decreases in body weight gain occurred at doses that were associated with diarrhea and histological alterations in the gastrointestinal tract (Arnold et al. 2003; Waner and Nyska 1988). One DMA study in nonpregnant animals reported decreases in body weight gain in rats administered 57 mg

DMA/kg/day via gavage 5 days/week for 4 weeks (Murai et al. 1993); other DMA studies have not reported decreases in body weight gain in rats following exposure to 11 mg DMA/kg/day for acute durations (Cohen et al. 2001), 3.7–60 mg DMA/kg/day for intermediate durations (Arnold et al. 1999; Crown et al. 1987; Rubin et al. 1989; Siewicki 1981; Wanibuchi et al. 1996; Yamamoto et al. 1995), or 0.77 mg DMA/kg/day for chronic durations (Arnold et al. 2006). No alterations in body weight gain were observed in mice exposed to 94 mg DMA/kg/day for 2 years (Arnold et al. 2006). The lowest doses of roxarsone to produce a decrease in growth were 32 and 16 mg/kg/day in rats following acute- or intermediate-duration exposure, respectively, and 168 and 136 mg/kg/day in mice following acute or intermediate exposure (NTP 1989b); at the highest dose tested in chronic studies, no significant alterations in body weight gain were observed in rats at 4 mg/kg/day or in mice at 43 mg/kg/day (NTP 1989b).

3.2.2.3 Immunological and Lymphoreticular Effects

Inorganic Arsenicals. No studies were located regarding immunological and lymphoreticular effects in humans after oral exposure to inorganic arsenicals. No evidence of immunosuppression was detected in mice exposed to arsenate at levels up to 100 ppm (20 mg As/kg/day) in drinking water (Kerkvliet et al. 1980). This NOAEL is shown in Table 3-3 and Figure 3-3. Gallium arsenide at doses of 52–260 mg As/kg/day produced significant, dose-related decreases in relative spleen weight, spleen cellularity, humoral immune response (antibody forming cell response to sheep RBC), and delayed type hypersensitivity in rats (Flora et al. 1998). However, it is not clear to what extent these effects are due to the arsenic moiety.

Organic Arsenicals. No studies were located regarding immunological and lymphoreticular effects in humans or animals after oral exposure to organic arsenicals. No histological alterations were observed in immunological or lymphoreticular tissues following intermediate-duration exposure of rats to 43.2 mg DMA/kg/day in the diet (Crown et al. 1987) or rats and mice to 18.23 or 38.7 mg As/kg/day as roxarsone, respectively (NTP 1989b) or following chronic-duration exposure of rats and mice to 72.4 or 67.1 mg MMA/kg/day (Arnold et al. 2003), 7.8 or 94 mg DMA/kg/day (Arnold et al. 2006), or 4 or 43 mg/kg/day roxarsone (NTP 1989b). No studies examined immune function following oral exposure to organic arsenicals.

3.2.2.4 Neurological Effects

Inorganic Arsenicals. A large number of epidemiological studies and case reports indicate that ingestion of inorganic arsenic can cause injury to the nervous system. Acute, high-dose exposures (2 mg As/kg/day or above) often lead to encephalopathy, with signs and symptoms such as headache, lethargy, mental confusion, hallucination, seizures, and coma (Armstrong et al. 1984; Bartolome et al. 1999; Civantos et al. 1995; Cullen et al. 1995; Danan et al. 1984; Fincher and Koerker 1987; Levin-Scherz et al. 1987; Quatrehomme et al. 1992; Uede and Furukawa 2003; Vantroyen et al. 2004). Repeated exposures to lower levels (0.03–0.1 mg As/kg/day) are typically characterized by a symmetrical peripheral neuropathy (Chakraborti et al. 2003a, 2003b; Foy et al. 1992; Franzblau and Lilis 1989; Guha Mazumder et al. 1988; Hindmarsh et al. 1977; Huang et al. 1985; Lewis et al. 1999; Mizuta et al. 1956; Muzi et al. 2001; Silver and Wainman 1952; Szuler et al. 1979; Wagner et al. 1979). This neuropathy usually begins as numbness in the hands and feet, but later may develop into a painful "pins and needles" sensation. Both sensory and motor nerves are affected, and muscle weakness often develops, sometimes leading to wrist-drop or ankle-drop (Chhuttani et al. 1967; Heyman et al. 1956). Diminished sensitivity to stimulation and abnormal patellar reflexes have also been reported (Mizuta et al. 1956). Histological examination of nerves from affected individuals reveals a dying-back axonopathy with demyelination (Goebel et al. 1990; Hindmarsh and McCurdy 1986). Some recovery may occur following cessation of exposure, but this is a slow process and recovery is usually incomplete (Fincher and Koerker 1987; Le Quesne and McLeod 1977; Murphy et al. 1981). Peripheral neuropathy is also sometimes seen following acute high-dose exposures, with or without the previously described encephalopathy (Armstrong et al. 1984; Baker et al. 2005; Fincher and Koerker 1987; Goebel et al. 1990; Hantson et al. 1996; Kamijo et al. 1998). Neurological effects were not generally found in populations chronically exposed to doses of 0.006 mg As/kg/day or less (EPA 1981b; Harrington et al. 1978; Hindmarsh et al. 1977), although fatigue, headache, dizziness, insomnia, nightmare, and numbness of the extremities were among the symptoms reported at 0.005, but not 0.004 mg As/kg/day in a study of 31,141 inhabitants of 77 villages in Xinjiang, China (Lianfang and Jianzhong 1994), and depression was reported in some Wisconsin residents exposed to 2–10 µg As/L in the drinking water for 20 years or longer (Zierold et al. 2004).

There is emerging evidence suggesting that exposure to arsenic may be associated with intellectual deficits in children. For example, Wasserman et al. (2004) conducted a cross-sectional evaluation of intellectual function in 201 children 10 years of age whose parents were part of a larger cohort in Bangladesh. Intellectual function was measured using tests drawn from the Wechsler Intelligence Scale for Children; results were assessed by summing related items into Verbal, Performance, and Full-Scale

raw scores. The mean arsenic concentration in the water was 0.118 mg/L. The children were divided into four exposure groups, representing <5.5, 5.6–50, 50–176, or 177–790 µg As/L drinking water. After adjustment for confounding factors, a dose-related inverse effect of arsenic exposure was seen on both Performance and Full-Scale subset scores; for both end points, exposure to ≥50 µg/L resulted in statistically significant differences (p<0.05) relative to the lowest exposure group (<5.5 µg/L). In a later report, the same group of investigators examined 301 6-year-old children from the same area (Wasserman et al. 2007). In this case, the children were categorized into the following quartiles based on water arsenic concentration: 0.1–20.9, 21–77.9, 78–184.9, and 185–864 µg/L. After adjustment for water Mn, blood lead, and sociodemographic features known to contribute to intellectual function, water arsenic was significantly negatively associated with both Performance and Processing speed raw scores. Analyses of the dose-response showed that compared to the first quartile, those in the second and third categories had significantly lower Performance raw scores (p<0.03 and p=0.05, respectively). Those in the fourth category had marginally significantly lower Full-Scale and Processing Speed raw scores. It should be mentioned, however, that in general, arsenic in the water explained <1% of the variance in test scores. Water arsenic made no contribution to IQ outcomes. A study of 351 children age 5–15 years from West Bengal, India, found significant associations between urinary arsenic concentrations and reductions in scores of tests of vocabulary, object assembly, and picture completion; the magnitude of the reductions varied between 12 and 21% (von Ehrenstein et al. 2007). In this cohort, the average lifetime peak arsenic concentration in well water was 0.147 mg/L. However, no clear pattern was found for increasing categories of peak arsenic water concentrations since birth and children's scores in the various neurobehavioral tests conducted. Furthermore, using peak arsenic as a continuous variable in the regression models also did not support an adverse effect on the tests results. Exposure to arsenic *in utero* also did not suggest an association with the tests scores. Von Ehrestein et al. (2007) concluded that the study provided little evidence for an effect of long-term arsenic concentrations in drinking water and that the lack of findings with past exposures via drinking water may be due to incomplete assessment of past exposure, particularly exposure originating from food. Wasserman's results are consistent with those of ecological studies in children in Taiwan (Tsai et al. 2003) and in China (Wang et al. 2007). In the former, adolescents exposed to low (0.0017–0.0018 mg As/kg/day; n=20) levels of inorganic arsenic in the drinking water showed decreased performance in the switching attention task, while children in the high exposure group (0.0034–0.0042 mg As/kg/day; n=29) showed decreased performance in both the switching attention task and in tests of pattern memory, relative to unexposed controls (n=60). In the study in China (age 8– 12 years), 87 children whose mean arsenic concentration in the drinking water was 0.190 mg/L had a mean IQ score of 95 compared with 101 for children (n=253) with 0.142 mg/L arsenic in the water and 105 for control children (n=196) with 0.002 mg/L arsenic in the drinking water (Wang et

al. 2007). The differences in IQ scores between the two exposure groups and the control group were statistically significant.

Neurological effects have also been observed in animal studies. Rodriguez et al. (2001) evaluated neurobehavioral changes in male Sprague-Dawley rats exposed to 0, 5, 10, or 20 mg As/kg/day as sodium arsenite by gavage for 2 or 4 weeks; significant effects were seen in spontaneous locomotor activity and the food pellet manipulation test in the high-dose animals, while no effects were seen in the low- or mid-dose rats. Decreased performance in open field tests were also seen in rats exposed to 26.6 mg As/kg/day, but not to 13.3 mg/kg/day or less, as sodium arsenite for 4 weeks (Schulz et al. 2002); curiously, the behavioral changes were no longer present at 8 and 12 weeks of exposure, which may suggest an adaptive response. Heywood and Sortwell (1979) reported salivation and uncontrolled head shaking in two monkeys given several doses of 6 mg As/kg/day as arsenate, while no such effects were noted in monkeys given 3 mg As/kg/day for 2 weeks. Nemec et al. (1998) observed ataxia and prostration in pregnant female rabbits treated with 1.5 mg As/kg/day repeatedly during gestation, but not in rabbits treated with 0.4 mg As/kg/day. Some changes in levels of neurotransmitters (dopamine, norepinephrine, and 5-hydroxytryptamine) were seen in rats exposed to 2.3 mg As/kg/day as sodium arsenite and guinea pigs exposed to 1.7 mg As/kg/day as sodium arsenite in the drinking water for 16 weeks (Kannan et al. 2001) or in rats exposed to 0.14 mg As/kg/day as sodium arsenite by gavage for 28 days (Chattopadhyay et al. 2001), but the functional significance of these changes is not clear.

The highest NOAEL values and all reliable LOAEL values for neurological effects from inorganic arsenic in each species and duration category are recorded in Table 3-3 and plotted in Figure 3-3.

Organic Arsenicals. Numbness and tingling of the fingertips, toes, and circumoral region were reported by a women exposed to an unspecified amount of organic arsenic in bird's nest soup. Discontinuation of exposure resulted in the disappearance of symptoms (Luong and Nguyen 1999). Decreased absolute brain weights were seen in male rats exposed to 25.7 mg MMA/kg/day and female rats exposed to ≥33.9 mg MMA/kg/day, but decreased body weight also occurred at these exposure levels, and relative brain weights were increased in the males at 25.7 mg MMA/kg/day and the females at ≥33.9 mg MMA/kg/day in this study (Arnold et al. 2003). No neurological clinical signs or brain lesions were observed following chronic exposure of rats to 72.4 mg MMA/kg/day or mice to 67.1 mg MMA/kg/day (Arnold et al. 2003). Decreased spontaneous motility, increased startle response, and ataxia were observed in mice receiving a single gavage dose of 1,757 mg DMA/kg/day (Kaise et al. 1989); no other evidence (clinical signs or histological alterations) were observed in chronic studies of DMA in which

rats and mice were exposed to 7.8 or 94 mg DMA/kg/day, respectively (Arnold et al. 2006). Two studies in pigs indicate that repeated oral doses of roxarsone (6.3–20 mg/kg/day for 1 month) can cause significant neurotoxicity (Edmonds and Baker 1986; Kennedy et al. 1986; Rice et al. 1985). The main signs were time-dependent degenerations of myelin and axons (Kennedy et al. 1986; Rice et al. 1985). Evidence of neurological effects (hyperexcitability, ataxia, trembling) was noted in some rat and mouse studies (Kerr et al. 1963; NTP 1989b). Reliable NOAELs and LOAELs are presented in Tables 3-4, 3-5, and 3-6, and Figures 3-4, 3-5, and 3-6.

3.2.2.5 Reproductive Effects

Inorganic Arsenicals. Exposure to arsenic in drinking water has been associated with adverse reproductive outcomes in some studies. For example, a study of 96 women in Bangladesh who had been drinking water containing ≥0.10 mg As/L (approximately 0.008 mg As/kg/day) for 5–10 years reported a significant increase in spontaneous abortions (p=0.008), stillbirth (p=0.046), and preterm birth (p=0.018) compared to a nonexposed group (Ahmad et al. 2001). Similar results were reported by Milton et al. (2005) who found a significant association between concentrations of arsenic in the water >0.05 mg/L (approximately 0.006 mg As/kg/day) and spontaneous abortion (odds ratio [OR]=2.5; 95% CI=1.5–4.3) in a study of 533 women, also from Bangladesh. A study of 202 women from West Bengal, India, reported that exposure to arsenic concentrations of arsenic ≥0.2 mg/L in drinking water (approximately 0.02 mg As/kg/day) during pregnancy were associated with a 6-fold increased risk of stillbirth (OR=6.1; 95% CI=1.54–24.0) after adjustment for confounders (von Ehrenstein et al. 2006). No association was found between arsenic exposure and risk of spontaneous abortion (OR=1.01; 95% CI=0.73–10.8). An earlier study of 286 women in the United States also found no significant association between arsenic in the drinking water (0.0016 mg/L; approximately 0.00005 mg As/kg/day) and spontaneous abortion (OR=1.7; 95% CI=0.7–4.2) (Aschengrau et al. 1989).

Lugo et al. (1969) reported a case of a 17-year-old mother who ingested inorganic arsenic (Cowley's Rat and Mouse Poison) at week 30 of pregnancy. Twenty-four hours after ingestion of approximately 30 mL of arsenic trioxide (0.39 mg As/kg), she was admitted for treatment of acute renal failure. She went into labor and delivered a live female infant weighing 2 pounds, 7 ounces with a 1-minute Apgar score of 4. The infant's clinical condition deteriorated and she died at 11 hours of age.

Reproductive performance was not affected in female rats that received gavage doses of 8 mg As/kg/day (as As_2O_3) from 14 days prior to mating through gestation day 19 (Holson et al. 2000). Reproductive

indices that were evaluated included the precoital interval (time to mating), mating index (percentage of rats mated), and fertility index (percentage of matings resulting in pregnancy). In a 3-generation study in mice given sodium arsenite in drinking water at an average dose of 1 mg As/kg/day, there was a significant increase in the incidence of small litters and a trend toward a decreased number of pups per litter in all three generations of the treated group (Schroeder and Mitchener 1971). This finding is consistent with the results of developmental toxicity studies reported in Section 3.2.2.6. Female rats exposed to 0.24 mg As/kg/day (as arsenite) for 28 days showed changes in several reproductive system end points, including decreases in wet weights of the ovary and uterus, inhibition of steroidogenic enzymes, decreased ovarian and uterine peroxidase activities, and decreased estradiol levels relative to controls (Chattopadhyay et al. 2001). NOAEL and LOAEL values from these studies are shown in Table 3-3 and Figure 3-3.

Organic Arsenicals. No studies were located regarding reproductive effects in humans after oral exposure to organic arsenicals. No histological alterations in male or female reproductive tissues were observed in laboratory animals following exposure to MMA (Arnold et al. 2003), DMA (Arnold et al. 2006), or roxarsone (NTP 1989b) and no alterations in sperm parameters were observed in male rats exposed to 76 mg MMA/kg/day for at least 14 weeks (Schroeder 1994). However, some functional alterations have been reported in animals exposed to MMA or DMA. A decrease in estrus was observed in dogs exposed to 35 mg MMA/kg/day for 52 weeks (Waner and Nyska 1988); decreases in body weight gain (terminal body weight was 59% lower than controls) were also observed at this dose level and the effect may have been secondary to systemic toxicity. Decreases in pregnancy rate and male fertility index were observed in F_0 and F_1 rats exposed to 76 mg MMA/kg/day for 14 weeks prior to mating and during the mating, gestation, and lactation periods (Schroeder 1994). In the F_0 animals, the pregnancy rate and male fertility index were not statistically different from controls; however, the values were below historical controls and the investigators considered the effect to be treatment-related. In the F_1 animals, the male fertility index was statistically different from controls but the pregnancy rate was not; both parameters were within the range found in historical controls, but the investigators considered the effect to be treatment-related due to the consistency of the findings in the F_0 and F_1 animals. Impaired fertility, as evidenced by a decreased number of litters, was observed in male mice dosed with MSMA (119 mg/kg/day) during a 19-day mating period with unexposed females (Prukop and Savage 1986); the poor reporting of the study protocol and results precludes drawing conclusions from this study. An increase in the number of does with aborted fetuses was observed in rabbits exposed to 48 mg DMA/kg/day as DMA (Irvine et al. 2006); severe maternal toxicity (weight loss, reduced food intake, and

diarrhea) was also observed at this dose level. No reproductive effects were observed in a 2-generation rat study in which rats were exposed to 16.5 mg DMA/kg/day (Rubin et al. 1989).

3.2.2.6 Developmental Effects

Inorganic Arsenicals. Whether ingestion of inorganic arsenic may cause developmental effects in humans has not been extensively investigated. Lugo et al. (1969) reported a case of a mother who ingested inorganic arsenic (Cowley's Rat and Mouse Poison) at 30 weeks of gestation. Twenty-four hours after ingestion, she went into labor and delivered a live female infant weighing 2 pounds, 7 ounces with a 1-minute Apgar score of 4. The infant's clinical condition deteriorated with frequent episodes of apnea and bradycardia; subsequent venous blood gas determinations documented hypoxia, hypercapnea, and acidosis. The infant died at 11 hours of age. Autopsy performed 8 hours after death showed organ immaturity, generalized petechial hemorrhages, and hyaline membrane disease. Severe intra-alveolar pulmonary hemorrhage was remarkable. High arsenic levels were found in the infant's liver, kidney, and brain, demonstrating easy passage of inorganic arsenic across the placenta. The authors considered most of the findings in the neonate to be attributable to immaturity, but suggested that arsenic may have played a role in the severe intra-alveolar hemorrhaging that contributed to death.

Chronic exposure of women to arsenic in the drinking water has been associated with infants with low birth weights in Taiwan (Yang et al. 2003) and Chile (Hopenhayn et al. 2003a). Similar associations have been made between late fetal mortality, neonatal mortality, and postneonatal mortality and exposure to high levels of arsenic in the drinking water (up to 0.86 mg/L during over a decade), based on comparisons between subjects in low- and high-arsenic areas of Chile (Hopenhayn-Rich et al. 2000). More recently, von Ehrenstein et al. (2006) reported no significant association between exposure to concentrations of ≥0.1 mg/L arsenic in drinking water (approximately 0.008 mg As/kg/day) (n=117; 29 women were exposed to ≥0.5 mg/L) and increased risk for neonatal death or infant mortality during the first year of life in a study of a population in West Bengal, India. The same group of investigators reported significantly increased SMRs for lung cancer and bronchiectasis among subjects in a city in Chile who had probable exposure *in utero* (maternal exposure) or during childhood to high levels of arsenic (near 0.9 mg/L) in the drinking water (Smith et al. 2006). For those exposed in early childhood, the SMR for lung cancer was 7.0 (95% CI=5.4–8.9, $p<0.001$) and for bronchiecstasis 12.4 (95% CI=3.3–31.7, $p<0.001$). For those born during the high-exposure period, the corresponding SMRs were 6.1 (95% CI=3.5–9.9, $p<0.001$) and 46.2 (95% CI=21.1–87.7, $p<0.001$). The mortality data analyzed were for the age range 30–49 years.

No overall association between arsenic in drinking water and congenital heart defects was detected in a case-control study in Boston (Zierler et al. 1988), although an association with one specific lesion (coarctation of the aorta) was noted (OR=3.4, 95% CI=1.3–8.9). A study of 184 women with neural tube defects in the offspring living in a Texas county bordering Mexico found that exposure to levels of arsenic in the drinking water >0.010 mg/L (range or upper limit not specified) did not significantly increase the risk for neural tube defects (OR=2.0, 95% CI=0.1–3.1) (Brender et al. 2006).

Studies in animals, however, suggest that ingested inorganic arsenic may produce developmental effects at high doses that also produce overt maternal toxicity. Rats treated with a single gavage dose of 23 mg As/kg as arsenic trioxide on day 9 of gestation had a significant increase in postimplantation loss and a decrease in viable fetuses per litter, while those treated with 15 mg As/kg showed no effects (Stump et al. 1999). Rats treated by daily gavage with 8 mg As/kg/day starting 14 days before mating and continuing through gestation had significantly reduced fetal body weights and significantly increased incidences of several skeletal variations (unossified sternebrae #5 or #6, slight or moderate sternebrae malalignment, 7th cervical ribs) that the researchers considered to be consequences of developmental growth retardation (Holson et al. 2000). No developmental effects were found at 4 mg As/kg/day in this study. Exposure of rats to 2.93–4.20 mg As/kg/day throughout gestation and for 4 months postnatally resulted in alterations in neurobehavioral parameters in the offspring, including increased spontaneous locomotor activity and number of errors in a delayed alternation task; maternal behavior was not affected (Rodriguez et al. 2002). Studies in mice found increased fetal mortality, decreased fetal body weight, a low incidence of gross malformations (primarily exencephaly), and an increase in skeletal malformations in mice given single gavage doses of 23–48 mg As/kg during gestation (Baxley et al. 1981; Hood et al. 1978), with no effects at 11 mg As/kg. Similarly, in mice treated with 24 mg As/kg/day as arsenic acid on days 6–15 of gestation, there was a significant increase in the number of resorptions per litter (42% vs. 4% in controls) and significant decreases in the number of live pups per litter (6.6 vs. 12.3 in controls) and mean fetal weight (1.0 g vs. 1.3 g in controls), while no developmental effects were found at 12 mg As/kg/day (Nemec et al. 1998). Hamsters treated with a single gavage dose of 14 mg As/kg during gestation also had increased fetal mortality and decreased fetal body weight (Hood and Harrison 1982), with no effect at 11 mg As/kg. However, the most sensitive species was the rabbit, which had increased resorptions and decreased viable fetuses per litter at 1.5 mg As/kg/day and a developmental NOAEL of 0.4 mg As/kg/day, following repeated gavage dosing with arsenic acid during gestation (Nemec et al. 1998). In each of these studies (except Hood et al. 1978, which failed to report maternal effects), overt maternal toxicity, including death in some cases, was found at the same or lower doses as the developmental

effects (Baxley et al. 1981; Holson et al. 2000; Hood and Harrison 1982; Nemec et al. 1998; Stump et al. 1999).

It is noteworthy that the effect in the 3-generation reproduction study in mice by Schroeder and Mitchener (1971), decreased pups per litter (all generations), is consistent with the findings of many of these shorter-term studies (Baxley et al. 1981; Hood and Harrison 1982; Hood et al. 1978; Nemec et al. 1998; Stump et al. 1999). The dose in this long-term study was 1 mg As/kg/day; in a 2-year study by these researchers, this dose produced effects such as decreased body weight gain and increased mortality (Schroeder and Balassa 1967).

A series of studies presented evidence that inorganic arsenic may be a transplacental carcinogen in animals. Waalkes et al. (2003, 2004a, 2004b, 2004c) exposed timed-pregnant AJ mice to 0, 42.5, or 85 ppm of sodium arsenite in the drinking water from gestation day 8 through 18 and observed the offspring for 90 weeks following birth; the study authors estimated daily doses at 9.55 and 19.3 mg As/kg/day. A dose-related increase was reported in the incidence of hepatocellular carcinomas and adrenal tumors in the male offspring from both treatment levels, while male offspring from high-dose animals showed an increase in total number of tumors. In female offspring, an increase in uterine hyperplasia was seen in the offspring of both treated groups while the offspring of high-dose animals showed increased incidence of lung carcinomas. For both exposed groups, regardless of gender, the offspring showed a significant increase in the number of malignant tumors (Waalkes et al. 2003). More recent studies from the same group of investigators have suggested that aberrant estrogen signaling, potentially through inappropriate estrogen receptor-α (ER-α), may play a role in arsenic-induced liver tumors in male offspring (Waalkes et al. 2006a) and in arsenic-induced uterine and bladder carcinoma in female offspring (Waalkes et al. 2006b). The latter was based on the observation of over-expression of ER-α and *pS*2, an estrogen-regulated gene, in the respective tissues.

These studies (shown in Table 3-3 and Figure 3-3) indicate that the fetus may be affected by ingested arsenic.

Organic Arsenicals. No studies were located regarding developmental effects in humans after oral exposure to organic arsenicals. The developmental toxicity of organic arsenicals has been investigated in rats and rabbits for MMA and in rats, mice, and rabbits for DMA. Decreased fetal weights and an increased incidence of fetuses with incomplete ossification of thoracic vertebrae were observed in the offspring of rats administered via gavage 500 mg MMA/kg/day on gestational days 6–15; no

developmental effects were observed at 100 mg MMA/kg/day (Irvine et al. 2006). Decreases in maternal body weight gain were observed at 100 and 500 mg MMA/kg/day. A decrease in pup survival was observed in F_1 and F_2 offspring of rats exposed to 76 mg MMA/kg/day (Schroeder 1994); although pup survival was not statistically different from controls, the investigators considered the effect to be biologically significant because survival in the MMA pups was outside the lower range of survival in historical controls. Increases in the number of fetuses with supernumerary thoracic ribs and eight lumbar vertebrae were observed in the offspring of rabbits administered to 12 mg MMA/kg/day on gestational days 7–19 (Irvine et al. 2006); the investigators noted that these effects were probably secondary to maternal stress.

No developmental effects were observed in the offspring of rats administered via gavage 15 mg DMA/kg/day on gestational days 7–16 (Rogers et al. 1981). At 30 mg DMA/kg/day, there was an increase in the percentage of fetuses with irregular palatine rugae; no maternal effects were observed at this dose level (Rogers et al. 1981). The investigators noted that the functional significance of aberrant rugae in rats is not known. Doses of ≥36 mg DMA/kg/day resulted in decreases in fetal weights and delays in ossification (Chernoff et al. 1990; Irvine et al. 2006; Rogers et al. 1981); decreases in maternal body weight gain were often observed at the same dose levels. Irvine et al. (2006) also reported an increase in the occurrence of diaphragmatic hernia in the offspring of rats exposed to 36 mg DMA/kg/day as DMA on gestational days 6–15. Mice appear to be less sensitive than rats to the developmental toxicity of DMA. No developmental effects were observed in the offspring of mice administered 200 mg DMA/kg/day on gestational days 7–16 (Rogers et al. 1981); at higher doses, decreases in fetal body weight, delays in ossification, and cleft palate were observed (Kavlock et al. 1985; Rogers et al. 1981). In rabbits, a NOAEL of 12 mg DMA/kg/day was identified (Irvine et al. 2006); at 48 mg DMA/kg/day, there were increased maternal deaths and abortions.

3.2.2.7 Cancer

Inorganic Arsenicals. There is convincing evidence from a large number of epidemiological studies and case reports that ingestion of inorganic arsenic increases the risk of developing skin cancer (Alain et al. 1993; Beane Freeman et al. 2004; Bickley and Papa 1989; Cebrián et al. 1983; Chen et al. 2003; Guo et al. 2001a; Haupert et al. 1996; Hsueh et al. 1995; Lewis et al. 1999; Lüchtrath 1983; Mitra et al. 2004; Morris et al. 1974; Piontek et al. 1989; Sommers and McManus 1953; Tay and Seah 1975; Tsai et al. 1998a, 1999; Tseng 1977; Tseng et al. 1968; Zaldívar 1974; Zaldívar et al. 1981). Lesions commonly observed are multiple squamous cell carcinomas, some of which appear to develop from the

hyperkeratotic warts or corns described in Section 3.2.2.2. In addition, multiple basal cell carcinomas may occur, typically arising from cells not associated with hyperkeratinization. In most cases, skin cancer develops only after prolonged exposure, but one study has reported skin cancer in people exposed for <1 year (Reymann et al. 1978). Although both types of skin cancer can be removed surgically, they may develop into painful lesions that may be fatal if left untreated (Shannon and Strayer 1989).

A number of studies that identify CELs in exposed humans are summarized in Table 3-3 and shown in Figure 3-3. The EPA reviewed the studies that provided dose-response data on the risk of skin cancer (EPA 1988d) and concluded that the most useful study for the purposes of quantitative risk assessment was the ecologic epidemiology study by Tseng et al. (1968). In this study, the incidence of skin cancer was measured as a function of exposure level in over 40,000 people residing in 37 villages in Taiwan, and compared to a control group of over 7,500 people. Beyond the very large sample size, other strengths of this study include excellent case ascertainment (physical examination), inclusion of both males and females, and lifetime exposure duration. Weaknesses and uncertainties include poor nutritional status of the exposed populations, their genetic susceptibility, their exposure to inorganic arsenic from nonwater sources, and the applicability of extrapolating data from Taiwanese to the U.S. population because of different background rates of cancer, possibly genetically determined, and differences in diet other than arsenic (e.g., low protein and fat and high carbohydrate) (EPA 1988d). Because of a lack of information on the amount of individual exposure, subjects were classified into three exposure groups (i.e., high, medium, and low). Based upon pooled data for skin cancer incidence and average well concentrations for each village in the Tseng et al. (1968) study, the EPA calculated a unit risk (the upper-bound excess cancer risk from lifetime exposure to water containing 1 µg As/L) of $5x10^{-5}$ (IRIS 2007). The average daily doses (expressed as mg As/kg/day) that correspond to excess cancer risks of $1x10^{-4}$–$1x10^{-7}$ are shown in Figure 3-3.

The use of a cancer risk estimate derived from the Tseng et al. (1968) study for a U.S. population has been the source of intense debate. Some have argued and have provided data in support of the view that there is persuasive evidence that inorganic arsenic is a cause of human cancer at several sites (i.e., Smith et al. 1992, 1995, 2002). On the other hand, a number of concerns have been raised regarding the strength, or lack of strength, of the database, including: the adequacy of the model used by EPA and the accuracy and reliability of the exposure data (Brown et al. 1997a, 1997b); a number of host and environmental factors among the Taiwanese not applicable elsewhere (Carlson-Lynch et al. 1994); a possible threshold for arsenic carcinogenicity and nonlinearities in the dose-response curve (Abernathy et al. 1996; Slayton et al. 1996); differences in health and nutrition between Taiwan and the United States

that might increase cancer risk in Taiwan (Beck et al. 1995); the possibility that lower doses of arsenic may be beneficial role in some physiological processes (EPA 1988d; FNB/IOM 2001; NRC 1999, 2001); and the possibility of significant exposure to arsenic from sources other than the well water (Chappell et al. 1997). Many of these factors were recognized by EPA (1988d). A report by NRC (2001) suggested that the risks calculated based on increases in incidence of lung and bladder cancers may be greater than those calculated by the EPA based on incidences of skin cancer.

Several early epidemiological studies performed in the United States did not report an increased frequency of skin cancer in small populations consuming water containing arsenic at levels of around 0.1–0.2 ppm (EPA 1981b; Goldsmith et al. 1972; Harrington et al. 1978; Morton et al. 1976). These early data suggested that arsenic-associated skin cancer is not a common problem in this country, but these studies lacked sufficient statistical power to detect small increases in skin cancer incidence that might have occurred at these low doses (EPA 1983g). Later studies in exposed U.S. populations from Utah (Lewis et al. 1999) and Iowa (Beane Freeman et al. 2004) have suggested that arsenic-exposed individuals within the United States may have increased incidence or risk of mortality from some skin cancers, melanoma in particular; however, exposure data from these studies are generally insufficient for dose-response analysis. Another study found a suggestion of an arsenic-induced effect on the development of skin cancer, but the association did not achieve statistical significance (Karagas et al. 2001). Therefore, the risk of arsenic-induced skin cancers in U.S. populations, while it may appear to be less than in some other evaluated populations, may be the reflection that, in most studies, exposures were lower.

In addition to the risk of skin cancer, there is mounting evidence that ingestion of arsenic may increase the risks of internal cancers as well. Many case studies have noted the occurrence of internal tumors of the liver and other tissues in patients with arsenic-induced skin cancer (Falk et al. 1981b; Kasper et al. 1984; Koh et al. 1989; Lander et al. 1975; Regelson et al. 1968; Sommers and McManus 1953; Tay and Seah 1975; Zaldívar et al. 1981). These studies are supported by large-scale epidemiological studies, where associations and/or dose response trends have been detected for tumors of the bladder, kidney, liver, lung, and prostate (Chen and Wang 1990; Chen et al. 1985, 1986, 1988a, 1988b, 1992; Chiou et al. 1995; Cuzick et al. 1992; Ferreccio et al. 1998; Guo et al. 1997; Hopenhayn-Rich et al. 1998; Kurttio et al. 1999; Lewis et al. 1999; Moore et al. 2002; Rivara et al. 1997; Smith et al. 1998; Tsuda et al. 1995a; Wu et al. 1989). The EPA has not yet calculated a unit risk value or slope factor for arsenic-induced internal tumors.

There is increasingly convincing evidence that long-term exposure to arsenic can result in the development of bladder cancer (Bates et al. 2004; Chen et al. 1992, 2003; Chiou et al. 1995, 2001; Cuzick et al. 1992; Guo et al. 2001b; Karagas et al. 2004; Lamm et al. 2004; Michaud et al. 2004; Steinmaus et al. 2003), with transitional cell cancers being the most prevalent. Chiou et al. (1995) reported a dose-response relationship between long-term arsenic exposure from drinking artesian well water and the incidence of lung cancer, bladder cancer, and cancers of all sites combined (after adjustment for age, sex, and cigarette smoking) in four townships in Taiwan exposed to inorganic arsenic in drinking water (0–1.14 mg/L). In a later followup study of the same cohort, the increase in bladder cancer was found to be statistically significant only in subjects exposed for 40 years or longer (Chiou et al. 2001). Cuzick et al. (1992) evaluated a cohort treated with Fowler's solution (potassium arsenite) in Lancashire, England, during the period 1945–1969 and followed through 1991; the cohort of 478 patients showed a significant excess of bladder cancer, but no excess for other causes of death. Of a subcohort of 142 patients examined for signs of arsenicism around 1970 (Cuzick et al. 1992), all 11 subsequent cancer deaths occurred in those with signs of arsenicism (p=0.0009). Hopenhayn-Rich et al. (1996a) investigated bladder cancer mortality for the years 1986–1991 in the 26 counties of Cordoba, Argentina, and reported that bladder cancer SMRs were consistently higher in counties with documented arsenic exposure; a later case-control study by the same authors (Bates et al. 2004) did not report statistically significant increases in bladder cancers resulting from arsenic exposure, except in individuals exposed for 50 years or longer. Guo et al. (2001a) reported significantly increased rate differences for bladder cancer in men and women in Taiwan exposed to 0.64 mg arsenic/L in the drinking water, but not at lower exposure levels. The arsenic-induced bladder tumors do not appear to be histologically different than similar bladder tumor types of nonarsenic origin (Chow et al. 1997), although they tended to be more pronounced. In contrast, Michaud et al. (2004) reported no correlation between arsenic levels in toenails and the incidence of bladder cancers in Finnish workers. Among evaluated U.S. cohorts, there has generally been no association between arsenic exposure (~60–100 µg As/L) and the incidence of mortality from bladder cancers (Lamm et al. 2004; Steinmaus et al. 2003), although it is possible that smoking may render individuals more susceptible to arsenic-induced bladder tumors (Karagas et al. 2004; Steinmaus et al. 2003).

Studies have also suggested that chronic oral exposure to arsenic may result in the development of respiratory tumors and increased incidence of lung cancer (Ferreccio et al. 2000; Guo 2004; Nakadaira et al. 2002; Smith et al. 1998; Viren and Silvers 1999). A study of arsenic-exposed individuals in northern Chile reported significantly increased odds ratios for lung cancer among subjects with ≥30 µg As/L of drinking water (Ferreccio et al. 2000), although when adjusted for socioeconomic status, smoking, and

other factors, the increase was only significant at 60 µg As/L or greater. Guo (2004) reported significantly increased rates differences (RD) for lung cancer for Taiwanese men and women exposed to 0.64 mg As/L or greater, with those subjects >50 years of age being particularly at risk. Nakadaira et al. (2002) suggested that even comparatively short exposure durations (≤5 years) may be sufficient for the development of arsenic-induced lung cancer.

Studies in U.S. populations exposed to arsenic in drinking water (EPA 1981b; Lamm et al. 2004; Lewis et al. 1999; Morton et al. 1976; Steinmaus et al. 2003; Valentine et al. 1992) have not yielded the cancer incidences and health effects noted in Taiwan, Mexico, and Chile. Whether this difference is due to a smaller population of subjects compared to Taiwan, to overall lower doses in exposed U.S. populations, or to differences in nutritional or socioeconomic conditions has not been resolved. It should be noted that exposed populations in Mexico and Chile are also smaller than those in Taiwan.

Most studies of animals exposed to arsenate or arsenite by the oral route have not detected any clear evidence for an increased incidence of skin cancer or other cancers (Byron et al. 1967; Kroes et al. 1974; Schroeder et al. 1968). Arsenic has sometimes been called a "paradoxical" human carcinogen because of this lack of animal data (Jager and Ostrosky-Wegman 1997). The basis for the lack of tumorigenicity in animals is not known, but could be related to species-specific differences in arsenic distribution, and induction of cell proliferation (Byrd et al. 1996) (see Section 3.5). As discussed in Section 3.5 below, the carcinogenic effects of arsenic may partially result from its function as a cocarcinogen, which would not manifest in most animal carcinogenicity studies.

One mouse study using transgenic mice (which carry the v-Ha-ras oncogene) administered 48 mg As/kg/day as sodium arsenite in drinking water for 4 weeks followed by dermal application of 12-O-tetradecanoylphorbol-13-acetage (TPA) to shaved back skin twice a day for 2 weeks showed an increase in the incidence of skin papillomas when compared to transgenic mice receiving only TPA treatment, only arsenic, or to wild-type mice receiving both TPA and arsenic (Germolec et al. 1998); arsenic treatment alone did not result in increased papilloma incidence. Increases in mRNA transcripts for the growth factors transforming growth factor-α (TGF-α) and granulocyte/ macrophage-colony stimulating factor (GM-CSF) were detected in the epidermis of the arsenic-treated mice.

A few studies in mice have noted that arsenic ingestion may actually decrease the incidence of some tumor types. For example, arsenic exposure caused decreased incidence of urethane-induced pulmonary tumors (Blakley 1987), spontaneous mammary tumors (Schrauzer and Ishmael 1974; Schrauzer et al.

1976), and tumors resulting from injection of mouse sarcoma cells (Kerkvliet et al. 1980). However, arsenic also increased the growth rate of the tumors that did occur, resulting in a net decrease in survival time in tumor-bearing animals (Kerkvliet et al. 1980; Schrauzer and Ishmael 1974). These observations suggest that arsenic may affect different types of neoplastic cells differently, perhaps acting mainly as a tumor promoter (Schrauzer and Ishmael 1974; Shirachi et al. 1983), although some studies have suggested that arsenic's actions are not consistent with tumor promotion (Baroni et al. 1963; Boutwell 1963).

There is evidence suggesting that inorganic arsenic can induce cancer in the offspring from mice exposed to arsenic during gestation (transplacental carcinogen) (Waalkes et al. 2003, 2004a, 2004b, 2004c, 2006a, 2006b). These studies are summarized in Section 3.2.2.6, Developmental Effects.

Organic Arsenicals. No studies were located regarding cancer in humans after oral exposure to organic arsenicals. Two lifetime carcinogenicity studies with MMA did not find significant increases in tumors in rats exposed to 72.4 mg MMA/kg/day in the diet for 2 years (Arnold et al. 2003) or 8.4 mg MMA/kg/day in drinking water for 2 years (Shen et al. 2003). No significant increases in neoplastic lesions were observed in mice exposed to 67.1 mg MMA/kg/day in the diet for 2 years (Arnold et al. 2003).

In contrast, significant increases in the incidence of urinary bladder tumors have been observed in rats exposed for 2 years to 7.8 mg DMA/kg/day in the diet (Arnold et al. 2006) or 3.4 mg DMA/kg/day in drinking water (Wei et al. 1999, 2002). The incidence of bladder tumors was similar to controls in the rats exposed to 0.77 mg DMA/kg/day (Arnold et al. 2006) or 0.75 mg DMA/kg/day (Wei et al. 1999, 2002). Neither study reported significant increases in the incidence of neoplastic lesions in other tissues. Arnold et al. (2006) did not find increases in the incidence of neoplastic lesions in mice exposed to doses as high as 94 mg DMA/kg/day in the diet for 2 years. Hayashi et al. (1998) reported that exposure of A/J mice (a strain susceptible to lung tumorigenesis) to 10.4 mg DMA/kg/day (but not 1.3 or 5.2 mg DMA/kg/day) in drinking water for 50 weeks resulted in an increased incidence of papillary adenomas and/or adenocarcinomas and an increased number of lung tumors per mouse.

The incidence of basophilic foci (believed to be a precancerous lesion) in the liver of rats initiated with diethylnitrosamine was increased by subsequent 6-month drinking water exposure to 11 mg DMA/kg/day, suggesting that this compound could act as a cancer promoter (Johansen et al. 1984). Additional evidence for the possible role of DMA as a promoter comes from Yamamoto et al. (1995), who reported that 15 or 60 mg DMA/kg/day in the drinking water for 24 weeks significantly enhanced the tumor induction in the

urinary bladder, kidney, liver, and thyroid gland in male F344 rats treated with a series of initiators. Wanibuchi et al. (1996) reported that treatment of F344 rats for 32 weeks with up to 14.3 mg DMA/kg/day DMA in the drinking water did not result in increased incidences of urinary bladder papillomas or carcinomas, but that incidence of these tumors was elevated if the animals were first pretreated with an initiating compound (BBN). A later study by Li et al. (1998) reported that NBR rats (which do not synthesize $\alpha_{2\mu}$-globulin) exposed to an initiator for 4 weeks followed by DMA for 32 weeks, similar to the Wanibuchi et al. (1996) study, showed a statistically significant increase in simple hyperplasia and papillary or nodular hyperplasia of the bladder. A study by Salim et al. (2003) suggested that DMA primarily exerts its carcinogenic effects on spontaneous tumor development.

No increases in tumor incidence were observed in rats, mice, or dogs exposed to 10, 13, or 5 mg/kg/day roxarsone, respectively, in the diet for 2 years (Prier et al. 1963). Similarly, no evidence of carcinogenicity was observed in female rats or male or female mice exposed to 4 or 43 mg/kg/day as roxarsone in the diet for 2 years (NTP 1989b). However, a slight increase in pancreatic tumors was noted in male rats exposed to 4 mg/kg/day (NTP 1989b); this was considered to constitute equivocal evidence of carcinogenicity.

3.2.3 Dermal Exposure

Adverse effects from dermal exposure to inorganic or organic arsenicals have not been extensively investigated. Table 3-7 summarizes studies in animals and humans that provide quantitative data on dermal exposure-effect relationships for inorganic arsenicals. No quantitative data on dermal exposure to organic arsenicals were located. Available quantitative and qualitative data are discussed in greater detail below.

3.2.3.1 Death

Inorganic Arsenicals. No studies were located regarding death in humans after dermal exposure to inorganic arsenicals. In rats, no deaths resulted from dermal exposure to arsenate or arsenite at doses up to 1,000 mg As/kg (Gaines 1960). These data indicate that dermal exposure to inorganic arsenic compounds is very unlikely to result in death.

Organic Arsenicals. No studies were located regarding death in humans after dermal exposure to organic arsenicals. No deaths were observed in rabbits receiving daily dermal applications of 540 mg As/kg as

Table 3-7 Levels of Significant Exposure to Inorganic Arsenic - Dermal

Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL	LOAEL Less Serious	LOAEL Serious	Reference Chemical Form	Comments
ACUTE EXPOSURE							
Immuno/ Lymphoret							
Gn Pig (Hartley)	once		580 mg/L			Wahlberg and Boman 1986 As(+3)	
Gn Pig (Hartley)	once		4000 mg/L			Wahlberg and Boman 1986 As(+5)	
INTERMEDIATE EXPOSURE							
Systemic							
Mouse (Rockland)	30 wk 11 x/wk	Dermal		6 F mg/kg/day (gross hyperplasia, ulceration)		Boutwell 1963 As(+3)	

F = female; Gn pig = guinea pig; Immuno/Lymphoret = immunological/lymphoreticular; wk = week(s); x = time(s)

MMA 5 days/week for 21 days (Margitich and Ackerman 1991b) or 1,000 mg DMA/kg/day 5 days/week for 21 days (Margitich and Ackerman 1991a).

3.2.3.2 Systemic Effects

No studies were located that have associated respiratory, cardiovascular, gastrointestinal, hematological, musculoskeletal, hepatic, renal, endocrine, ocular, or body weight effects in humans or animals with dermal exposure to inorganic arsenicals.

Respiratory Effects. No studies were located regarding respiratory effects in humans after dermal exposure to organic arsenicals. No histological effects were observed in the respiratory tracts of rabbits following dermal application of 1,000 mg/kg/day MMA or DMA 5 days/week for 21 days (Margitich and Ackerman 1991a, 1991b).

Cardiovascular Effects. No studies were located regarding cardiovascular effects in humans after dermal exposure to organic arsenicals. No histological effects were observed in the hearts of rabbits following dermal application of 1,000 mg/kg/day MMA or DMA 5 days/week for 21 days (Margitich and Ackerman 1991a, 1991b).

Hematological Effects. No studies were located regarding hematological effects in humans after dermal exposure to organic arsenicals. No treatment-related hematological alterations were observed in rabbits receiving dermal applications of 1,000 mg MMA/kg/day (Margitich and Ackerman 1991a) or 1,000 mg DMA/kg/day 5 days/week for 21 days (Margitich and Ackerman 1991b).

Hepatic Effects. No studies were located regarding hepatic effects in humans after dermal exposure to organic arsenicals. No significant alterations in blood clinical chemistry, liver weights, or histopathology were observed in rabbits dermally exposed to 1,000 mg/kg/day MMA or DMA 5 days/week for 21 days (Margitich and Ackerman 1991a, 1991b).

Renal Effects. No studies were located regarding renal effects in humans after dermal exposure to organic arsenicals. No significant alterations in urinalysis, kidney weights, or histopathology were observed in rabbits following dermal exposure to 1,000 mg MMA/kg/day (Margitich and Ackerman 1991a) or 1,000 mg DMA/kg/day 5 days/week for 21 days (Margitich and Ackerman 1991b).

Endocrine Effects. No studies were located regarding endocrine effects in humans after dermal exposure to organic arsenicals. No alterations in adrenal gland weight or histopathology of the adrenal glands, pancreas, pituitary gland, thyroid gland, and parathyroid gland were observed in rabbits following dermal application of 1,000 mg/kg/day MMA or DMA 5 days/week for 21 days (Margitich and Ackerman 1991a, 1991b).

Dermal Effects.

Inorganic Arsenicals. Several studies of humans exposed to arsenic dusts in the workplace have reported that inorganic arsenic (usually arsenic trioxide) can cause contact dermatitis (Holmqvist 1951; Pinto and McGill 1953). Typical responses included erythema and swelling, with papules and vesicles in more severe cases (Holmqvist 1951). The dermal contact rates that cause these effects in humans have not been quantified, but a similar direct irritation of the skin has been noted in mice exposed to 4 mg As/kg/day as potassium arsenite for 30 weeks (Boutwell 1963). In contrast, no significant dermal irritation was noted in guinea pigs exposed to aqueous solutions containing 4,000 mg As/L as arsenate or 580 mg As/L as arsenite (Wahlberg and Boman 1986). These studies indicate that direct contact may be of concern at high exposure levels, but do not suggest that lower levels are likely to cause significant irritation.

Studies on possible dermal sensitization by inorganic arsenicals are discussed in Section 3.2.3.3 below.

Organic Arsenicals. Contact dermatitis was reported in workers involved in the application of an organic arsenical herbicide, which is a mixture of DMA and its sodium salt (Peoples et al. 1979).

Application of an unspecified amount of MMA to the skin of rabbits was reported to result in mild dermal irritation in a Draize test (Jaghabir et al. 1988). No dermal irritation was reported in rabbits repeatedly exposed to 1,000 mg MMA/kg/day (Margitich and Ackerman 1991a) or 1,000 mg DMA/kg/day 5 days/week for 21 days (Margitich and Ackerman 1991b).

Ocular Effects. No studies were located regarding ocular effects in humans or animals after dermal exposure to organic arsenicals.

Body Weight Effects. No studies were located regarding body weight effects in humans after dermal exposure to organic arsenicals. No significant alterations in body weight gain were observed in rabbits

following a 5 day/week exposure to 1,000 mg/kg/day MMA or DMA for 21 days (Margitich and Ackerman 1991a, 1991b).

3.2.3.3 Immunological and Lymphoreticular Effects

Inorganic Arsenicals. Examination of workers exposed to arsenic trioxide dusts in a copper smelter led Holmqvist (1951) to suspect that repeated dermal contact could lead to dermal sensitization. In support of this, Holmqvist (1951) found a positive patch test in 80% of the exposed workers compared to 30% in a control population. These data do suggest that workers may be sensitized to arsenic, but the high response rate in controls seems unusual. A much lower response rate (0.5%) was noted in another patch test study of dermal sensitization (Wahlberg and Boman 1986), and the few positive responses seemed to be due to a cross-reactivity with nickel. Mohamed (1998) evaluated 11 male workers at a tin smelting factory where arsenic trioxide levels ranged from 5.2 to 14.4 mg/m^3. The workers experienced symptoms of generalized itch, dry and hyperpigmented skin, folliculitis, and superficial ulcerations. The authors concluded that arsenic-containing dust collected on the sweat on the workers' skin, causing contact dermatitis. Studies in guinea pigs did not yield evidence of a sensitization reaction to inorganic arsenic (Wahlberg and Boman 1986).

Organic Arsenicals. Support for sensitization to DMA is provided in a case report of a 26-year-old woman who was occupationally exposed to DMA and experienced eczema on her face (Bourrain et al. 1998). Patch testing confirmed an allergic reaction to DMA, and avoidance of DMA resulted in disappearance of the symptoms. No studies were located regarding immunological or lymphoreticular effects in animals after dermal exposure to organic arsenicals.

No studies were located that have associated any of the following effects in humans or animals with dermal exposure to inorganic or organic arsenicals:

3.2.3.4 Neurological Effects

3.2.3.5 Reproductive Effects

3.2.3.6 Developmental Effects

3.2.3.7 Cancer

Inorganic Arsenicals. No studies were found that have associated cancer in humans with dermal exposure to arsenic. Application of arsenic acid to the skin of mice pretreated with dimethylbenz-

anthracene did not result in any skin tumors (Kurokawa et al. 1989), suggesting that arsenic does not act as a promoter in this test system.

Organic Arsenicals. No studies were located regarding cancer in humans or animals after dermal exposure to organic arsenicals.

3.3 GENOTOXICITY

Inorganic Arsenicals. There have been a large number of studies of the genotoxic effects of arsenic. Tables 3-8 and 3-9 summarize a number of reports on the *in vitro* and *in vivo* genotoxicity of inorganic arsenicals, respectively. In general, *in vitro* studies in prokaryotic organisms have been negative for gene mutations (Lantzsch and Gebel 1997; Löfroth and Ames 1978; Nishioka 1975; Rossman et al. 1980; Ulitzur and Barak 1988). Studies in human fibroblasts, lymphocytes, and leukocytes, mouse lymphoma cells, Chinese hamster ovary cells, and Syrian hamster embryo cells demonstrate that *in vitro* arsenic exposure can induce chromosomal aberrations and sister chromatid exchange (see Table 3-8 for citations). *I vitro* studies in human, mouse, and hamster cells have also been positive for DNA damage and repair and enhancement or inhibition of DNA synthesis.

Studies of humans have detected a higher-than-average incidence of chromosomal aberrations in peripheral lymphocytes, both after inhalation exposure (Beckman et al. 1977; Nordenson et al. 1978) and oral exposure (Burgdorf et al. 1977; Nordenson et al. 1979). These studies must be interpreted with caution, since in most cases, there were only a small number of subjects and a number of other chemical exposures were possible (EPA 1984a). Human and animal data are available indicating that inhaled inorganic arsenic is clastogenic. Workers exposed to unspecified concentrations of arsenic trioxide at the Ronnskar copper smelter in Sweden were found to have a significant increase in the frequency of chromosomal aberrations in peripheral lymphocytes (Beckman et al. 1977; Nordenson et al. 1978). This result is supported by an animal study that found increased chromosomal aberrations in the livers of fetuses from pregnant mice exposed to 22, but not 2.2 or 0.20, mg As/m^3 as arsenic trioxide on days 9–12 of gestation (Nagymajtényi et al. 1985). Workers in the arsenic-based glass making industry in southern India had a significantly increased frequency of micronuclei in buccal cells and increased DNA damage in leukocytes compared to a control group (Vuyyuri et al. 2006). Exposure levels were not available, but the concentration of arsenic in the blood from workers was approximately 5 times higher than in the reference group.

Table 3-8. Genotoxicity of Inorganic Arsenic *In Vitro*

Valence	Species (test system)	End point	Results		Reference
			With activation	Without activation	
Prokaryotic organisms:					
As^{+3}	*Escherichia coli*	Reverse mutation	No data	+	Nishioka 1975
As^{+3}	*E. coli* PQ37	Gene mutation	No data	–	Lantzsch and Gebel 1997
As^{+3}	*E. coli* (six strains)	Reverse mutation	No data	–	Rossman et al. 1980
As^{+3}	*Salmonella typhimurium*	Gene mutation	No data	–	Löfroth and Ames 1978
As^{+3}	*Photobacterium fischeri*	Gene mutation	No data	–	Ulitzur and Barak 1988
As^{+5}	*S. typhimurium*	Gene mutation	No data	–	Löfroth and Ames 1978
As^{+5}	*P. fischeri*	Gene mutation	No data	+	Ulitzur and Barak 1988
Eukaryotic organisms:					
	Fungi:				
As^{+3}; As^{+5}	*Saccharomyces cerevisiae*	Gene mutation	No data	–	Singh 1983
	Mammalian cells:				
As^{+3}	Human fibroblasts	DNA repair inhibition	No data	+	Okui and Fujiwara 1986
As^{+3}	Human fibroblasts	DNA repair and mutant frequencies	+	+	Wiencke et al. 1997
As^{+3}	Human fibroblasts	DNA repair inhibition	+	+	Hartwig et al. 1997
As^{+3}	Human fibroblasts (MRC5CV1)	DNA migration	No data	+	Hartmann and Speit 1996
As^{+3}	Human fibroblasts (HFW cells)	Cytotoxicity	No data	+	Lee and Ho 1994
As^{+3}	Human skin fibroblasts (HFW)	Chromosome endoreduplication	No data	+	Huang et al. 1995
As^{+3}	Human skin fibroblasts	Chromosomal aberrations	No data	+	Yih et al. 1997
As^{+3}	Human fetal lung fibroblasts	DNA strand breaks	No data	+	Dong and Luo 1993
As^{+3}	Human fetal lung fibroblasts (2BS cells)	DNA damage and repair	No data	+	Dong and Luo 1994
As^{+3}; As^{+5}	Human umbilical cord fibroblasts	Chromosomal aberrations	No data	+	Oya-Ohta et al. 1996
As^{+3}	Diploid human fibroblasts	Morphological transformation	No data	+	Landolph 1994

Table 3-8. Genotoxicity of Inorganic Arsenic *In Vitro*

Valence	Species (test system)	End point	Results		Reference
			With activation	Without activation	
As^{+3}	Human leukocytes	Chromosomal aberration	No data	+	Nakamuro and Sayato 1981
As^{+3}	Human lymphocytes	DNA protein cross-links	–	–	Costa et al. 1997
As^{+3}; As^{+5}	Human lymphocytes	Enhancement or inhibition on DNA synthesis	No data	+	Meng 1993a
As^{+3}; As^{+5}	Human lymphocytes	Enhancement or inhibition on DNA synthesis	No data	+	Meng 1993b
As^{+3}; As^{+5}	Human lymphocytes	Enhancement or inhibition on DNA synthesis	No data	+	Meng 1994
As^{+3}	Human lymphocytes	Hyperdiploidy and chromosomal breakage	No data	(+)	Rupa et al. 1997
As^{+3}	Human lymphocytes	Hyperdiploid nuclei	No data	+	Ramirez et al. 1997
As^{+3}	Human lymphocytes	Chromosomal aberration	No data	+	Beckman and Nordenson 1986
As^{+3}	Human lymphocytes	Chromosomal aberrations and sister chromatid exchange	No data	+	Nordenson et al. 1981
As^{+3}	Human lymphocytes	Chromosomal aberration	No data	+	Sweins 1983
As^{+3}	Human lymphocytes	Chromosomal aberrations	No data	+	Yager and Wiencke 1993
As^{+3}	Human lymphocytes	Chromosomal aberrations	No data	+	Vega et al. 1995
As^{+3}	Human lymphocytes	Chromosomal aberrations	No data	+	Wan et al. 1982
As^{+3}	Human lymphocytes	Chromosomal aberrations and sister chromatic exchange	No data	+	Wiencke and Yager 1992
As^{+3}	Human lymphocytes	Chromosome aberrations and sister chromatid exchanges	No data	+	Larramendy et al. 1981
As^{+3}	Human lymphocytes	Sister chromatid exchange	No data	+	Gebel et al. 1997
As^{+5}	Human lymphocytes	Sister chromatid exchange	No data	–	Gebel et al. 1997
As^{+3}	Human lymphocytes	Sister chromatid exchange	No data	+	Hartmann and Speit 1994
As^{+3}	Human lymphocytes	Sister chromatid exchange	No data	+	Jha et al. 1992
As^{+3}	Human lymphocytes	Sister chromatid exchange	No data	+	Rasmussen and Menzel 1997

Table 3-8. Genotoxicity of Inorganic Arsenic *In Vitro*

Valence	Species (test system)	End point	Results		Reference
			With activation	Without activation	
As^{+3}; As^{+5}	Human T-cell lymphoma-derived cell line (Molt-3)	PARP activity inhibition	No data	+	Yager and Wiencke 1997
As^{+3}	Human cervix carcinoma HeLa and cisplatin-resistant HeLa/CPR variant cells	DNA repair modification	+	+	Chao 1996
As^{+3}	Human cervix carcinoma cells (HeLa)	DNA damage recognition	No data	-	Hartwig et al. 1998
As^{+3}	Human osteosarcoma cells (HOS)	DNA repair	No data	+	Hu et al. 1998
As^{+3}	Human osteosarcoma cells (HOS)	Cell transformation	No data	+	Mure et al. 2003
As^{+3}	Human-hamster hybrid A_1 cells	DNA adducts	No data	+	Kessel et al. 2002
As^{+3}	Mouse lymphoma cells	Enhanced viral forward mutation	No data	(+)	Oberly et al. 1982
As^{+3}; As^{+5}	Mouse lymphoma cells [L5178Y/$TK^{+/-}$ (-3.7.2C)]	Chromosomal mutations	No data	+	Moore et al. 1997a
As^{+3}	Mouse lymphoma cells [L5178Y $tk^{+/-}$ (3.7.sc)]	Mutagenicity	No data	+	Oberly et al. 1996
As^{+3}; As^{+5}	Mouse lymphoma cells	Chromosomal aberrations	No data	+	Moore et al. 1994a
As^{+3}	Mouse lymphoma cells	Chromosomal aberrations	No data	+	Sofuni et al. 1996
As^{+3}	Mouse 3T6 cells	Gene amplification	No data	+	Lee et al. 1988
As^{+3}	Mouse embryo fibroblasts (C3H/10T/2 Cl8)	Morphological transformation	No data	+	Landolph 1994
As^{+3}	Chinese hamster V79 cells	Gene mutation	No data	–	Li and Rossman 1991
As^{+3}	Chinese hamster V79 cells	Gene mutation	No data	–	Rossman et al. 1980
As^{+3}	Chinese hamster V79 cells	DNA damage, DNA-protein cross-linking, micronucleus induction	No data	+	Gebel et al. 1998a
As^{+3}	Chinese hamster V79 cells	DNA repair and mutant frequencies	No data	+	Li and Rossman 1991
As^{+3}	Chinese hamster V79 cells	Intrachromosomal homologous recombination	No data	+	Helleday et al. 2000
As^{+3}	Chinese hamster ovary cells (CHO-AL)	Gene mutation	No data	+	Hei et al. 1998
As^{+3}	Chinese hamster ovary cells (CHO-AS52)	Mutagenicity	No data	+	Meng and Hsie 1996

Table 3-8. Genotoxicity of Inorganic Arsenic *In Vitro*

Valence	Species (test system)	End point	Results		Reference
			With activation	Without activation	
As^{+3}	Chinese hamster ovary cells	Gene mutation	No data	+	Yang et al. 1992
As^{+3}	Chinese hamster ovary cells	DNA repair inhibition	No data	+	Lee-Chen et al. 1993
As^{+3}	Chinese hamster ovary cells	DNA repair inhibition	No data	–	Lee-Chen et al. 1992
As^{+3}	Chinese hamster ovary cells (CHO-K1)	DNA strand breaks	+	+	Lee-Chen et al. 1994
As^{+3}	Chinese hamster ovary cells (CHO-K1)	DNA strand breaks	No data	+	Lynn et al. 1997
As^{+3}	Chinese hamster ovary cells	Aberrant metaphases	No data	+	Jan et al. 1986
As^{+3}	Chinese hamster ovary cells	Aberrant metaphases	No data	+	Lee et al. 1986
As^{+3}	Chinese hamster ovary cells	Chromosomal aberrations	+	+	Huang et al. 1992
As^{+3}	Chinese hamster ovary cells (CHO-K1)	Chromosomal aberrations	No data	+	Huang et al. 1993
As^{+3}, As^{+5}	Chinese hamster ovary cells (CHO-K1)	Chromosomal aberrations and sister chromatid exchange	No data	+	Kochhar et al. 1996
As^{+3}	Chinese hamster ovary cells	Chromosomal aberrations and sister chromatid exchange	+	+	Lin and Tseng 1992
As^{+3}	Chinese hamster ovary cells	Chromosomal aberrations and sister chromatid exchange	No data	+	Wan et al. 1982
As^{+3}	Chinese hamster ovary cells	Sister chromatid exchange and micronucleus induction	No data	+	Fan et al. 1996
As^{+3}	Chinese hamster ovary cells	Cell-killing and micronucleus induction	No data	+	Wang and Huang 1994
As^{+3}	Chinese hamster ovary cells	Micronuclei	No data	+	Liu and Huang 1997
As^{+3}	Chinese hamster ovary cells	Micronuclei formation	No data	+	Yee-Chien and Haimei 1996
As^{+3}	Chinese hamster ovary cells	Micronuclei induction	No data	+	Wang et al. 1997
As^{+3}	Chinese hamster ovary cells	Cytotoxicity	No data	–	Lee and Ho 1994
As^{+3}	Syrian hamster embryo cells	Gene mutation	No data	–	Lee et al. 1985

Table 3-8. Genotoxicity of Inorganic Arsenic *In Vitro*

Valence	Species (test system)	End point	Results		Reference
			With activation	Without activation	
As^{+3}	Syrian hamster embryo cells	Chromosome aberrations and sister chromatid exchanges	No data	+	Larramendy et al. 1981
As^{+3}	Syrian hamster embryo cells	Chromosomal aberration	No data	+	Lee et al. 1985
As^{+3}	Syrian hamster embryo cells	Sister chromatid exchange	No data	+	Lee et al. 1985
As^{+3}	Syrian hamster embryo cells	Micronuclei induction	No data	–	Gibson et al. 1997
As^{+3}	Syrian hamster embryo cells	Micronuclei induction	No data	–	Gibson et al. 1997
As^{+3}	Syrian hamster embryo cells	Morphological transformation	No data	+	Kerckaert et al. 1996
As^{+3}	Syrian hamster embryo cells	Morphological transformation	No data	+	Lee et al. 1985
As^{+3}	Syrian hamster embryo cells	Morphological transformation	No data	+	Casto et al. 1979
As^{+5}	Human fibroblasts	DNA repair inhibition	No data	–	Okui and Fujiwara 1986
As^{+5}	Human leukocytes	Chromosomal aberrations	No data	(+)	Nakamuro and Sayato 1981
As^{+5}	Human lymphocytes	Chromosomal aberrations	No data	–	Nordenson et al. 1981
As^{+5}	Human lymphocytes	Chromosome aberrations and sister chromatid exchanges	No data	+	Larramendy et al. 1981
As^{+5}	Human lymphocytes	Sister chromatid exchange	No data	–	Rasmussen and Menzel 1997
As^{+5}	Human peripheral lymphocytes	Sister chromatid exchange	No data	+	Zanzoni and Jung 1980
As^{+5}	Human keratinocyte line SCC-9 cells	Keratinocyte programming and transcriptional activity	No data	+	Kachinskas et al. 1997
As^{+5}	Mouse lymphoma cells	Gene mutation	No data	–	Amacher and Paillet 1980
As^{+5}	Mouse lymphoma cells	Gene mutation	No data	–	Amacher and Paillet 1980
As^{+5}	Chinese hamster ovary cells	Chromosomal aberrations	No data	+	Wan et al. 1982
As^{+5}	Syrian hamster embryo cells	Gene mutation	No data	–	Lee et al. 1985
As^{+5}	Syrian hamster embryo cells	Chromosome aberrations and sister chromatid exchanges	No data	+	Larramendy et al. 1981

Table 3-8. Genotoxicity of Inorganic Arsenic *In Vitro*

Valence	Species (test system)	End point	Results		Reference
			With activation	Without activation	
As^{+5}	Syrian hamster embryo cells	Chromosomal aberrations	No data	+	Lee et al. 1985
As^{+5}	Syrian hamster embryo cells	Sister chromatid exchange	No data	+	Lee et al. 1985
As^{+5}	Syrian hamster embryo cells	Morphological transformation	No data	+	Lee et al. 1985
As^{+5}	Syrian hamster embryo cells	Morphological transformation	No data	+	DiPaolo and Casto 1979

(+) = weakly positive or marginal result; – = negative result; + = positive result; DNA = deoxyribonucleic acid

Table 3-9. Genotoxicity of Inorganic Arsenic *In Vivo*

Valence	Exposure route	Species (test system)	End point	Results	Reference
Nonmammalian					
As^{+3} As^{+5}	Injection	*Drosophila melanogaster*	Somatic mutations and mitotic recombination	+	Ramos-Morales and Rodriguez-Arnaiz 1995
As^{+3} As^{+5}	Larval feeding	*D. melanogaster*	Somatic mutations and mitotic recombination	+	Ramos-Morales and Rodriguez-Arnaiz 1995
As^{+5}	Larvae	*D. melanogaster*	Mitotic recombinations	+	de la Rosa et al. 1994
Mammalian					
As^{+3}	Inhalation	Human (lymphocytes)	Chromosomal aberrations	–	Beckman et al. 1977
As^{+3}	Inhalation	Human (lymphocytes)	Chromosomal aberrations	+	Nordenson et al. 1978
As^{+3}	Oral	Human (lymphocytes)	Chromosomal aberrations	–	Burgdorf et al. 1977
No data	Oral	Human (lymphocytes)	Chromosomal aberrations	–	Vig et al. 1984
No data	Oral	Human (skin)	DNA adducts	+	Matsui et al. 1999
As^{+3}	Oral	Human (lymphocytes)	Sister chromatid exchange	–	Burgdorf et al. 1977
As^{+3}	Oral	Human (lymphocytes)	Sister chromatid exchange	+	Hsu et al. 1997
No data	Oral	Human (lymphocytes)	Sister chromatid exchange	+	Lerda 1994
No data	Oral	Human (lymphocytes)	Sister chromatid exchange	+	Liou et al. 1999
No data	Oral	Human (lymphocytes)	Sister chromatid exchange	+	Mahata et al. 2003
As^{+3}	Oral	Human (lymphocytes)	Sister chromatid exchange	–	Nordenson et al. 1978
No data	Oral	Human (lymphocytes)	Sister chromatid exchange	–	Vig et al. 1984
No data	Oral	Human skin carcinoma	Mutation and overexpression of p53	+	Hsu et al. 1999
As^{+3}	Oral	Exfoliated human epithelial cells	Micronuclei	+	Moore et al. 1996
As^{+3}	Oral	Exfoliated human epithelial cells	Micronuclei	+	Tian et al. 2001
No data	Oral	Human (bladder cells)	Micronuclei	+	Moore et al. 1995
No data	Oral	Human (lymphocytes)	Micronuclei	+	Martínez et al. 2004
No data	Oral	Human (lymphocytes)	Micronuclei	+	Basu et al. 2004
No data	Oral	Human (oral mucosa cells)	Micronuclei	+	Basu et al. 2004

Table 3-9. Genotoxicity of Inorganic Arsenic *In Vivo*

Valence	Exposure route	Species (test system)	End point	Results	Reference
No data	Oral	Human (urothelial cells)	Micronuclei	+	Basu et al. 2004
As^{+5}	Oral	Rat (bone marrow cells)	Chromosomal aberrations	+	Datta et al. 1986
As^{+3}	Inhalation	Mouse (fetal liver)	Chromosomal aberrations	(+)	Nagymajtényi et al. 1985
As^{+3}	Oral	Mouse (bone marrow cells)	Chromosomal aberrations	+	Das et al. 1993
As^{+3}	Oral	Mouse (bone marrow cells)	Chromosomal aberrations	+	Poddar et al. 2000
As^{+3}	Oral	Mouse (bone marrow cells)	Chromosomal breaks, exchanges	–	Poma et al. 1987
As^{+3}	Oral	Mouse (spermatogonia)	Chromosomal aberrations	–	Poma et al. 1987
As^{+3}	Oral	Mouse (leukocytes)	Chromosomal breaks	+	McDorman et al. 2002
As^{+3}	Intraperitoneal	Mouse (bone marrow cells)	Chromosomal breaks, exchanges	–	Poma et al. 1981
As^{+3}	Intraperitoneal	Mouse (bone marrow cells)	Micronuclei	+	DeKnudt et al. 1986
As^{+3}	Intraperitoneal	Mouse (spermatogonia)	Spermatongonia	–	Poma et al. 1981
As^{+3}	Intraperitoneal	Mouse (spermatogonia)	Sperm morphology	–	DeKnudt et al. 1986
As^{+3}	Intraperitoneal	Mouse (spermatogenesis)	Dominant lethal mutations	–	DeKnudt et al. 1986

(+) = weakly positive or marginal result; – = negative result; + = positive result; DNA = deoxyribonucleic acid

Investigations of genotoxic effects of ingested arsenic have yielded mixed results possibly due to the different types of cells examined and the different exposure levels experienced by the populations studied. A study of p53 mutations in arsenic-related skin cancers from patients in Taiwan exposed to arsenic from drinking water found a high rate of p53 mutations and different types of p53 mutations compared with those seen in UV-induced skin cancers (Hsu et al. 1999); similar results have been found in mice (Salim et al. 2003). In humans exposed to Fowler's solution (potassium arsenite, usually taken at a dose of about 0.3 mg As/kg/day [Holland 1904]), increased sister chromatid exchanges, but no increase in chromosomal aberrations, was reported in one study (Burgdorf et al. 1977), while just the converse (increased aberrations but no increase in sister chromatid exchange) was reported in another (Nordenson et al. 1979). Moore et al. (1997a) reported an exposure-dependent increase in the occurrence of micronucleated cells in epithelial cells from the bladder in a male population in northern Chile chronically exposed to high and low arsenic levels in their drinking water (average concentrations, 600 and 15 µg As/L, respectively), and noted that chromosome breakage was the major cause of micronucleus (MN) formation. Similar results were reported by Martínez et al. (2004) who evaluated micronuclei formation in peripheral lymphocytes from people in northern Chile exposed to up to 0.75 mg As/L in their drinking water. In contrast, Martínez et al. (2005) did not find a significant increase in micronuclei in buccal cells from subjects from the same area relative to a low exposure group. Vig et al. (1984) found no significant differences in the frequency of chromosomal aberrations or sister chromatid exchanges between two populations in Nevada with differing levels of arsenic in their drinking water (mean concentrations of 5 and 109 µg/L). In animal studies, an increased incidence of chromosomal abnormalities was detected in rats given oral doses of sodium arsenate (4 mg As/kg/day) for 2–3 weeks (Datta et al. 1986), but no consistent increase in chromosomal aberrations was detected in bone marrow cells or spermatogonia from mice given sodium arsenite (about 50 mg As/kg/day) for up to 8 weeks (Poma et al. 1987). These studies suggest that ingested arsenic may cause chromosomal effects, but these data are too limited to draw a firm conclusion.

Organic Arsenicals. The genotoxicity of the organic arsenicals has been investigated in a number of studies (see Table 3-10). Several tests indicate that DMA and roxarsone may be able to cause chromosome aberrations, mutations, and deoxyribonucleic acid (DNA) strand breaks; *in vitro* studies with MMA did not find significant increases in the occurrence of chromosome aberrations, forward or reverse mutations, unscheduled DNA synthesis (Chun and Killeen 1989a, 1989b, 1989c, 1989d). An increased number of DNA strand breaks were detected in lung and other tissues of mice and rats given oral doses of ~1,500 mg/kg DMA (Okada and Yamanaka 1994; Yamanaka et al. 1989a); this effect appeared to be related to the formation of some active oxygen species. These breaks were largely repaired within 24 hours, so the relevance with respect to health risk is uncertain.

Table 3-10. Genotoxicity of Organic Arsenic

Chemical form	Species (test system)	End point	Results		Reference
			With activation	Without activation	
Prokaryotic organisms (*in vitro*):					
MMA	*Salmonella typhimurium*	Gene mutation	–	–	Chun and Killeen 1989c
DMA	*Escherichia coli*	Gene mutation	No data	+	Yamanaka et al. 1989b
Roxarsone	*S. typhimurium*	Gene mutation	–	–	NTP 1989b
Eukaryotic organisms (*in vitro*):					
MMA	Chinese hamster ovary cells	Chromosome aberrations	–	–	Chun and Killeen 1989a
MMA	Mouse lymphoma cells ($L5178Y/TK^{+/-}$)	Forward mutation	–	–	Chun and Killeen 1989b
MMA	Rat heptocytes	Unscheduled DNA synthesis	No data	–	Chun and Killeen 1989d
DMA	Human peripheral lymphocytes	Mitogenesis inhibited	No data	–	Endo et al. 1992
DMA	Human lymphocytes	Sister chromatid exchange	No data	–	Rasmussen and Menzel 1997
DMA	Human alveolar (L-132) cells	Lung-specific DNA damage	No data	+	Kato et al. 1994
DMA	Human alveolar type II (L-132) cells	DNA single-strand breaks	+	+	Kawaguchi et al. 1996
DMA	Human diploid L-132 epithelial cells	DNA single-strand breaks	No data	+	Rin et al. 1995
DMA	Human alveolar type II (L-132) cells	DNA strand breaks	No data	+	Tezuka et al. 1993
DMA	Human embryonic cell line of type II alveolar epithelial cells (L-132)	DNA single-strand breaks and DNA-protein crosslinks	No data	+	Yamanaka et al. 1993
DMA	Human alveolar epithelial (L-132) cells	DNA single-strand breaks and DNA-protein crosslinks	No data	+	Yamanaka et al. 1995
DMA	Human pulmonary epithelial (L-132) cells	DNA single-strand breaks	No data	+	Yamanaka et al. 1997
DMA	Human umbilical cord fibroblasts	Chromosomal aberrations	No data	+	Oya-Ohta et al. 1996
DMA	Mouse lymphoma cells ($L5178Y/TK^{+/-}$ -3.7.2C)	Chromosomal mutations	No data	+	Moore et al. 1997a
DMA	Chinese hamster lung and diploid cells (V79)	Mitotic arrest and tetraploid formation	No data	+	Endo et al. 1992

Table 3-10. Genotoxicity of Organic Arsenic

Chemical form	Species (test system)	End point	Results		Reference
			With activation	Without activation	
DMA	Chinese hamster V79 cells	Chromosomal aberrations	No data	+	Ueda et al. 1997
DMA	Chinese hamster lung and diploid cells (V79)	Chromosomal aberrations	No data	+	Kitamura et al. 2002
DMA	Chinese hamster lung and diploid cells (V79)	Chromosomal aberrations	+	+	Kuroda et al. 2004
DMA	Chinese hamster V79 cells	Tetraploids and mitotic arrest	No data	+	Eguchi et al. 1997
MMA	Human umbilical cord fibroblasts	Chromosomal aberrations	No data	+	Oya-Ohta et al. 1996
MMA	Chinese hamster V79 cells	Tetraploids and mitotic arrest	No data	+	Eguchi et al. 1997
Roxarsone	*Drosophila melanogaster*	Sex linked recessive	No data	–	NTP 1989b
Roxarsone	Rat hepatocyte	DNA double-strand breaks	No data	+	Storer et al. 1996
Roxarsone	A31-1-13 clone of BALB/c-3T3 cells	Transformation response and mutagenicity	No data	–	Matthews et al. 1993
Roxarsone	Mouse lymphoma (L5178Y) cells	Trifluorothymidine resistance	No data	+	NTP 1989b
Eukaryotic organisms (*in vivo*):					
DMA	Rat (oral exposure)	DNA single-stand breaks in lung	No data	+	Yamanaka and Okada 1994
DMA	Mouse (oral exposure)	DNA strand breaks in tissues	No data	+	Yamanaka et al. 1989b
DMA	Mouse (oral exposure)	DNA single-stand breaks in lung	No data	+	Yamanaka et al. 1993
DMA	Mouse (oral exposure)	DNA single-strand breaks in lung	No data	–	Yamanaka et al. 1989a
DMA	Mouse (oral exposure)	DNA adduct formation	No data	+	Yamanaka et al. 2001
DMA	Mouse (injection)	Aneuploidy in bone marrow cells	No data	+	Kashiwada et al. 1998

– = negative result; + = positive result; DMA = dimethylarsinic acid; DNA = deoxyribonucleic acid; MMA = monomethylarsonic acid

3.4 TOXICOKINETICS

There is an extensive database on the toxicokinetics of inorganic arsenic. Most studies have been performed in animals, but there are a number of studies in humans as well. These studies reveal the following main points:

- Both arsenate and arsenite are well absorbed by both the oral and inhalation routes. Absorption by the dermal route has not been well characterized, but is low compared to the other routes. Inorganic arsenic in soil is absorbed to a lesser extent than solutions of arsenic salts.
- The rate of absorption of arsenic in highly insoluble forms (e.g., arsenic sulfide, lead arsenate) is much lower than that of more soluble forms via both oral and inhalation routes.
- Once absorbed, arsenites are oxidized to arsenates and methylated. This process may then be repeated to result in dimethylated arsenic metabolites.
- Distribution of arsenic in the rat is quite different from other animal species, suggesting that the rat is probably not an appropriate toxicokinetic model for distribution, metabolism, or excretion of arsenic by humans.
- The As(+3) form undergoes enzymic methylation primarily in the liver to form MMA and DMA. The rate and relative proportion of methylation production varies among species. The rate of methylation varies considerably among tissues.
- Most arsenic is promptly excreted in the urine as a mixture of As(+3), As(+5), MMA, and DMA; DMA is usually the primary form in the urine. Smaller amounts are excreted in feces. Some arsenic may remain bound to tissues, depending inversely on the rate and extent of methylation.

Less information is available for the organic arsenicals. It appears that both MMA and DMA are well absorbed, but are rapidly excreted in the urine and feces. MMA may be methylated to DMA, but neither MMA nor DMA are demethylated to yield inorganic arsenic.

A review of the evidence that supports these conclusions is presented below.

3.4.1 Absorption

3.4.1.1 Inhalation Exposure

Inorganic Arsenicals. Since arsenic exists in air as particulate matter, absorption across the lung involves two processes: deposition of the particles onto the lung surface, and absorption of arsenic from the deposited material. In lung cancer patients exposed to arsenic in cigarette smoke, deposition was

estimated to be about 40% and absorption was 75–85% (Holland et al. 1959). Thus, overall absorption (expressed as a percentage of inhaled arsenic) was about 30–34%. In workers exposed to arsenic trioxide dusts in smelters, the amount of arsenic excreted in the urine (the main route of excretion; see Section 3.4.4) was about 40–60% of the estimated inhaled dose (Pinto et al. 1976; Vahter et al. 1986). Absorption of arsenic trioxide dusts and fumes (assessed by measurement of urinary metabolites) correlated with time weighted average arsenic air concentrations from personal breathing zone air samplers (Offergelt et al. 1992). Correlations were best immediately after a shift and just before the start of the next shift. Although the percent deposition was not measured in these cases, it seems likely that nearly all of the deposited arsenic was absorbed. This conclusion is supported by intratracheal instillation studies in rats and hamsters, where clearance of oxy compounds of arsenic (sodium arsenite, sodium arsenate, arsenic trioxide) from the lung was rapid and nearly complete (60–90% within 1 day) (Marafante and Vahter 1987; Rhoads and Sanders 1985). In contrast, arsenic sulfide and lead arsenate were cleared more slowly (Marafante and Vahter 1987), indicating that the rate of absorption may be lower if the inhaled arsenic is in a highly insoluble form. There are no data to suggest that absorption of inhaled arsenic in children differs from that in adults.

Organic Arsenicals. No studies were located regarding absorption of organic arsenicals in humans or animals after inhalation exposure. However, DMA instilled in the lungs of rats was absorbed very rapidly (half-time of 2.2 minutes) and nearly completely (at least 92%) (Stevens et al. 1977). This indicates that organic arsenicals are likely to be well absorbed by the inhalation route.

3.4.1.2 Oral Exposure

Inorganic Arsenicals. Several studies in humans indicate that arsenates and arsenites are well absorbed across the gastrointestinal tract. The most direct evidence is from a study that evaluated the 6-day elimination of arsenic in healthy humans who were given water from a high-arsenic sampling site (arsenic species not specified) and that reported approximately 95% absorption (Zheng et al. 2002). A similar absorption efficiency can be estimated from measurements of fecal excretion in humans given oral doses of arsenite, where <5% was recovered in the feces (Bettley and O'Shea 1975). This indicates absorption was at least 95%. These results are supported by studies in which urinary excretion in humans was found to account for 55–87% of daily oral intakes of arsenate or arsenite (Buchet et al. 1981b; Crecelius 1977; Kumana et al. 2002; Mappes 1977; Tam et al. 1979b). In contrast, ingestion of arsenic triselenide (As_2Se_3) did not lead to a measurable increase in urinary excretion (Mappes 1977), indicating that

gastrointestinal absorption may be much lower if highly insoluble forms of arsenic are ingested. There are no data to suggest that absorption of arsenic from the gut in children differs from that in adults.

These observations in humans are supported by a number of studies in animals. Fecal excretion of arsenates and arsenites ranged from 2 to 10% in monkeys and mice, with 70% or more appearing in urine (Charbonneau et al. 1978; Roberts et al. 2002; Vahter 1981; Vahter and Norin 1980). Oral absorption of [^{73}As] labeled sodium arsenate in mice was unaffected by dose (0.0005–5 mg/kg) as reflected in percentage of dose excreted in feces over 48 hours (Hughes et al. 1994). Absorption ranged from 82 to 89% at all doses. Gonzalez et al. (1995) found that the percentage of arsenate that was absorbed in rats decreased as the dose increased from 6 to 480 µg, suggesting saturable, zero-order absorption of arsenate in this species. Hamsters appear to absorb somewhat less than humans, monkeys, and mice, since fecal excretion usually ranges from 10 to 40% (Marafante and Vahter 1987; Marafante et al. 1987a; Yamauchi and Yamamura 1985). Rabbits also appear to absorb less arsenate than humans, monkeys, or mice after oral exposure (Freeman et al. 1993). After a gavage dose of 1.95 mg/kg sodium arsenate, 45% of the arsenate was recovered in feces in males and 52% in females. As in humans, when highly insoluble arsenic compounds are administered (arsenic trisulfide, lead arsenate), gastrointestinal absorption is reduced 20–30% (Marafante and Vahter 1987).

Bioavailability of arsenic was measured in rabbits ingesting doses of smelting soils that contained arsenic primarily in the form of sulfides (Freeman et al. 1993). Bioavailability was assessed by comparing the amounts of arsenic that was excreted after ingestion of the soil to that excreted after an intravenous dose of sodium arsenate. The bioavailability of the arsenic in the ingested soil was 24±3.2% and that of sodium arsenate in the gavage dose was 50±5.7%. Approximately 80% of the arsenic from ingested soil was eliminated in the feces compared with 50% of the soluble oral dose and 10% of the injected dose. In another study, rabbits dosed with sodium arsenite (0.8 mg As/kg) had 5 times greater blood arsenic concentrations than rabbits dosed with arsenic-containing soil (2.8 mg As/kg), suggesting a lower bioavailability of the arsenic in soil (Davis et al. 1992).

Studies of the bioavailability of arsenic suggest that absorption of arsenic in ingested dust or soil is likely to be considerably less than absorption of arsenic from ingested salts (Davis et al. 1992, 1996; EPA 1997g; Freeman et al. 1993, 1995; Pascoe et al. 1994; Roberts et al. 2002, 2007; Rodriguez et al. 1999). Oral absorption of arsenic in a group of three female Cynomolgus monkeys from a soluble salt, soil, and household dust was compared with absorption of an intravenous dose of sodium arsenate (Freeman et al. 1995). Mean absolute percentage bioavailability based on urine arsenic excretion was reported at

67.6±2.6% (gavage), 19.2±1.5% (oral dust), and 13.8±3.3% (oral soil). Mean absolute percentage bioavailability based on blood arsenic levels was reported at 91.3±12.4% (gavage), 9.8±4.3% (oral dust), and 10.9±5.2% (oral soil). The arsenic in the dust and soil was approximately 3.5–5-fold (based on levels in the urine) and 8–9-fold (based on levels in the blood) less bioavailable than arsenic in solution. Two other studies in monkeys reported relative bioavailability of arsenic in soil from a number of locations (electrical substation, wood preserving sites, pesticide sites, cattle-dip sites, volcanic soil, and mining sites) ranged from 5 to 31% (Roberts et al. 2002, 2007). A study in beagle dogs fed with soil containing As_2O_5 or treated with intravenous soluble arsenic found that compared to injection the bioavailability of arsenic from ingested soil was 8.3±2.0% (Groen et al. 1993). The bioavailability of arsenic in soil has been studied in juvenile swine that received daily oral doses of soil or sodium arsenate (in food or by gavage) for 15 days (EPA 1997g). The soils were obtained from various mining and smelting sites and contained, in addition to arsenic at concentrations of 100–300 µg/g, lead at concentrations of 3,000–14,000 µg/g. The arsenic doses ranged from 1 to 65.4 µg/kg/day. The fraction of the arsenic dose excreted in urine was measured on days 7 and 14 and the relative bioavailability of the soil-borne arsenic was estimated as the ratio of urinary excretion fractions, soil arsenic:sodium arsenate. The mean relative bioavailability of soil-borne arsenic ranged from 0 to 98% in soils from seven different sites (mean±SD, 45%±32). Estimates for relative bioavailability of arsenic in samples of smelter slag and mine tailings ranged from 7 to 51% (mean±SD, 35%±27). Rodriguez et al. (1999) used a similar approach to estimate the relative bioavailability of arsenic in mine and smelter wastes (soils and solid materials) in juvenile swine. Samples included iron slag deposits and calcine deposits and had arsenic concentrations that ranged from 330 to 17,500 µg/g. Relative bioavailability (waste:sodium arsenate) ranged from 3 to 43% for 13 samples (mean, 21%) and was higher in iron slag wastes (mean, 25%) than in calcine wastes (mean, 13%).

Bioavailability of arsenic from soil is reduced by low solubility and inaccessibility due to the presence of secondary reaction products or insoluble matrix components (Davis et al. 1992). This is supported by studies conducted with *in vitro* simulations of the gastric and/or intestinal fluids (Hamel et al. 1998; Pouschat and Zagury 2006; Rodriguez et al. 1999; Ruby et al. 1996, 1999; Williams et al. 1998). When soils containing arsenic are incubated in simulated gastrointestinal fluids, only a fraction of the arsenic becomes soluble. Estimates of the soluble, or bioaccessible, arsenic fraction have ranged from 3 to 50% for various soils and mining and smelter waste materials (Pouschat and Zagury 2006; Rodriguez et al. 1999; Ruby et al. 1996); these estimates are similar to *in vivo* estimates of the relative bioavailability of arsenic in these same materials (Ruby et al. 1999).

Organic Arsenicals. Based on urinary excretion studies in volunteers, it appears that both MMA and DMA are well absorbed (at least 75–85%) across the gastrointestinal tract (Buchet et al. 1981a; Marafante et al. 1987b). This is supported by studies in animals, where at least 75% absorption has been observed for DMA (Marafante et al. 1987b; Stevens et al. 1977; Vahter et al. 1984; Yamauchi and Yamamura 1984) and MMA (Hughes et al. 2005; Yamauchi et al. 1988). In mice, the relative bioavailability of MMA appears to be dose-dependent; 81% was absorbed following a single gavage dose of 0.4 mg MMA/kg/day compared to 60% following administration of 4 mg MMA/kg/day (Hughes et al. 2005).

3.4.1.3 Dermal Exposure

Inorganic Arsenicals. No quantitative studies were located on absorption of inorganic arsenicals in humans after dermal exposure. Percutaneous absorption of [^{73}As] as arsenic acid (H_3AsO_4) alone and mixed with soil has been measured in skin from cadavers (Wester et al. 1993). Labeled arsenic was applied to skin in diffusion cells and transit through the skin into receptor fluid measured. After 24 hours, 0.93% of the dose passed through the skin and 0.98% remained in the skin after washing. Absorption was lower with [^{73}As] mixed with soil: 0.43% passed through the skin over 24 hours and 0.33% remained in the skin after washing.

Dermal absorption of arsenic has been measured in Rhesus monkeys (Lowney et al. 2005; Wester et al. 1993). After 24 hours, 6.4% of [^{73}As] as arsenic acid was absorbed systemically, as was 4.5% of [^{73}As] mixed with soil (Wester et al. 1993). Similarly, 2.8% of soluble arsenic in water was detected in the urine 24 hours after exposure (Lowney et al. 2005). However, arsenic from soil was poorly absorbed; 0.12% was detected in the urine after 24 hours. Differences between the Wester et al. (1993) and Lowney et al. (2005) studies in terms of uptake from soil may be due to the differences in forms of arsenic in the soil. In the Wester et al. (1993) study, soil was mixed with radiolabelled arsenic acid in water; Lowney et al. (2005) used soil samples from a pesticide manufacturing facility that historically manufactured arsenical pesticides (the arsenic was primarily in the iron oxide and iron silicate mineral phases). Lowney et al. (2005) also measured urinary levels of arsenic following dermal application of CCA residues and found that the levels did not increase from background. Uptake of arsenic into blood or tissues was undetectable for up to 24 hours in rats whose tails were immersed in solutions of sodium arsenate for 1 hour. However, arsenic began to increase in blood, liver, and spleen over the next 5 days (Dutkiewicz 1977). The rate of uptake was estimated to be 1–33 μg/cm^2/hour. These findings suggest that dermal exposure leads initially to arsenic binding to skin, and that the bound arsenic may slowly be taken up into the blood, even after exposure ends.

Organic Arsenicals. No studies were located on absorption of organic arsenicals in humans or animals after dermal exposure.

3.4.2 Distribution

3.4.2.1 Inhalation Exposure

Inorganic Arsenicals. No studies were located on the distribution of arsenic in humans or animals after inhalation exposure, but intratracheal administration of arsenic trioxide to rats resulted in distribution of arsenic to the liver, kidney, skeleton, gastrointestinal tract, and other tissues (Rhoads and Sanders 1985). This is consistent with data from oral and parenteral studies (below), which indicate that absorbed arsenic is distributed throughout the body.

Organic Arsenicals. No studies were located regarding the distribution of organic arsenicals in humans or animals after inhalation exposure. However, DMA administered to rats by the intratracheal route was distributed throughout the body (Stevens et al. 1977), suggesting that inhalation of organic arsenicals would also lead to widespread distribution.

3.4.2.2 Oral Exposure

Inorganic Arsenicals. Analysis of tissues taken at autopsy from people who were exposed to background levels of arsenic in food and water revealed that arsenic is present in all tissues of the body (Liebscher and Smith 1968). Most tissues had about the same concentration level (0.05–0.15 ppm), while levels in hair (0.65 ppm) and nails (0.36 ppm) were somewhat higher. This indicates that there is little tendency for arsenic to accumulate preferentially in any internal organs. However, exposure levels may not have been high enough to cause elevated levels in tissues. Arsenic exposure may have been low enough that the methylation process in the body resulted in limited accumulation in internal organs. Tissue analysis of organs taken from an individual following death from ingestion of 8 g of arsenic trioxide (about 3 g of arsenic) showed a much higher concentration of arsenic in liver (147 μg/g) than in kidney (27 μg/g) or muscle, heart, spleen, pancreas, lungs, or cerebellum (11–12 μg/g) (Benramdane et al. 1999a). Small amounts were also found in other parts of the brain (8 μg/g), skin (3 μg/g), and hemolyzed blood (0.4 μg/g). Many studies have been performed where arsenic levels in hair and nails have been measured and correlations with exposure analyzed. Some of these studies are discussed in Section 3.8, Biomarkers of Exposure.

Inorganic arsenic passes easily through the placenta. High levels of arsenic were found in the liver, kidney, and brain during autopsy of an infant prematurely born to a young mother who had ingested inorganic arsenic at week 30 of gestation (Lugo et al. 1969). Arsenic was detected in human breast milk at concentrations of 0.00013–0.00082 ppm in a World Health Organization study (Somogyi and Beck 1993). Arsenic concentrations were 0.0001–0.0044 ppm in human milk sampled from 88 mothers on the Faroe Islands whose diets were predominantly seafood (Grandjean et al. 1995). Exposures to arsenic from the seafood diet in this population was most likely to organic "fish arsenic." In a population of Andean women exposed to high concentrations (about 200 ppb) of inorganic arsenic in drinking water, concentrations of arsenic in breast milk ranged from about 0.0008 to 0.008 ppm (Concha et al. 1998b).

Studies in mice and hamsters given oral doses of arsenate or arsenite have found elevated levels of arsenic in all tissues examined (Hughes et al. 2003; Vahter and Norin 1980; Yamauchi and Yamamura 1985), including the placenta and fetus of pregnant females (Hood et al. 1987, 1988). Inorganic arsenic crosses the placental barrier and selectively accumulates in the neuroepithelium of the developing animal embryo (Hanlon and Ferm 1977; Lindgren et al. 1984). In mice, radiolabel from orally administered 74-As was widely distributed to all tissues, with the highest levels in skin, kidney, and liver (Hughes et al. 2003). No obvious differences between As(+3) and As(+5) were found, although residual levels after 24 hours tended to be higher for As(+3) than As(+5) (Vahter and Norin 1980). However, *in vitro* studies have found that the cellular uptake of As(+3) was higher than that of As(+5) (Bertolero et al. 1987; Dopp et al. 2004); in mouse cells, the difference was 4-fold (Bertolero et al. 1987). In hamsters, increases in tissue levels were noted after oral treatment with As(+3) for most tissues (hair, kidney, liver, lung, skin, muscle), with the largest increases in liver and lung (Yamauchi and Yamamura 1985). Liver and kidney arsenic concentrations increased with dose in dogs fed arsenite in the diet for 6 months (Neiger and Osweiler 1992). A study examining the speciation of arsenic following a single dose exposure to sodium arsenate to mice (Kenyon et al. 2005) found that the levels of inorganic arsenic and DMA were similar in the blood, liver, and kidney; much lower levels of MMA were found in these tissues. The concentration of DMA in the lungs exceeded inorganic arsenic and the levels of inorganic arsenic and MMA were similar; the DMA concentration was about 6 times higher than that of inorganic arsenic.

Inorganic arsenic crosses the placental barrier and selectively accumulates in the neuroepithelium of the developing animal embryo (Hanlon and Ferm 1977; Lindgren et al. 1984). Following maternal exposure to arsenite or arsenate throughout gestation and lactation, inorganic arsenic and DMA were detected in the

newborn mouse brains (Jin et al. 2006). The levels of inorganic arsenic in the brain were similar to those in the newborn livers; however, the levels of DMA in the brain were about twice as high as in the liver.

Organic Arsenicals. No studies were located on the distribution of organic arsenicals in humans following oral exposure. Studies in animals found MMA and DMA distributed to all tissues after acute oral doses (Hughes et al. 2005; Stevens et al. 1977; Vahter et al. 1984; Yamauchi and Yamamura 1984; Yamauchi et al. 1988). In mice, MMA is rapidly distributed throughout the body with peak tissue concentrations occurring between 0.25 and 4 hours after administration of a single gavage dose of 0.4 or 4 mg MMA/kg (Hughes et al. 2005). The peak levels of MMA in the bladder, kidneys, and lungs were higher than blood, with the highest levels occurring in the bladder. The terminal half-lives of MMA were 4.2–4.9 hours in the liver, lung, and blood, 9.0 hours in the urinary bladder, and 15.9 hours in the kidney in mice dosed with 0.4 mg MMA/kg; similar half-lives were measured in the 4.0 mg MMA/kg mice. Two hours after dosing, most of the methylated arsenic in the tissues was in the form of MMA. In rats exposed to 100 mg/kg DMA in the diet for 72 days, high levels of arsenic was detected in the blood (Lu et al. 2004a). The arsenic was primarily found in the erythrocyte; the concentration in the erythrocyte was 150 times higher than the arsenic concentration in the plasma.

3.4.2.3 Dermal Exposure

No studies were located regarding distribution of inorganic or organic arsenicals in humans or animals after dermal exposure.

3.4.2.4 Other Routes of Exposure

Inorganic Arsenicals. Studies in mice, rabbits, and monkeys injected intravenously with solutions of arsenite or arsenate confirm that arsenic is widely distributed throughout the body (Lindgren et al. 1982; Marafante and Vahter 1986; Vahter and Marafante 1983; Vahter et al. 1982). Shortly after exposure, the concentration of arsenic tends to be somewhat higher in liver, kidney, lung, and gastrointestinal epithelium (Hughes et al. 2000; Lindgren et al. 1982; Vahter and Marafante 1983; Vahter et al. 1982), but levels tend to equilibrate over time. Arsenate shows a tendency to deposit in skeletal tissue that is not shared by arsenite (Lindgren et al. 1982, 1984), presumably because arsenate is an analog of phosphate.

The distribution of arsenic in the rat is quite different from other animal species. Following intramuscular injection of carrier-free radio-arsenate in rats, most of the injected arsenic became bound to hemoglobin in red blood cells, and very little reached other tissues (Lanz et al. 1950). However, similar experiments

in dogs, mice, guinea pigs, rabbits, and chicks found very little uptake of arsenic into the blood in these species (cats gave intermediate results).

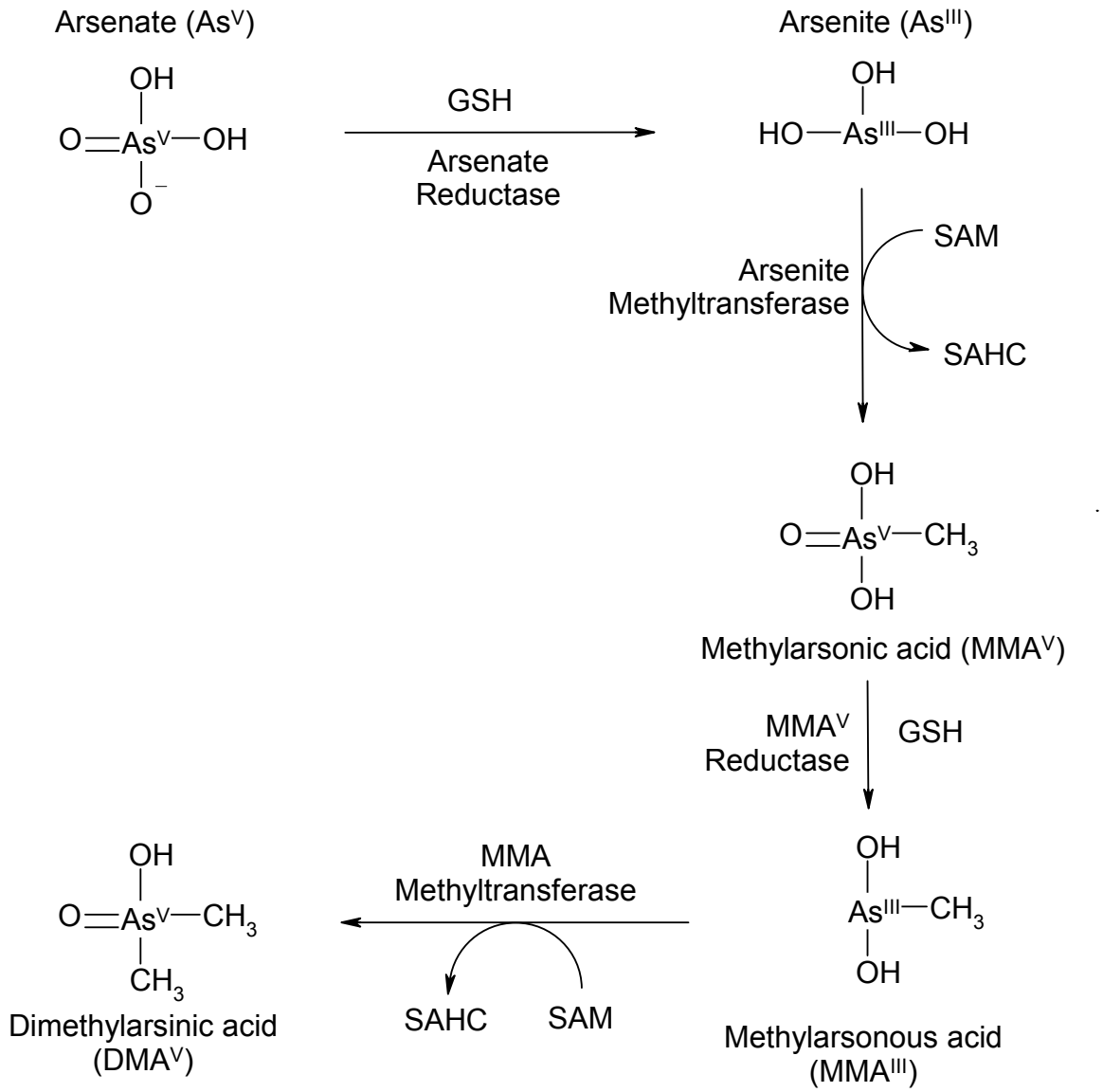
Organic Arsenicals. Following intravenous administration of DMA in mice, DMA is rapidly distributed throughout the body (Hughes et al. 2000). In the blood, the DMA was initially detected in the plasma, but fairly rapidly equilibrated between the plasma and erythrocytes. Blood, plasma, erythrocyte, liver, and kidney distribution and elimination of DMA did not differ in groups of mice administered 1.11 or 111 mg DMA/kg. However, a significant difference in DMA elimination from the lungs was observed; the elimination half-time increased from 91 minutes in the 1.11 mg DMA/kg group to 6,930 minutes in the 111 mg DMA/kg group.

3.4.3 Metabolism

Inorganic Arsenicals. The metabolism of inorganic arsenic has been extensively studied in humans and animals, and is diagrammed in Figure 3-7. Two basic processes are involved: (1) reduction/oxidation reactions that interconvert As(III) and As(V), and (2) methylation reactions, which convert arsenite to MMA and DMA. The resulting series of reactions results in the reduction of inorganic arsenate to arsenite (if necessary), methylation to MMA(V), reduction to MMA(III), and methylation to DMA(V). These processes appear to be similar whether exposure is by the inhalation, oral, or parenteral route. The human body has the ability to change inorganic arsenic to organic forms (i.e., by methylation) that are more readily excreted in urine. In addition, inorganic arsenic is also directly excreted in the urine. It is estimated that by means of these two processes, >75% of the absorbed arsenic dose is excreted in the urine (Marcus and Rispin 1988), although this may vary with the dose and exposure duration. This mechanism is thought to have an upper-dose limit which, when overwhelmed, results in a higher incidence of arsenic toxicity. This is supported by a case report of an individual who died 3 days after ingesting 8 g of arsenic trioxide (about 3 g of arsenic) (Benramdane et al. 1999a). Only 20% of the total arsenic in all tissues analyzed was methylated (14% MMA, 6% DMA), while 78% remained as arsenite and 2% as arsenate.

The majority of the evidence characterizing the metabolic pathways of arsenic is derived from analysis of urinary excretion products. Exposure of humans to either arsenates or arsenites results in increased levels of inorganic As(+3), inorganic As(+5), MMA, and DMA in urine (Aposhian et al. 2000a, 2000b; Buchet et al. 1981a, 1981b; Concha et al. 1998a, 1998b; Crecelius 1977; Kurttio et al. 1998; Lovell and Farmer

Figure 3-7. Inorganic Arsenic Biotransformation Pathway



SAHC = S-adenosylhomocysteine; SAM = S-adenosylmethionine

Source: adapted from Aposhian et al. 2000b

1985; Smith et al. 1977; Tam et al. 1979b; Vahter 1986). Similar results are obtained from studies in mice (Vahter 1981; Vahter and Envall 1983), hamsters (Hirata et al. 1988; Marafante and Vahter 1987; Takahashi et al. 1988), and rabbits (Maiorino and Aposhian 1985; Marafante et al. 1985; Vahter and Marafante 1983). Historically, little distinction was made between MMA(V) and MMA(III) in the urine in most studies, and the assumption was that the majority of MMA in the urine was MMA(V); however, Aposhian et al. (2000a, 2000b) demonstrated that the methylated arsenic atom may be in either valance state.

The relative proportions of As(+3), As(+5), MMA, and DMA in urine can vary depending upon the chemical administered, time after exposure, route of exposure, dose level, and exposed species. In general, however, DMA is the principal metabolite following long-term exposure, with lower levels of inorganic arsenic [As(+3) and As(+5)] and MMA. In humans, the relative proportions are usually about 40–75% DMA, 20–25% inorganic arsenic, and 15–25% MMA (Buchet et al. 1981a; Hopenhayn et al. 2003b; Loffredo et al. 2003; Mandal et al. 2001; Smith et al. 1977; Tam et al. 1979b; Tokunaga et al. 2002; Vahter 1986). With relatively constant exposure levels, these metabolic proportions remain similar over time (Concha et al. 2002), and appear to be similar among family members (Chung et al. 2002). One study of groups of women and children in two villages in Argentina showed that children ingesting large amounts of arsenic in their drinking water (200 µg/L) excreted about 49% inorganic arsenic and 47% DMA (Concha et al. 1998b). This compared to 32% inorganic arsenic and 66% DMA for the women in the study. This may indicate that metabolism of arsenic in children is less efficient than in adults. The rabbit has a ratio of metabolites similar to human adults (Maiorino and Aposhian 1985), suggesting that this may be a good animal model for toxicokinetics in humans. Mice may also be a good human toxicokinetic model based on the similarity of arsenic metabolism and deposition (Vahter et al. 2002). In contrast, the guinea pig, marmoset, and tamarin monkey do not methylate inorganic arsenic (Healy et al. 1998; Vahter and Marafante 1985; Vahter et al. 1982; Zakharyan et al. 1996); thus, they may be poor models for humans.

Reduction of arsenate to arsenite can be mediated by glutathione (Menzel et al. 1994). Scott et al. (1993) showed that glutathione forms complexes with both arsenate and arsenite *in vitro*, and that glutathione is oxidized (and arsenate reduced) in the glutathione-arsenate reaction. Studies *in vitro* indicate that the substrate for methylation is As(+3), and that As(+5) is not methylated unless it is first reduced to As(+3) (Buchet and Lauwerys 1985, 1988; Lerman et al. 1983). The main site of methylation appears to be the liver, where the methylation process is mediated by enzymes that utilize S-adenosylmethionine as cosubstrate (Buchet and Lauwerys 1985, 1988). Under normal conditions, the availability of methyl

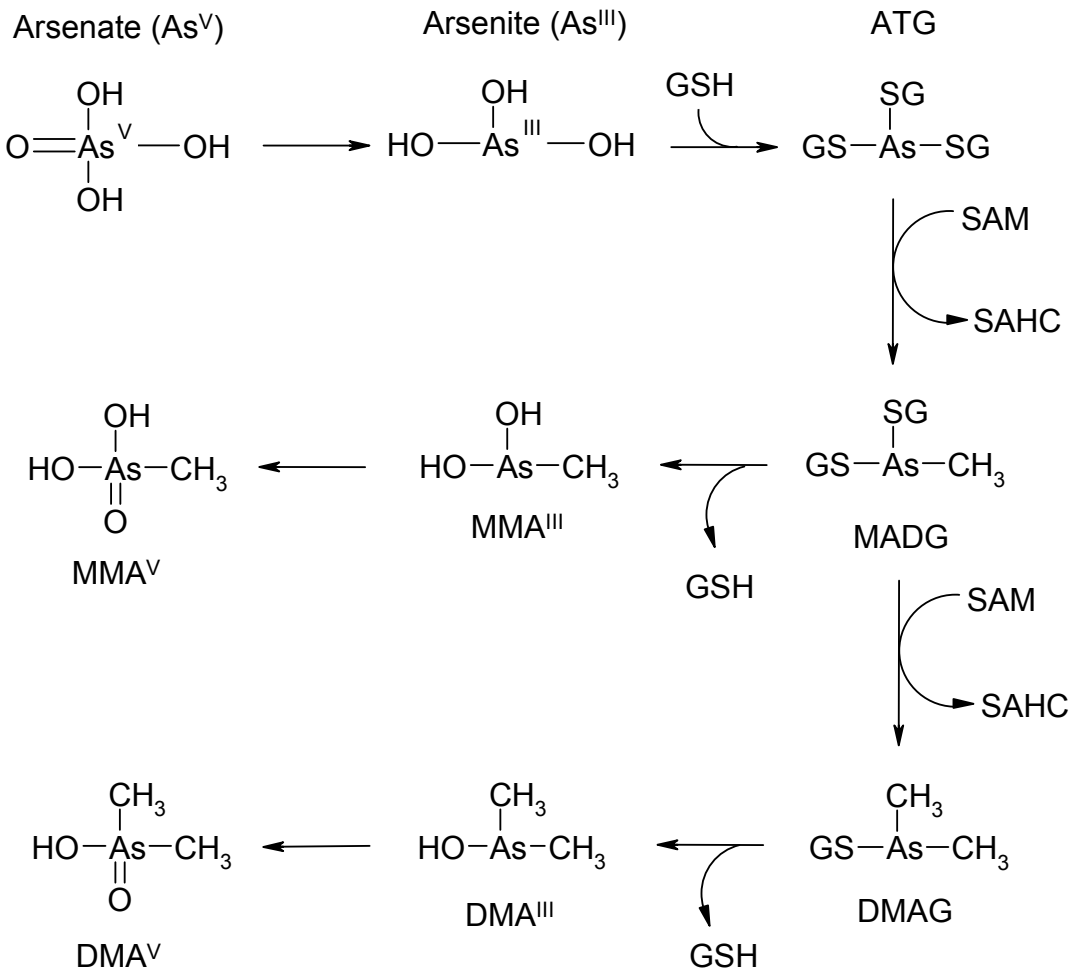
donors (e.g., methionine, choline, cysteine) does not appear to be rate limiting in methylating capacity, either in humans (Buchet et al. 1982) or in animals (Buchet and Lauwerys 1987; Buchet et al. 1981a). However, severe dietary restriction of methyl donor intake can result in significant decreases in methylating capacity (Buchet and Lauwerys 1987; Vahter and Marafante 1987).

Arsenic methyltransferase and MMA methyltransferase activities have been purified to homogeneity from cytosol of rabbit liver (Zakharyan et al. 1995), Rhesus monkey liver (Zakharyan et al. 1996), and rat liver (Thomas et al. 2004). It appears that a single protein catalyzes both activities. This activity transfers a methyl group from S-adenosylmethionine to As(+3) yielding MMA, which is then further methylated to DMA. Reduced glutathione is probably a co-factor *in vivo*, but other thiols can substitute *in vitro* (L-cysteine, dithiothreitol). The substrate saturation concentration for rabbit arsenite methyltransferase is 50 μM, for MMA methyltransferase it is 1,000 μM. The purified activity is specific for arsenite and MMA; selenite, selenate, selenide, and catechols do not serve as substrates. Thomas et al. (2004) reported cloning the gene for an S-adenosylmethionine-dependent methyltransferase from rat liver cytosol that catalyzes the conversion of arsenic to methylated and dimethylated species. It bears a high similarity to translations of *cyt19* genes in both the mouse and the human; both this gene and protein are now termed arsenic (+3 oxidation state) methyltransferase (AS3MT).

Studies in mice indicate that exposure to arsenic does not induce arsenic methylation activity (Healy et al. 1998). Mice receiving up to 0.87 mg As/kg/day as sodium arsenate in drinking water for 91 days had the same arsenic methylating activity as unexposed controls. Specific activities were highest in testis (1.45 U/mg) followed by kidney (0.70 U/mg), liver (0.40 U/mg), and lung (0.20 U/mg). None were affected by arsenic exposure.

An alternative biotransformation pathway (Figure 3-8) has recently been proposed for arsenic (Hayakawa et al. 2005) based on the nonenzymatic formation of glutathione complexes with arsenite resulting in the formation of arsenic triglutathione. The arsenic triglutathione is subsequently methylated by AS3MT to form monomethyl arsenic glutathione. At low glutathione levels (1 mM), the monomethyl arsenic glutathione is hydrolyzed to form MMA(III). At high glutathione levels (5 mM), the monomethyl arsenic glutathione is methylated to dimethylarsinic glutathione by AS3MT. Dimethylarsinic glutathione is quickly hydrolyzed to form DMA(III) (Hayakawa et al. 2005; Thomas et al. 2007). In the classical inorganic arsenic biotransformation pathway (Figure 3-7), MMA(V) is converted to the more toxic MMA(III); in contrast, in the alternative pathway, MMA(III) is converted to the less toxic MMA(V).

Figure 3-8. Alternative Inorganic Arsenic Biotransformation Pathway



ATG = arsenic triglutathione; DMAG = dimethylarsinic glutathione; GSH = glutathione; MADG = monomethyl arsenic glutathione; SAHC = S-adenosylhomocysteine; SAM = S-adenosylmethionine

Source: Hayakawa et al. 2005; Thomas et al. 2007

Since methylation tends to result in lower tissue retention of inorganic arsenic (Marafante and Vahter 1984, 1986; Marafante et al. 1985; Vahter and Marafante 1987), the methylation process is usually viewed as a detoxification mechanism. However, several studies showing an elevated toxicity of MMA(III) relative even to As(III) in cultured human liver cells (Petrick et al. 2000, 2001) have called this assumption into question. Because methylation is an enzymic process, an important issue is the dose of arsenic that saturates the methylation capacity of an organism, resulting in a possible increased level of the more toxic As(III) in tissues, or whether or not such a dose exists. Limited data from studies in humans suggest that methylation may begin to become limiting at doses of about 0.2–1 mg/day (0.003–0.015 mg/kg/day) (Buchet et al. 1981b; Marcus and Rispin 1988). However, these observations are relatively uncertain since they are based on data from only a few subjects, and the pattern of urinary excretion products in humans who ingested high (near lethal) oral doses or were exposed to elevated levels in the workplace is not much different from that in the general population (Lovell and Farmer 1985; Vahter 1986). Furthermore, the nutrient intakes reported by Engel and Receveur (1993) were sufficient to accommodate the body stores of methyl groups needed for arsenic biomethylation. At the highest arsenic level reported in the endemic area, the biomethylation process required only a few percent of the total daily methyl intake (Mushak and Crocetti 1995). Thus, the dose rate at which methylation capacity becomes saturated cannot be precisely defined with current data.

Organic Arsenicals. With the exception of arsenosugars, which may undergo extensive metabolism, organic arsenicals appear to undergo little metabolism. Humans who ingested a dose of MMA converted a small amount (about 13%) to DMA (Buchet et al. 1981a). Similarly, in mice and hamsters, DMA and MMA are primarily excreted unchanged in the urine (Hughes et al. 2005; Marafante et al. 1987b; Vahter et al. 1984). In mice, a small percentage of MMA is methylated to DMA and some is further methylated to trimethylarsine oxide (TMAO) (Hughes et al. 2005). In contrast, administration of MMA(III) to mice resulted in the excretion of mostly DMA(V) and smaller amounts of MMA(V), MMA(III), and DMA(III) (Hughes et al. 2005). As with MMA, only a small percentage (<10%) of the DMA is methylated to TMAO (Hughes et al. 2005; Marafante et al. 1987b; Yamauchi and Yamamura 1984; Yamauchi et al. 1988).

MMA and DMA are more extensively methylated in rats compared to other animal species. After 1 week of exposure to 100 mg As/kg/day as MMA in drinking water, rats excreted 50.6% of the total arsenic in urine as MMA, 19.0% as DMA, 6.9% as TMAO, and 0.4% as tetramethylarsonium (Yoshida et al. 1998). In contrast, mice exposed to a single dose of 40 mg As/kg as MMA excreted 89.6% of the dose as MMA, 6.2% as DMA, and 1.9% as TMAO (Hughes et al. 2005). Similarly, 24 hours after administration of a

single oral dose of 50 mg As/kg as MMA in hamsters, 26.9% was excreted in urine as MMA, 1.43% as DMA, and 0.07% as trimethylarsenic compound (Yamauchi et al. 1988). As with MMA, oral exposure of mice and hamsters to DMA results in most of the dose being excreted in the urine in the form of DMA (or DMA complex) (Marafante et al. 1987b); in rats, the levels of DMA and TMAO are about equal (Yoshida et al. 1998).

The available data suggest that the methylarsenates are not demethylated to inorganic arsenic either in humans (Buchet et al. 1981a; Marafante et al. 1987b) or in animals (rats and hamsters) (Stevens et al. 1977; Yamauchi and Yamamura 1984; Yoshida et al. 2001).

3.4.4 Elimination and Excretion

3.4.4.1 Inhalation Exposure

Inorganic Arsenicals. As noted previously (see Section 3.4.1.1), urinary excretion of arsenic appears to account for 30–60% of the inhaled dose (Holland et al. 1959; Pinto et al. 1976; Vahter et al. 1986). Since the deposition fraction usually ranges from about 30 to 60% for most respirable particles (EPA 1989b), this suggests that nearly all arsenic that is deposited in the lung is excreted in the urine. The time course of excretion in humans exposed by inhalation has not been thoroughly investigated, but urinary arsenic levels in workers in a smelter rose within hours after they came to work on Monday and then fell over the weekend (Vahter et al. 1986). This implies that excretion is fairly rapid, and this is supported by intratracheal studies in rats (Rhoads and Sanders 1985) and hamsters (Marafante and Vahter 1987), where whole-body clearance of administered arsenate or arsenite occurred with a half-time of 1 day or less. However, the study in rats (Rhoads and Sanders 1985) found that the clearance of arsenic trioxide was biphasic, with 95% cleared with a half-time of 29 minutes and the remaining arsenic cleared with a half-time of 75 days. For sodium arsenate and sodium arsenite, <0.1% of the dose was retained in the lung 3 days after exposure of hamsters; 1.3% of the arsenic trisulfide dose was retained after 3 days (Marafante and Vahter 1987). The Marafante and Vahter (1987) study suggested that lung clearance was influenced by compound solubility. The primary forms of arsenic found in the urine of inhalation-exposed humans are DMA and MMA, with inorganic arsenic comprising <25% of the total urinary arsenic (Apostoli et al. 1999).

Organic Arsenicals. No studies were located regarding the excretion of organic arsenicals by humans or animals after inhalation exposure. However, rats that were given a single intratracheal dose of DMA

excreted about 60% in the urine and about 8% in the feces within 24 hours (Stevens et al. 1977). This indicates that organic arsenicals are likely to be promptly excreted after inhalation exposure.

3.4.4.2 Oral Exposure

Inorganic Arsenicals. Direct measurements of arsenic excretion in humans who ingested known amounts of arsenite or arsenate indicate that very little is excreted in the feces (Bettley and O'Shea 1975), and that 45–85% is excreted in urine within 1–3 days (Apostoli et al. 1999; Buchet et al. 1981a; Crecelius 1977; Mappes 1977; Tam et al. 1979b). At low exposure levels, urinary arsenic levels generally increase linearly with increasing arsenic intake (Calderon et al. 1999). During lactation, a very small percent of ingested arsenic may also be excreted in the breast milk (Concha et al. 1998a). A similar pattern of urinary and fecal excretion is observed in hamsters (Marafante and Vahter 1987; Yamauchi and Yamamura 1985) and mice (Vahter and Norin 1980); this pattern is typically modeled as a biphasic process (e.g., Hughes et al. 2003). Generally, whole body clearance is fairly rapid, with half-times of 40–60 hours in humans (Buchet et al. 1981b; Mappes 1977). Clearance is even more rapid in mice and hamsters, with 90% removed in 2 days (Hughes et al. 2003; Marafante and Vahter 1987; Vahter 1981; Vahter and Norin 1980).

A study in pregnant women exposed to elevated levels of inorganic arsenic in drinking water found that most of the ingested arsenic was excreted in the urine as DMA (79–85%), with smaller amounts excreted as inorganic arsenic (8–16%) or MMA (5–6%) (Christian et al. 2006). Similarly, in mice, arsenate is primarily excreted in the urine as DMA, with lesser amounts of inorganic arsenic and MMA (Kenyon et al. 2005). Following a single oral dose of 10 µmol/kg sodium arsenate, 78.4% was excreted as DMA, 20.2% as inorganic arsenic, and 1.45% as MMA; at a 10-fold higher dose, the ratio of DMA to inorganic arsenic decreased (57.7% DMA, 39.8% inorganic arsenic, and 2.59% MMA).

Arsenic is also excreted in the bile via the formation of two arsenic-glutathione complexes (arsenic triglutathione and methylarsenic diglutathione) (Kala et al. 2000). In rats administered 5.0 mg/kg sodium arsenite, equal amounts of arsenic triglutathione and methylarsenic diglutathione were found in the bile 18–20 minutes after exposure. At a lower arsenic dose (0.5 mg/kg), only methylarsenic diglutathione was found. As discussed in Section 3.4.4.4, biliary excretion of arsenic has also been detected in mice, hamsters, guinea pigs, and rabbits following parenteral exposure (Csanaky and Gregus 2002).

Organic Arsenicals. Studies in humans indicate that ingested MMA and DMA are excreted mainly in the urine (75–85%), and this occurs mostly within 1 day (Buchet et al. 1981a; Marafante et al. 1987b). This is supported by studies in rats, mice, and hamsters, although in animals, excretion is more evenly distributed between urine and feces (Hughes et al. 2005; Marafante et al. 1987b; Stevens et al. 1977; Yamauchi and Yamamura 1984; Yamauchi et al. 1988). In mice administered 40 mg As/kg as DMA, 56.4% was excreted in the urine as DMA, 7.7% as a DMA complex, and 3.5% as TMAO during a 48-hour period after dosing; in the feces, 24.3% was DMA and 4.9% as DMA complex (Marafante et al. 1987b). In hamsters, 38.7% was DMA, 11.2% as DMA complex, and 6.4% as TMAO in the urine; in the feces, 37.3% as DMA and 4.9% as DMA complex. As with DMA, most MMA is excreted in the urine and feces as parent compound. In the urine of mice administered 0.4 mg As/kg as MMA, 98.2% of the urinary arsenicals was in the form of MMA(V) and 1.8% as MMA(III) (Hughes et al. 2005); at a 10-fold higher dose, 89.6% was excreted as MMA(V), 1.2% as MMA(III), 6.2% as DMA(V), 1.1% as DMA(III), and 1.9% as TMAO. As discussed previously, exposure of rats to MMA or DMA results in the excretion of a higher percentage of metabolites. After 1 week exposure to MMA, 50.6% of the dose was excreted as MMA, 19.0% as DMA, and 6.9% of TMAO (Yoshida et al. 1998). A 1-week exposure to DMA, 44.9% was excreted as DMA in the urine and 40.0% as TMAO (Yoshida et al. 1998). A longer-term exposure to DMA (>7 months) resulted in a higher percentage of the amount of parent compound excreted; 56–65% as DMA and 23–35% as TMAO (Li et al. 1998; Wanibuchi et al. 1996; Yoshida et al. 1998).

In mice and hamsters, DMA and MMA are rapidly cleared from the body (Hughes et al. 2005; Marafante et al. 1987b; Vahter et al. 1984). In mice, 85% of the initial oral dose of DMA was eliminated from the body with a half-life of 2.5 hours (Vahter et al. 1984). In contrast to the mouse data, 45% on the initial DMA dose to rats was eliminated with a half-time of 13 hours and the remaining 55% had an elimination half-time of 50 days (Vahter et al. 1984).

3.4.4.3 Dermal Exposure

Inorganic Arsenicals. No studies were located regarding excretion of inorganic arsenicals in humans or animals following dermal exposure. In rats, arsenic absorbed through the tail was excreted approximately equally in urine and feces, similar to the excretion pattern following oral exposure (Dutkiewicz 1977).

Organic Arsenicals. No studies were located regarding excretion of organic arsenicals in humans or animals following dermal exposure.

3.4.4.4 Other Routes of Exposure

Inorganic Arsenicals. Excretion of arsenate and arsenite following parenteral exposure of animals is similar to that seen following oral exposure. In rabbits and mice, urinary excretion within 8 hours usually accounts for about 50–80% of the dose (Maehashi and Murata 1986; Maiorino and Aposhian 1985; Vahter and Marafante 1983). Somewhat lower levels (30–40%) are excreted in the urine of marmoset monkeys (Vahter and Marafante 1985; Vahter et al. 1982), probably because of the absence of methylation in this species. Whole-body clearance studies in mice indicate that arsenate is over 65% removed within 24 hours, while arsenite is about 86% removed at 24 hours (Lindgren et al. 1982). A relatively small proportion of an injected dose of arsenic V (10% for rats, 4% for mice, and <2% for hamsters, guinea pigs, and rabbits) was found to be excreted into the bile within the first 2 hours postinjection (Csanaky and Gregus 2002). Following arsenic III injection, a much greater percentage (92% for guinea pigs and 75% for rats) of the arsenic was found in the bile in the first 2 hours after administration (Csanaky and Gregus 2002). Similarly, approximately 40% of an intravenous dose of sodium arsenite was excreted into the bile of rats, most of it occurring during the first hour after exposure (Kala et al. 2000). Kala et al. (2000) determined that the biliary transport of arsenic was dependent on the formation of arsenic-glutathione complexes, which were transported out of hepatocytes by multidrug resistance associated protein 2 (MRP2/cMOAT); most of the arsenic in bile was in the form of arsenic triglutathione or methylarsenic diglutathione.

3.4.5 Physiologically Based Pharmacokinetic (PBPK)/Pharmacodynamic (PD) Models

Physiologically based pharmacokinetic (PBPK) models use mathematical descriptions of the uptake and disposition of chemical substances to quantitatively describe the relationships among critical biological processes (Krishnan et al. 1994). PBPK models are also called biologically based tissue dosimetry models. PBPK models are increasingly used in risk assessments, primarily to predict the concentration of potentially toxic moieties of a chemical that will be delivered to any given target tissue following various combinations of route, dose level, and test species (Clewell and Andersen 1985). Physiologically based pharmacodynamic (PBPD) models use mathematical descriptions of the dose-response function to quantitatively describe the relationship between target tissue dose and toxic end points.

PBPK/PD models refine our understanding of complex quantitative dose behaviors by helping to delineate and characterize the relationships between: (1) the external/exposure concentration and target tissue dose of the toxic moiety, and (2) the target tissue dose and observed responses (Andersen and

Krishnan 1994; Andersen et al. 1987). These models are biologically and mechanistically based and can be used to extrapolate the pharmacokinetic behavior of chemical substances from high to low dose, from route to route, between species, and between subpopulations within a species. The biological basis of PBPK models results in more meaningful extrapolations than those generated with the more conventional use of uncertainty factors.

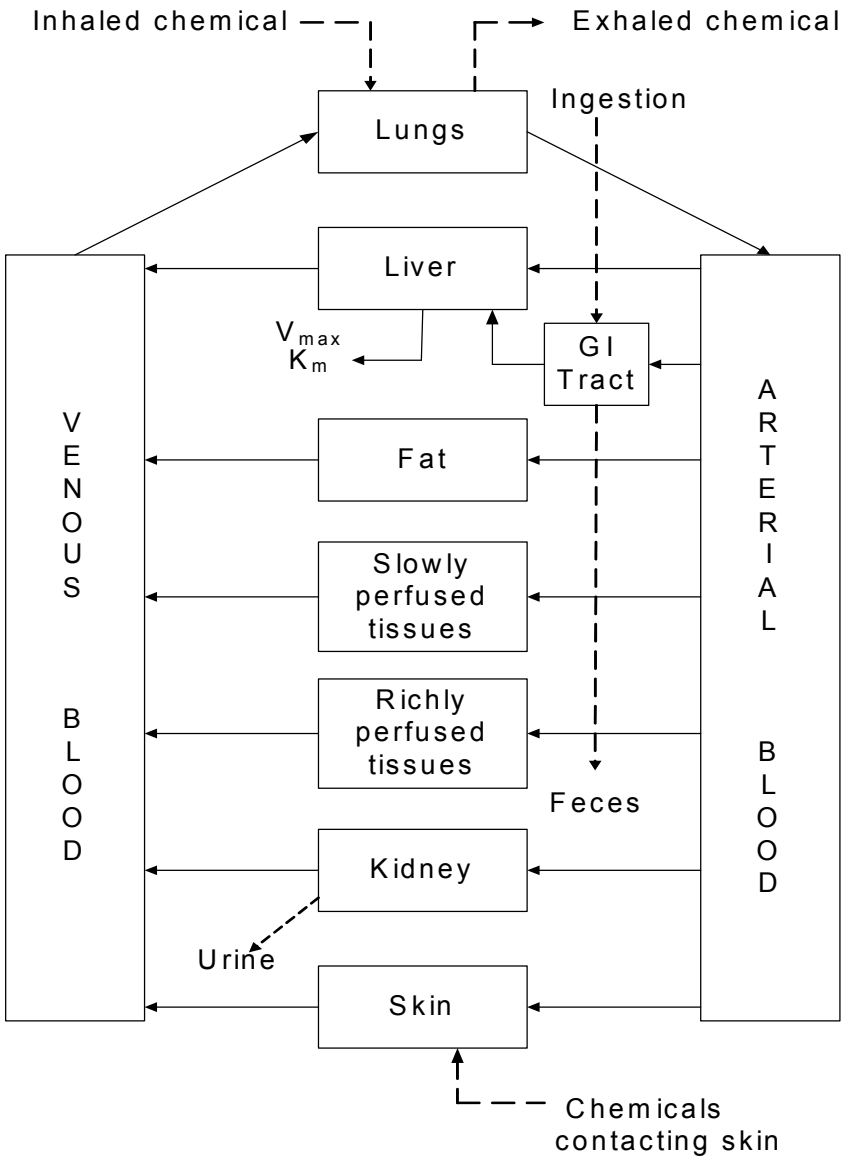
The PBPK model for a chemical substance is developed in four interconnected steps: (1) model representation, (2) model parameterization, (3) model simulation, and (4) model validation (Krishnan and Andersen 1994). In the early 1990s, validated PBPK models were developed for a number of toxicologically important chemical substances, both volatile and nonvolatile (Krishnan and Andersen 1994; Leung 1993). PBPK models for a particular substance require estimates of the chemical substance-specific physicochemical parameters, and species-specific physiological and biological parameters. The numerical estimates of these model parameters are incorporated within a set of differential and algebraic equations that describe the pharmacokinetic processes. Solving these differential and algebraic equations provides the predictions of tissue dose. Computers then provide process simulations based on these solutions.

The structure and mathematical expressions used in PBPK models significantly simplify the true complexities of biological systems. If the uptake and disposition of the chemical substance(s) are adequately described, however, this simplification is desirable because data are often unavailable for many biological processes. A simplified scheme reduces the magnitude of cumulative uncertainty. The adequacy of the model is, therefore, of great importance, and model validation is essential to the use of PBPK models in risk assessment.

PBPK models improve the pharmacokinetic extrapolations used in risk assessments that identify the maximal (i.e., the safe) levels for human exposure to chemical substances (Andersen and Krishnan 1994). PBPK models provide a scientifically sound means to predict the target tissue dose of chemicals in humans who are exposed to environmental levels (for example, levels that might occur at hazardous waste sites) based on the results of studies where doses were higher or were administered in different species. Figure 3-9 shows a conceptualized representation of a PBPK model.

If PBPK models for arsenic exist, the overall results and individual models are discussed in this section in terms of their use in risk assessment, tissue dosimetry, and dose, route, and species extrapolations.

Figure 3-9. Conceptual Representation of a Physiologically Based Pharmacokinetic (PBPK) Model for a Hypothetical Chemical Substance



Note: This is a conceptual representation of a physiologically based pharmacokinetic (PBPK) model for a hypothetical chemical substance. The chemical substance is shown to be absorbed via the skin, by inhalation, or by ingestion, metabolized in the liver, and excreted in the urine or by exhalation.

Source: adapted from Krishnan and Andersen 1994

Several PBPK models have been developed for inorganic arsenic; the Mann, Yu, and Menzel models are discussed below. A joint research effort between the Chemical Industry Institute of Toxicology (CIIT) Centers for Health Research, EPA, ENVIRON International, and the Electric Power Research Institute (EPRI) is underway to develop a biologically based dose response model of carcinogenicity. Part of this effort involves refining the existing PBPK models (Clewell et al. 2007).

3.4.5.1 Summary of PBPK Models

The Mann model (Gentry et al. 2004; Mann et al. 1996a, 1996b), Yu model (Yu 1998a, 1998b; Yu 1999a, 1999b), and Menzel model (Menzel et al. 1994) are the PBPK models for arsenic currently available. The Mann model simulates the absorption, distribution, metabolism, elimination, and excretion of As(+3), As(+5), MMA, and DMA after oral and inhalation exposure in mice, hamsters, rabbits, and humans. The Yu model simulates the absorption, distribution, metabolism, elimination, and excretion of As(+3), As(+5), MMA, and DMA after oral exposure to inorganic arsenic in mice, rats, or humans. The Menzel model is a preliminary model that predicts internal organ burden of arsenic during specific oral exposures, simulating the metabolism, distribution to organs and binding to organs in mice, rats, and humans.

3.4.5.2 Arsenic PBPK Model Comparison

The Mann model is a well-derived model, consisting of multiple compartments and metabolic processes, and modeling four chemical forms of arsenic (two organic and two inorganic), which has been validated using experimental data. The Yu model has more compartments than the Mann model, also models metabolism and fate of four forms of arsenic, and has likewise been validated using experimental data. The Menzel model is still preliminary and has not been validated.

3.4.5.3 Discussion of Models

The Mann Model

Risk assessment. The Mann model was not used for risk assessment.

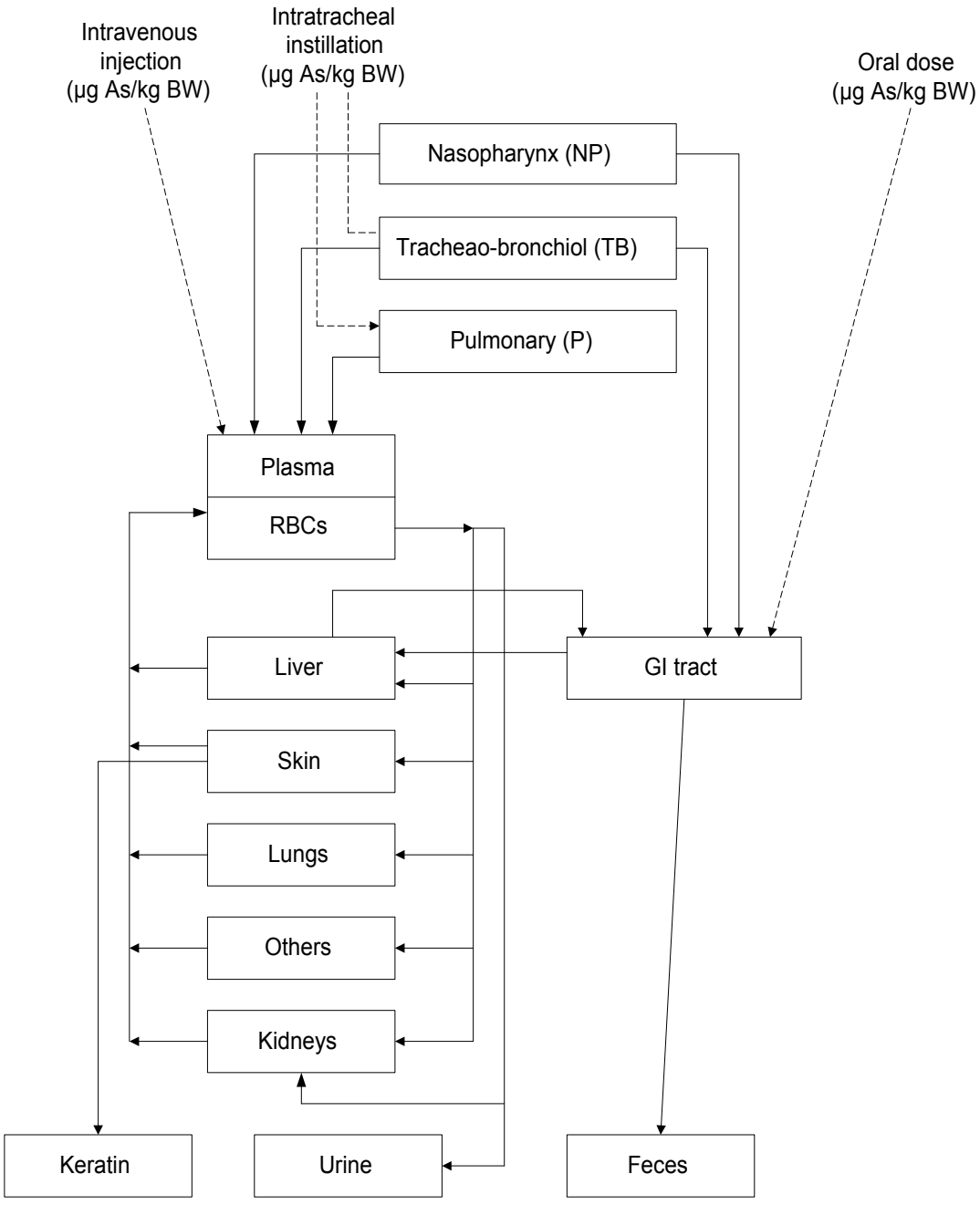
Description of the model. The Mann model was initially developed to simulate oral, intratracheal, and intravenous exposure to arsenic in rabbits and hamsters (Mann et al. 1996a). In a companion paper, the model was expanded to include inhalation exposure and extrapolated and applied to humans (Mann et al. 1996b). A subsequent paper further expanded the model to include mice (Gentry et al. 2004).

The model consists of six tissue compartments: blood, liver, kidneys, lungs, skin, and other tissues. The blood compartment is divided into plasma and red blood cell subcompartments, considered to be at equilibrium. Three routes of exposure are considered in the model. Oral exposure is considered to enter the liver from the gastrointestinal tract via first-order kinetics. Intratracheal exposure results in deposition into the pulmonary and tracheo-bronchial regions of the respiratory tract. Uptake into blood from the pulmonary region is considered to be via first order kinetics into plasma, uptake from the tracheo-bronchial region is by both transfer into plasma and transport into the gastrointestinal tract. Intravenous injection results in a single bolus dose into the plasma compartment.

Metabolism in the model consists of oxidation/reduction and two methylation reactions. The oxidation/reduction of inorganic arsenic was modeled as a first order process in the plasma, with reduction also included in the kidneys. Methylation of As(+3) was modeled as a two-step process occurring in the liver according to Michaelis-Menton kinetics.

Most physiological parameters were derived by scaling to body weight. In cases where parameters were not available (absorption rates, tissue affinity, biotransformation), estimates were obtained by fitting. This was done by duplicating the initial conditions of published experiments in the model, varying the unknown parameters and comparing the results of the simulation to the reported results. Tissue affinity constants were estimated using reported arsenic levels in tissues at various times after exposure. Metabolic rate constants and absorption rate constants were estimated using data for excretion of arsenic metabolites in urine and feces. Figure 3-10 shows the animal model and Tables 3-11, 3-12, 3-13, and 3-14 provide the parameters used in the animal model. The human model is similar to the animal models with adjustments for body weight and absorption and metabolic rates. A naso-pharynx compartment is included in the human model, which was not present in the animal models. Penetration and deposition in the respiratory tract are based on the log-normal particle size distribution of the aerosol. Metabolic and absorption rate constants were fitted using experimental data on urinary excretion of arsenic following a single oral dose of As(+3) (Buchet et al. 1981a) or As(+5) (Tam et al. 1979b) in volunteers. The lung absorption rate constant was obtained by fitting the total urinary excretion of arsenic as predicted with the model to experimental data obtained from occupational exposure to arsenic trioxide (Offergelt et al. 1992). Figure 3-11 shows the human model, and Tables 3-15 and 3-16 provide the data and constants used in the human model.

Figure 3-10. Parameters Used in the Mann PBPK Model for Animals



Source: Mann et al. 1996b

Table 3-11. Parameters Used in the Mann PBPK Model for Animals

Physiological parameter	Rabbit (body weight=3.5 kg)	Hamster (body weight=0.100 kg)
Blood volume (mL)	253	7.0
Organ weight (g)		
Liver	121	4.8
Kidneys	25	1.2
Lungs	31	1.0
Skin	420	17.1
Organ volume (mL)		
Others	2,386	62.0
Lumen volume (mL)		
Stomach	15	0.5
Small intestine	20	0.6
Blood flow (mL/minute)		
Cardiac output	556	38.3
Liver, hepatic	25	1.2
Liver, splanchic	98	6.0
Kidneys	100	7.0
Lungs	13	0.7
Skin	38	2.6
Others	282	20.8
Clearance (mL/minute)		
Glomerular Filtration Rate	10	0.6
Small intestine length (cm)	180	56.0
Total capillary surface area (cm^2)	93,835	2,681.0

PBPK = physiologically based pharmacokinetic

Source: Mann et al. 1996a

Table 3-12. Tissue Affinity Constants (K_{ij}) Obtained for the Mann PBPK Model for Animals by Fitting for Rabbits and Hamsters

	K_{ij} (unitless)			
Tissue (*i*)	As(V)	As(III)	MMA	DMA
Liver	1	200	10	1
Kidneys	40	20	100	5
Lungs	1	1	1	20
Skin	1	60	50	1
Others	10	40	1	1

DMA = dimethylarsinic acid; MMA = monomethylarsonic acid; PBPK = physiologically based pharmacokinetic

Source: Mann et al. 1996a

Table 3-13. Metabolic Rate Constants for the Mann PBPK Model for Animals Obtained by Fitting for Rabbits and Hamsters

Oxidation/reduction	First order	Rabbit	Hamster
Reduction	(1/hour)	3,000.00	100.00
Oxidation	(1/hour)	6,000.00	400.00
Kidney reduction	(1/hour)	30.00	1.00
Methylation	Michaelis–Menten		
1st step	$K_{M_{MMA}}$ (µmol/mL)	0.05	0.12
	$V_{MAX_{MMA}}$ (µmol/mL-hour)	4.00	0.12
2nd step	$K_{M_{DMA}}$ (µmol/mL)	0.90	0.08
	$V_{MAX_{DMA}}$ (µmol/mL-hour)	1.50	0.12

DMA = dimethylarsinic acid; MMA = monomethylarsonic acid; PBPK = physiologically based pharmacokinetic

Source: Mann et al. 1996a

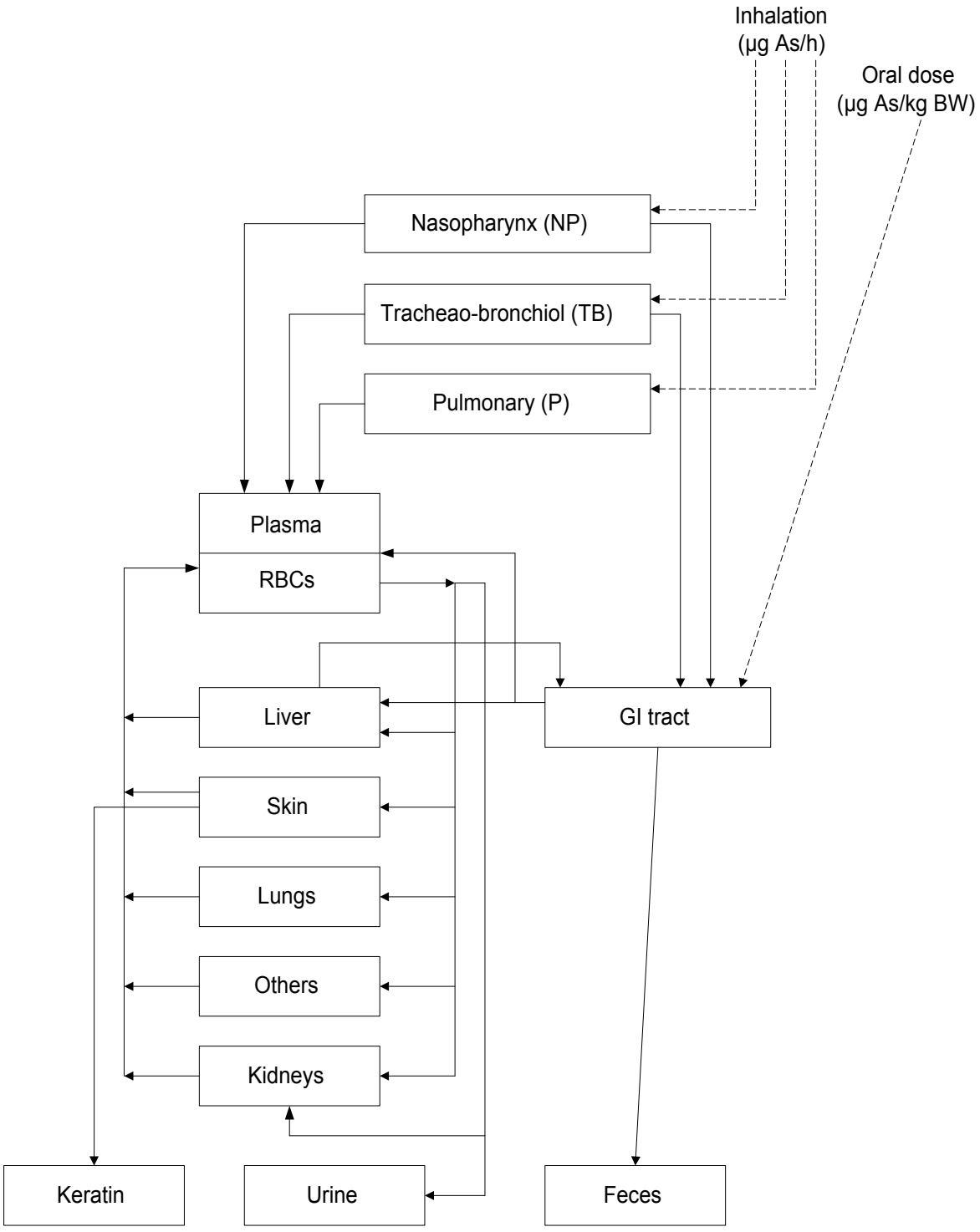
Table 3-14. Fitted Gastrointestinal Tract and Lung Absorption Half-time for the Hamster for the Mann PBPK Model

	Absorption, half-time (hour)	
Arsenic compound	Gastrointestinal tract	Lung
As(V)		
$Na_3(AsO_4)$	0.08	12
$Pb_3(AsO_4)$	0.39	690
As_2O_5	0.28	—
As(III)		
$NaAsO_2$	0.08	12
As_2S_3	0.48	12
As_2O_3	0.02	—
DMA	0.09	—

DMA = dimethylarsinic acid; PBPK = physiologically based pharmacokinetic

Source: Mann et al. 1996a

Figure 3-11. Parameters Used in the Mann PBPK Model for Humans



Source: Mann et al. 1996b

Table 3-15. Physiological Data Used in the Mann PBPK Model for Humans

Physiological parameter	Organ	Units	Human (body weight=70 kg)
Blood volume		mL	5,222
Organ weight	Liver	g	1,856
	Kidneys	g	314
	Lungs	g	584
	Skin	g	6,225
	Others	g	55,277
Lumen volume	Stomach	mL	274
	Small intestine	mL	393
Blood flow	Cardiac output	L/minute	5.29
	Liver, hepatic	L/minute	0.32
	Liver, splanchic	L/minute	1.02
	Kidneys	L/minute	0.95
	Lungs	L/minute	0.16
	Skin	L/minute	0.35
	Others	L/minute	2.49
Creatinine			
Male		g/day	1.7
Female		g/day	1.0
Clearance			
Glomerular filtration rate		mL/minute	156
Small intestine length		cm	481
Nasopharynx area		cm^2	177
Tracheobronchial area		cm^2	5,036
Pulmonary area		cm^2	712,471
Total capillary surface area		cm^2	$1,877x10^6$

DMA = dimethylarsinic acid; MMA = monomethylarsonic acid; PBPK = physiologically based pharmacokinetic

Source: Mann et al. 1996b

Table 3-16. Tissue Affinity Constants (K_{ij}) Obtained by Fitting the Mann PBPK Animal Model for Use with Humans

	K_{ij} (unitless)			
Tissue (i)	As(V)	As(III)	MMA	DMA
Liver	1	200	10	1
Kidneys	40	20	100	5
Lungs	1	1	1	20
Skin	1	60	50	1
Red blood cells	0.2	1.5	0.2	0.2
Others	10	40	1	1

DMA = dimethylarsinic acid; MMA = monomethylarsonic acid; PBPK = physiologically based pharmacokinetic

Source: Mann et al. 1996b

Validation of the model. The model was generally successful in describing the disposition of an intravenous dose of sodium arsenate in rabbits over a 24-hour period (Marafante et al. 1985). Discrepancies included a 6–7-fold overestimation of levels in skin at 24 hours and underestimation of As(+5) in plasma in the hour following injection. A statistical assessment of how well the model fit the empirical data was not presented. In hamsters, the model was also generally predictive of oral and intratracheal exposures (Marafante and Vahter 1987). Generally, predictions were better for the exposures to As(+5) than for those to As(+3).

The human model was validated using data from studies of repeated oral intake of sodium arsenite in volunteers (Buchet et al. 1981b), occupational exposure to arsenic trioxide and elemental arsenic (Vahter et al. 1986), and community exposure to As(+5) via drinking water (Harrington et al. 1978; Valentine et al. 1979). Simulations were generally in good agreement with the experimental data.

The predictions of tissue distribution, metabolism, and elimination of arsenic compounds from the mouse model were compared with experimental data, and showed generally good agreement. The model tended to overpredict the concentration of organic arsenicals in the lungs, and to a lesser extent in the kidneys and liver, while for inorganic arsenic, the model overpredicted the levels of arsenic (V) present in the urine of acutely-exposed mice.

Target tissues. Levels in skin were not well predicted by this model in animals. Results for the lung were not presented, except for the mouse model, which tended to overpredict lung levels. The human model was only used to predict urinary metabolites.

Species extrapolation. Species extrapolation was not attempted in this model. However, tissue affinities derived for the rabbit and hamster models were used in the human model.

Interroute extrapolation. Interroute extrapolation was not attempted in this model.

The Menzel Model

Risk assessment. The Menzel model was not used for risk assessment.

Description of the model. The Menzel model was developed to simulate oral exposure to arsenic from drinking water and food. Inhalation of arsenic in the particulate phase or as arsine gas is not considered. The chemical species in drinking water is assumed to be As(+5).

The model consists of two sets of compartments: those in which the pools of arsenic are not influenced by blood perfusion, and those in which blood perfusion does determine arsenic burden. The former set of compartments includes the gut, feces, hair, bladder, and urine. The latter set of compartments included lung, liver, fat, skin, kidney, and other tissues. Oral exposure is considered to enter the liver from the gastrointestinal tract.

The model followed that of Andersen and coworkers (Andersen et al. 1987; Ramsey and Andersen 1984). Data from mice were used to test predictions of absorption. Excretion is considered to be rapid and complete into the urine, with no reabsorption from the kidney. Fecal arsenic content accounts for unabsorbed arsenic excreted in the bile, and complex arsenic species from food. Metabolism includes reduction by glutathione and methylation. Arsenic accumulation in the skin, hair and nails was included by assuming that arsenic binds irreversibly to protein sulfide groups in hair and nails.

Validation of the model. The model was preliminary and has not been validated.

Target tissues. Target tissues have not yet been modeled.

Species extrapolation. Species extrapolation was not attempted in this model.

Interroute extrapolation. Interroute extrapolation was not attempted in this model.

The Yu Model

Risk assessment. The Yu model was not used for risk assessment.

Description of the model. The Yu model was developed to simulate oral exposure to arsenic in mice and rats (Yu 1998a, 1998b), and was later adapted for oral exposures in humans (Yu 1999a, 1999b). Inhalation of arsenic in the particulate phase or as arsine gas is not considered. As(+3), As(+5), MMA, and DMA were all considered in the model, though the movements of MMA and DMA were not considered.

The model consists of eight tissue compartments: intestine, skin, muscle, fat, kidney, liver, lung, and vessel-rich group (VRG, e.g., brain); in the human model, the VRG and kidney compartments were combined. Only oral exposure was considered. Absorption is based on absorption to the stomach, which then passes the arsenic to the gastrointestinal tract. From the gastrointestinal tract, arsenic is either transferred to the blood or excreted in the feces.

The physiological parameters for the model were obtained from published values in the literature. Tissue/blood partition coefficients were based on the postmortem blood and tissue concentrations from a fatal human poisoning case study (Saady et al. 1989). Tissue volumes and blood flow rates were based on published values from a number of sources (EPA 1988e; Reitz et al. 1990). Absorption and excretion rate constants were based on experimental observations of blood concentrations and urinary and fecal excretion following oral administration of inorganic arsenic (Odanaka et al. 1980; Pomroy et al. 1980). Metabolic rate constants for the methylation and dimethylation of inorganic arsenic were also based on experimental observations (Buchet et al. 1981a; Crecelius 1977). Figure 3-12 shows the model and Table 3-17 provides the parameters used for each species.

Validation of the model. The model was generally successful at predicting the urinary excretion 48 hours after administration of 5 mg/kg inorganic arsenic in both rats and mice. After 48 hours, the observed/predicted ratios associated with excreted doses ranged from 0.78 to 1.11 for the mouse and from 0.85 to 0.93 for the rat. However, the model overpredicted the amount of inorganic arsenic found in the feces of mice at 24 and 48 hours, and overpredicted the amount of DMA formed by exposed mice at 48 hours. In rats, the model overestimated the urinary and fecal excretion of inorganic arsenic at 24 hours postexposure, though at 48 hours, measured values all fell within the predicted ranges. The human model was also generally successful at predicting the urinary excretion of arsenic compounds following oral exposure, based on results of controlled human exposure studies (Buchet et al. 1981a; Vahter 1983). In general, however, the model underpredicts excretion at early time points and overpredicts at later time points, with 24 hours being the time at which its predictive capabilities agreed most strongly with available data.

The ability of the model to predict tissue burdens was not compared to actual data for any species.

Figure 3-12. Parameters Used in the Yu PBPK Model for Animals

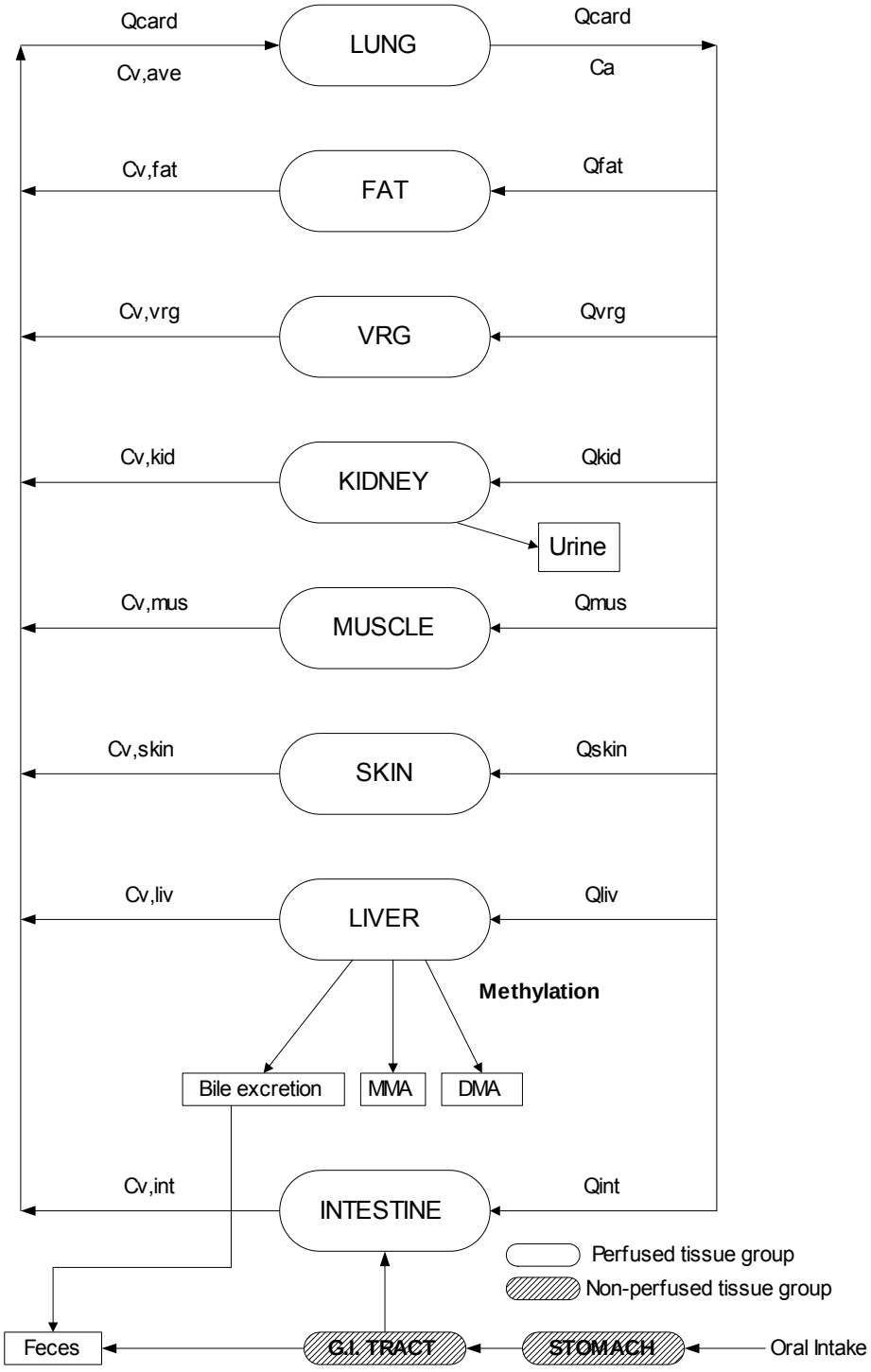


Table 3-17. Parameters Used in the Yu PBPK Model

	Mouse	Rat	Human
Partition coefficients			(As^{III}/As^{V}/MMA/DMA)
Intestine	6.0	6.0	2.8/2.8/1.2/1.4
Skin	5.0	5.0	2.5/2.5/1.25/1.25
VRG	6.0	6.0	Combined with kidney
Muscle	5.0	10.0	2.6/2.6/1.8/2.8
Fat	—	0.5	0.3/0.3/0.3/0.3
Kidney	8.5	7.5	4.15/4.15/1.8/2.075
Liver	10.0	10.0	5.5/5.3/2.35/2.65
Lung	4.0	4.0	4.15/4.15/1.8/2.075
Blood flow rate (mL/hour)			
Intestine	100	528	1,810
Skin	7.68	37.8	130
VRG	157	960	N/A
Muscle	153	1,260	25,850
Fat	—	253.2	6,467
Kidney	255	255	45,240
Liver	255	1,260	32,320
Lung	N/R	N/R	129,000
Tissue volume (mL)			
Intestine	1.94	6.9	558
Skin	1.83	15.4	606
VRG	0.81	23.0	N/A
Muscle	19.9	162	6,989
Fat	—	14.5	2,328
Kidney	0.484	1.63	248
Liver	1.67	5.82	422
Lung	0.124	1.0	400
Metabolism constants			
$Vmax_{(MMA)}$ (µmol/hour)	0.45	0.15	11.25
$Vmax_{(DMA)}$ (µmol/hour)	0.375	0.06	22.25
$Km_{(MMA)}$ (µmol/hour)	1.0	0.2	0.01
$Km_{(DMA)}$ (µmol/hour)	0.2	0.2	0.01

Table 3-17. Parameters Used in the Yu PBPK Model

	Mouse	Rat	Human
First-order rate constants			(As^{III}/As^{V}/MMA/DMA)
K_{SI} ($hour^{-1}$)	0.3	0.3	-/1.2/-/-
K_{AI} ($hour^{-1}$)	1.5	3.6	-/1.2/-/-
K_{fecal} ($hour^{-1}$)	0.33	0.048	-/0.0012/0.0/0.0
$K_{urinary}$ ($hour^{-1}$)	1.32	0.9	0.05/0.075/0.07/0.04
$K_{biliary}$ ($hour^{-1}$)	0.33	0.3	-/0.018/-/-

DMA = dimethylarsinic acid; MMA = monomethylarsonic acid; N/A = not applicable; N/R = not reported

Source: Yu 1998a, 1998b, 1999a, 1999b

Target tissues. Model predictions of tissue burdens were not compared to actual data. The model accurately predicted, with a few exceptions, the urinary and fecal excretion of inorganic arsenic and its metabolites in rats, mice, and humans.

Species extrapolation. Species extrapolation beyond rats and mice was not attempted using this model. The human model has not been compared to, or linked with, either of the rodent models.

Interroute extrapolation. Interroute extrapolation was not attempted using this model.

3.5 MECHANISMS OF ACTION

3.5.1 Pharmacokinetic Mechanisms

Arsenic absorption depends on its chemical form. In humans, As(+3), As(+5), MMA, and DMA are orally absorbed ≥75%. Arsenic is also easily absorbed via inhalation. Absorption appears to be by passive diffusion in humans and mice, although there is evidence (Gonzalez et al. 1995) for a saturable carrier-mediated cellular transport process for arsenate in rats (for review, see Rosen 2002). Dermal absorption appears to be much less than by the oral or inhalation routes. Bioavailability of arsenic from soil appears to be lower via the oral route than it is for sodium salts of arsenic. Arsenic in soil may form water insoluble compounds (e.g., sulfides), which are poorly absorbed.

Arsenic and its metabolites distribute to all organs in the body; preferential distribution has not been observed in human tissues at autopsy or in experiments with animal species other than rat (in which arsenic is concentrated in red blood cells). Since the liver is a major site for the methylation of inorganic arsenic, a “first-pass” effect is possible after gastrointestinal absorption; however, this has not been investigated in animal models.

Arsenic and its metabolites are largely excreted via the renal route. This excretion mechanism is not likely to be saturated within the dose range expected from human exposure. Excretion can also occur via feces after oral exposure; a minor excretion pathway is nails and hair. The methylation of inorganic arsenic is the major metabolism pathway. The proportion of metabolites recovered in urine (As(+3), As(+5), MMA, DMA) are roughly consistent in humans regardless of the exposure scenario. However, interindividual variation is great enough that it cannot be determined if capacity limitation may occur in some individuals.

The manifestation of arsenic toxicity depends on dose and duration of exposure. Single oral doses in the range of 2 mg As/kg and higher have caused death in humans. Doses as low as 0.05 mg As/kg/day over longer periods (weeks to months) have caused gastrointestinal, hematological, hepatic, dermal, and neurological effects. These effects appear to be a result of direct cytotoxicity. Long-term exposure (years) to drinking water at levels as low as 0.001 mg As/kg/day have been associated with skin diseases and skin, bladder, kidney, and liver cancer. Long-term inhalation exposure to arsenic has also been associated with lung cancer at air levels as low as 0.05–0.07 mg/m^3. It is not clear at this time why long-term toxicity is different between the oral and inhalation routes, given that arsenic is easily absorbed into the systemic circulation by both routes.

Studies in mice and rats have shown that arsenic compounds induce metallothionein, a metal-binding protein thought to detoxify cadmium and other heavy metals, *in vivo* (Albores et al. 1992; Hochadel and Waalkes 1997; Kreppel et al. 1993; Maitani et al. 1987a). The potency of arsenic compounds in inducing metallothionein parallels their toxicity (i.e., As(+3) > As(+5) > MMA > DMA). For cadmium, it is thought that metallothionein binds the metal, making it biologically inactive. For arsenic, however, only a small percentage of the administered arsenic is actually bound to metallothionein (Albores et al. 1992; Kreppel et al. 1994; Maitani et al. 1987a). *In vitro* studies have shown that affinity of arsenic for metallothionein is much lower than that of cadmium or zinc (Waalkes et al. 1984). It has been proposed that metallothionein might protect against arsenic toxicity by acting as an antioxidant against oxidative injury produced by arsenic (NRC 1999).

3.5.2 Mechanisms of Toxicity

Mechanisms of arsenic-induced toxicity and carcinogenicity have not been clearly identified. However, recent efforts to elucidate mechanisms of arsenic toxicity and carcinogenicity have resulted in numerous *in vitro* and *in vivo* reports. Whereas these mechanistic studies typically employed relatively high arsenic exposure levels, some of the most recent studies were performed using more environmentally-relevant exposure levels. Due to the extremely large amount of mechanistic data for arsenic, it is not feasible to include all pertinent primary studies that address issues concerning proposed mechanisms of arsenic toxicity and carcinogenicity. Therefore, the following discussion of mechanisms of arsenic toxicity represents a summary of information from several recent review articles (Chen et al. 2004, 2005; Florea et al. 2005; Hughes 2002; Kitchin 2001; Lantz and Hays 2006; Navas-Acien et al. 2005; Rossman 2003; Roy and Saha 2002; Thomas et al. 2007; Vahter 2002).

It is becoming increasingly evident that the toxicity and carcinogenicity of arsenic is likely to be closely associated with metabolic processes. Absorbed pentavalent arsenic (AsV) is rapidly reduced to trivalent arsenic (AsIII) at least partially in the blood. Much of the formed AsIII is distributed to tissues and taken up by cells (particularly hepatocytes). Many cell types appear to accumulate AsIII more rapidly than AsV. Because AsIII (as arsenite) is known to be more highly toxic than AsV (as arsenate), the reduction step may be considered bioactivation rather than detoxification. Glutathione appears to play a role in the reduction of AsV to AsIII, which is required prior to methylation. Methylation of arsenic ultimately forms relatively less toxic MMA and DMA; this process is accomplished by alternating between the reduction of AsV to AsIII and the addition of a methyl group; S-adenosylmethionine is considered to be the source of the methyl group. Both MMA and DMA are less reactive with tissue constituents than inorganic arsenic and both are readily excreted in the urine. The methylation process appears to include multiple intermediates, some of which are more reactive than inorganic arsenic. For example, reactive trivalent metabolites, MMAIII and DMAIII, have been detected in the urine of human subjects chronically exposed to arsenic in drinking water, and *in vitro* studies have demonstrated MMAIII to be more toxic than arsenite or arsenate to human hepatocytes, epidermal keratinocytes, and bronchial epithelial cells. Additional *in vitro* studies have demonstrated genotoxic and DNA damaging properties of both MMAIII and DMAIII.

AsV (as arsenate) has been demonstrated to: (1) replace phosphate in glucose-6-phosphate and 6-phosphogluconate *in vitro*, (2) replace phosphate in the sodium pump and the anion exchange transport system of human red blood cells, (3) diminish the *in vitro* formation of adenosine-t′-triphosphate (ATP) by replacing phosphate in enzymatic reactions, and (4) deplete ATP in some cellular systems, but not in human erythrocytes. However, it is becoming more apparent that the major source of arsenic toxicity and carcinogenicity is related to its reduction to arsenite.

AsIII (as arsenite) is known to react with thiol-containing molecules such as glutathione and cysteine *in vitro*. Methylated trivalent arsenics such as MMAIII are potent inhibitors of glutathione reductase and thioredoxin reductase. It has been suggested that binding of arsenite and methylated trivalent arsenicals to critical thiol groups could lead to the inhibition of essential biochemical reactions, alteration of cellular redox status, and eventual cytotoxicity. Binding of MMAIII and DMAIII to protein has also been demonstrated *in vitro*. Arsenite inhibits pyruvate dehydrogenase (PDH), a complex that oxidizes pyruvate to acetyl-CoA, a precursor to intermediates of the citric acid cycle that provides reducing equivalents to the electron transport system for ATP production. This property may explain the depletion of carbohydrates in arsenite-treated rats.

Evidence that arsenic may induce alterations in nitric oxide metabolism and endothelial function includes findings that persons exposed to high levels of arsenic in drinking water had decreased serum and urine concentrations of nitric oxide metabolites, which was reversed upon intervention with drinking water containing lower levels of arsenic. Urinary arsenic levels have been inversely associated with nitric oxide production in activated monocytes. Arsenite concentrations of 1–25 μM inhibited endothelial nitric oxide synthase activity and resulting decreased cell growth in human endothelial cells, although lower concentrations up-regulated the expression of constitutive nitric oxide synthase 3, which might serve as an explanation for observed arsenic-induced cell growth and angiogenesis.

Although epidemiological studies demonstrate the carcinogenicity of arsenic in humans, early animal cancer bioassays failed to demonstrate a carcinogenic effect following lifetime exposure to inorganic arsenic. However, more recent focus has resulted in the development of animal models that exhibit carcinogenic activity in skin, urinary bladder, liver, and lung, tissues implicated in arsenic-induced cancer in humans. This concordance in target sites among animal models and humans indicates that common mechanisms of action may be applicable to humans and laboratory animals.

Several modes of action have been proposed to explain, at least in part, the carcinogenicity of arsenic. It is likely that multiple mechanisms are involved, some of which may relate to noncancer effects as well.

Oxidative Stress. Mechanistic studies of arsenic toxicity have suggested a role of the generation of reactive oxygen species in the toxicity of inorganic arsenic. Results of both *in vivo* and *in vitro* studies of arsenic-exposed humans and animals suggest the possible involvement of increased lipid peroxidation, superoxide production, hydroxyl radical formation, blood nonprotein sulfhydrals, and/or oxidant-induced DNA damage. Reduction of cellular oxidant defense by treatment with glutathione-depleting agents results in an increased sensitivity of cells to arsenic toxicity. Support for mechanisms of toxicity that involves arsenic-induced oxidative stress includes findings that inhaled arsenic can predispose the lung to oxidative damage, chronic low-dose arsenic alters genes and proteins that are associated with oxidative stress and inflammation, and major transcriptional regulators of altered genes are redox sensitive.

Genotoxicity. Collectively, *in vitro* and *in vivo* genotoxicity assays have demonstrated that arsenics cause single strand breaks, formation of apurinic/apyrimidinic sites, DNA base and oxidative base damage, DNA-protein crosslinks, chromosomal aberrations, aneuploidy, sister chromatid exchanges, and micronuclei. Chromosomal aberrations, characterized by chromatid gaps, breaks and fragmentation,

endoreduplication, and chromosomal breaks, are dose-dependent and arsenite is more potent than arsenate. Both MMAIII and DMAIII are directly genotoxic and are many times more potent than arsenite at inducing DNA damage. Inorganic arsenic can potentiate the mutagenicity observed with other chemicals, although arsenic itself does not appear to induce point mutations. Arsenic-induced genotoxicity may involve oxidants or free radical species.

Altered Growth Factors→Cell Proliferation→Promotion of Carcinogenesis. Increased concentrations of growth factors can lead to cell proliferation and eventual promotion of carcinogenesis. Arsenic-induced cell death can also lead to compensatory cell regeneration and carcinogenesis. Altered growth factors, cell proliferation, and promotion of carcinogenesis have all been demonstrated in one or more systems exposed to arsenics. Altered growth factors and mitogenesis were noted in human keratinocytes. Cell death was observed in human hepatocytes and rat bladder epithelium. Cell proliferation was demonstrated in human keratinocytes and intact human skin and rodent bladder cells. Promotion of carcinogenesis was noted in rat bladder, kidney, liver, and thyroid, and mouse skin and lung.

Additional Mechanisms of Toxicity Data. Inorganic arsenic exposure has been shown to modify the expression of a variety of genes related to cell growth and defense, including the tumor suppressor gene p53, as well as to alter the binding of nuclear transcription factors. Carcinogenic effects of arsenic may result from a cocarcinogenic effect. Whereas arsenic exposure alone did not elicit skin tumors in mice, co-exposure to arsenic and ultraviolet light resulted in skin tumors that were greater in number and larger in size than those produced by ultraviolet light alone. Arsenate and arsenite enhanced the amplification of a gene that codes for the enzyme dihydrofolate reductase, arsenate being more potent than arsenite. Furthermore, inhibition of DNA repair has been demonstrated in arsenic-treated cells.

3.5.3 Animal-to-Human Extrapolations

The usefulness of animal models for toxicity studies with arsenic is significantly limited by two major factors. First and most importantly, no animal model exists for the health effect of greatest concern for human exposure: carcinogenicity in skin and other organs after oral exposure. Second, the pattern of metabolism in humans (significant excretion of the methylated forms of arsenic) is unlike that of most other mammalian species (the mouse and rabbit may be exceptions). The ratios of inorganic to organic arsenic excreted also vary between species. The rat sequesters arsenic in its erythrocytes and is not a suitable model for human toxicity.

3.6 TOXICITIES MEDIATED THROUGH THE NEUROENDOCRINE AXIS

Recently, attention has focused on the potential hazardous effects of certain chemicals on the endocrine system because of the ability of these chemicals to mimic or block endogenous hormones. Chemicals with this type of activity are most commonly referred to as *endocrine disruptors*. However, appropriate terminology to describe such effects remains controversial. The terminology *endocrine disruptors*, initially used by Thomas and Colborn (1992), was also used in 1996 when Congress mandated the EPA to develop a screening program for "...certain substances [which] may have an effect produced by a naturally occurring estrogen, or other such endocrine effect[s]...". To meet this mandate, EPA convened a panel called the Endocrine Disruptors Screening and Testing Advisory Committee (EDSTAC), and in 1998, the EDSTAC completed its deliberations and made recommendations to EPA concerning *endocrine disruptors*. In 1999, the National Academy of Sciences released a report that referred to these same types of chemicals as *hormonally active agents*. The terminology *endocrine modulators* has also been used to convey the fact that effects caused by such chemicals may not necessarily be adverse. Many scientists agree that chemicals with the ability to disrupt or modulate the endocrine system are a potential threat to the health of humans, aquatic animals, and wildlife. However, others think that endocrine-active chemicals do not pose a significant health risk, particularly in view of the fact that hormone mimics exist in the natural environment. Examples of natural hormone mimics are the isoflavinoid phytoestrogens (Adlercreutz 1995; Livingston 1978; Mayr et al. 1992). These chemicals are derived from plants and are similar in structure and action to endogenous estrogen. Although the public health significance and descriptive terminology of substances capable of affecting the endocrine system remains controversial, scientists agree that these chemicals may affect the synthesis, secretion, transport, binding, action, or elimination of natural hormones in the body responsible for maintaining homeostasis, reproduction, development, and/or behavior (EPA 1997h). Stated differently, such compounds may cause toxicities that are mediated through the neuroendocrine axis. As a result, these chemicals may play a role in altering, for example, metabolic, sexual, immune, and neurobehavioral function. Such chemicals are also thought to be involved in inducing breast, testicular, and prostate cancers, as well as endometriosis (Berger 1994; Giwercman et al. 1993; Hoel et al. 1992).

There is little evidence to suggest that arsenic functions as an endocrine disruptor. An association has been demonstrated between exposure to arsenic in drinking water and increased incidence of diabetes mellitus (Rahman et al. 1998; Tsai et al. 1999; Tseng et al. 2000; Wang et al. 2003), although dose-response relationships are not available and the mechanism of action for this response has not been characterized. Studies by Waalkes and coworkers (Waalkes et al. 2006a, 2006b) have suggested that in

mice, arsenic may interact with estrogens to enhance production of female urogenital cancers and male hepatocellular cancer following exposure to arsenic *in utero*. The mechanism by which this might happen has not been elucidated. No other relevant data were located in humans or animals. Data on general effects of arsenic compounds on the endocrine system are presented in Sections 3.2.1.2 and 3.2.2.2 above.

In vitro studies provide suggestive evidence that arsenic may act as an endocrine disruptor. Studies by Bodwell et al. (2004, 2006) and Davey et al. (2007) demonstrate that arsenic can alter gene regulation of steroid hormone receptors for glucocorticoids, mineralocorticoids, progesterone, and estrogen.

3.7 CHILDREN'S SUSCEPTIBILITY

This section discusses potential health effects from exposures during the period from conception to maturity at 18 years of age in humans, when all biological systems will have fully developed. Potential effects on offspring resulting from exposures of parental germ cells are considered, as well as any indirect effects on the fetus and neonate resulting from maternal exposure during gestation and lactation. Relevant animal and *in vitro* models are also discussed.

Children are not small adults. They differ from adults in their exposures and may differ in their susceptibility to hazardous chemicals. Children's unique physiology and behavior can influence the extent of their exposure. Exposures of children are discussed in Section 6.6, Exposures of Children.

Children sometimes differ from adults in their susceptibility to hazardous chemicals, but whether there is a difference depends on the chemical (Guzelian et al. 1992; NRC 1993). Children may be more or less susceptible than adults to health effects, and the relationship may change with developmental age (Guzelian et al. 1992; NRC 1993). Vulnerability often depends on developmental stage. There are critical periods of structural and functional development during both prenatal and postnatal life, and a particular structure or function will be most sensitive to disruption during its critical period(s). Damage may not be evident until a later stage of development. There are often differences in pharmacokinetics and metabolism between children and adults. For example, absorption may be different in neonates because of the immaturity of their gastrointestinal tract and their larger skin surface area in proportion to body weight (Morselli et al. 1980; NRC 1993); the gastrointestinal absorption of lead is greatest in infants and young children (Ziegler et al. 1978). Distribution of xenobiotics may be different; for example, infants have a larger proportion of their bodies as extracellular water, and their brains and livers are proportionately larger (Altman and Dittmer 1974; Fomon 1966; Fomon et al. 1982; Owen and Brozek

1966; Widdowson and Dickerson 1964). The infant also has an immature blood-brain barrier (Adinolfi 1985; Johanson 1980) and probably an immature blood-testis barrier (Setchell and Waites 1975). Many xenobiotic metabolizing enzymes have distinctive developmental patterns. At various stages of growth and development, levels of particular enzymes may be higher or lower than those of adults, and sometimes unique enzymes may exist at particular developmental stages (Komori et al. 1990; Leeder and Kearns 1997; NRC 1993; Vieira et al. 1996). Whether differences in xenobiotic metabolism make the child more or less susceptible also depends on whether the relevant enzymes are involved in activation of the parent compound to its toxic form or in detoxification. There may also be differences in excretion, particularly in newborns who all have a low glomerular filtration rate and have not developed efficient tubular secretion and resorption capacities (Altman and Dittmer 1974; NRC 1993; West et al. 1948). Children and adults may differ in their capacity to repair damage from chemical insults. Children also have a longer remaining lifetime in which to express damage from chemicals; this potential is particularly relevant to cancer.

Certain characteristics of the developing human may increase exposure or susceptibility, whereas others may decrease susceptibility to the same chemical. For example, although infants breathe more air per kilogram of body weight than adults breathe, this difference might be somewhat counterbalanced by their alveoli being less developed, which results in a disproportionately smaller surface area for alveolar absorption (NRC 1993).

Arsenic has been recognized as a human toxicant for many centuries, and the symptoms of acute poisoning are well known. Children who are exposed to high levels of arsenic exhibit symptoms similar to those seen in adults, including respiratory, cardiovascular, dermal, and neurological effects, and vomiting if the arsenic is ingested (Borgoño et al. 1980; Foy et al. 1992; Kersjes et al. 1987; Muzi et al. 2001; Rosenberg 1974; Zaldívar 1974; Zaldívar and Guillier 1977). Arterial thickening of the pancreas was observed in five children who died in Chile after chronic exposure to arsenic (Rosenberg 1974). Foy et al. (1992) described systemic effects of chronic arsenic exposure in children in a village near a tin and tungsten mining operation in Thailand. The arsenic concentration in water samples from 35 shallow wells averaged 0.82 mg As/L (range, 0.02–2.7 mg As/L). Piped water (available in some homes) had a concentration of 0.07 mg As/L. A survey of skin manifestations of arsenic poisonings was conducted in the autumn of 1987. The case reports of four children were presented. All of the children had hyperkeratosis and hyperpigmentation of the extremities, including tibia, palms, and soles. In addition, one child had developed weakness 3 years previously and had anorexia and a chronic cough for 1 year. She had been held back twice in school as a slow learner. On examination, she had a runny nose and

weakness of her wrist joints. The liver was about 4 finger-breadths below the right costal margin with a sharp but tender edge. Blood arsenic levels ranged from 0.087 to 0.46 μg/mL and the arsenic level in hair ranged from 14.4 to 20 μg/g. The authors concluded that the finding of typical skin manifestations of chronic arsenic poisoning suggests that it may take a considerably shorter period of time to develop these manifestations than previously thought. However, it is not known what effect co-exposure to tin and tungsten might have had on skin manifestations in these children. Exposure to high arsenic levels during gestation and/or during early childhood also was associated with significant increases in SMRs for lung cancer and bronchiectasis during adulthood in a study of residents in a city in Chile with high arsenic levels in the drinking water (near 0.9 mg/L) during several years (Smith et al. 2006).

As previously mentioned in Sections 3.2.1.4 and 3.2.2.4, exposure of children to arsenic also has been associated with neurological deficits in children. Studies by Wasserman et al. (2004, 2007) of 6- and 10-year-old children from Bangladesh reported small but significant decreases in some tests of cognitive function associated with levels of arsenic in the water ≥0.05 mg/L. A study of pre-school age children in West Bengal, India, reported an association between current urinary arsenic concentrations, but not long-term water arsenic, and small decrements in intellectual tests (von Ehrenstein et al. 2007). Similar results were reported in a study of children in Taiwan (Tsai et al. 2003) and in China (Wang et al. 2007). Neurological effects have also been associated with elevated levels of arsenic in the air. For example, Bencko et al. (1977) reported that children of approximately 10 years of age living near a power plant burning coal of high arsenic content showed significant hearing losses (increased threshold) compared to a control group of children living outside the polluted area (Bencko et al. 1977). Also, in a study of Mexican children, Calderón et al. (2001) reported that children living near a smelter complex had poor performance on tests evaluating verbal IQ than children who lived farther from the smelter. Thus, the limited data available suggest that exposure of children to inorganic arsenic may result in detrimental effects on neurobehavioral parameters.

Wulff et al. (1996) conducted a retrospective study of a cohort of children born between 1961 and 1990 in the municipality of Skelleftea, Sweden, where a smelter released arsenic and other pollutants including lead, copper, cadmium, and sulfur dioxide. Childhood cancer incidences among children born in the vicinity of the smelter (i.e., within 20 km) and distant from the smelter (>20 km) were compared with expected incidences based on Swedish national statistics. There appeared to be an increased risk of childhood cancer (all types combined) among children born in the vicinity of the smelter (SIR=195, 95% CI=88–300, based on 13 cases observed and 6.7 expected), but the increase was not statistically significant, and in any event, the role of arsenic in any finding from this study is confounded by the

presence of other metals. The number of cases (n=42) was very close to the expected number (n=41.8) among children born distant from the smelter. Similar results were reported in a study by Moore et al. (2002), which did not find increased incidence ratios for all childhood cancers or for childhood leukemias in children from an area of Nevada with high arsenic exposures.

Inorganic arsenic has been characterized as a developmental toxicant. It is known to cross the placental barrier and selectively accumulate in the neuroepithelium of the developing animal embryo (Hanlon and Ferm 1977; Lindgren et al. 1984). Studies in animals have also revealed that various fetal malformations occur after embryonic exposure to arsenic *in vitro*; neural tube defects are the predominant and consistent malformation in these studies (Chaineau et al. 1990; Mirkes and Cornel 1992; Morrissey and Mottet 1983; Mottet and Ferm 1983; Tabacova et al. 1996; Willhite and Ferm 1984; Wlodarczyk et al. 1996). *In vivo* studies have shown that high doses of ingested arsenic can produce developmental effects (fetal mortality, skeletal defects), but generally only at maternally toxic doses (Baxley et al. 1981; Holson et al. 1999, 2000; Hood and Harrison 1982; Hood et al. 1978; Nemec et al. 1998; Stump et al. 1999). A series of studies showed an increased incidence of tumors in the offspring of mice exposed to arsenic from gestational day 8 through day 18(Waalkes et al. 2003, 2004a, 2004b, 2004c, 2006a, 2006b) (see Section 3.2.2.6 for further details). In humans, acute prenatal exposure to high doses of inorganic arsenic can result in miscarriage and early neonatal death (Bolliger et al. 1992; Lugo et al. 1969). Although several studies have reported marginal associations between prolonged low-dose human arsenic exposure and adverse reproductive outcomes, including spontaneous abortion, stillbirth, developmental impairment, and congenital malformation (Ahmad et al. 2001; Aschengrau et al. 1989; Chakraborti et al. 2003c; Hopenhayn-Rich et al. 2000; Nordström et al. 1978a, 1979b; Yang et al. 2003; Zierler et al. 1988), none of these studies have provided convincing evidence for such effects or information concerning possible dose-response relationships.

There is no evidence for differences in absorption of arsenic in children and adults. Ingestion of arsenic in dirt may be an important route of exposure for young children. A study that used a synthetic gastric juice designed to mimic gastric conditions in a 2-year-old child found that absorption of arsenic from contaminated soil was likely to be up to 5 times lower than the total concentration of arsenic in the soil (Williams et al. 1998). As previously mentioned, arsenic crosses the placenta and preferentially accumulates in the embryonic neuroepithelium. In addition, arsenic is known to be present in breast milk at low concentrations. Arsenic concentrations were low in human milk sampled from 88 mothers in the Faroe Islands (0.0001–0.0044 ppm), where the diet is predominantly seafood (exposures were primarily to “fish arsenic” [Grandjean et al. 1995]), in a population of Andean women (0.0008–0.008 ppm) exposed

to high concentrations of inorganic arsenic in drinking water (Concha et al. 1998b), and in a World Health Organization survey (0.00013–0.00082 ppm) (Somogyi and Beck 1993). There is no information in the literature describing storage of arsenic in maternal tissues. There is some evidence that metabolism of arsenic in children is less efficient than in adults. Children in two villages in Argentina ingesting large amounts of arsenic in their drinking water (200 µg/L) excreted about 49% inorganic arsenic and 47% DMA, compared to 32% inorganic arsenic and 66% DMA for the women in the study (Concha et al. 1998b). No PBPK models specifically targeted at fetuses, infants, or children, or pregnant or lactating women were found in the literature. There are no biomarkers that have been specifically identified for children exposed to arsenic. In addition, no unique interactions of arsenic with other chemicals have been identified in children.

The mechanism of toxic action of arsenic in the mammalian cell may involve inhibition of proliferation of cells (Dong and Luo 1993; Jha et al. 1992; Petres et al. 1977). In addition, high-dose arsenic impairs assembly and disassembly of microtubules, thus interfering with mitotic spindle formation and embryonal cell division (Léonard and Lauwerys 1980; Li and Chou 1992; Mottet and Ferm 1983). Arsenic compounds also cause chromosomal aberrations (Jha et al. 1992; Léonard and Lauwerys 1980), which may disrupt cell cycling. The direct toxic effects of high levels of arsenic in the developing embryo result not from a difference in the mechanism of toxicity during development, but rather from the existence of a unique target tissue, the neuroepithelium. The process of neurulation involves cell shape changes, cytokinesis, and cell adhesion, which are dependent upon cytoskeletal elements that are functionally affected by arsenic (Dallaire and Béliveau 1992; Edelman 1992; Gunn et al. 1992; Li and Chou 1992; Moriss-Kay et al. 1994; Schoenwolf and Smith 1990; Taubeneck et al. 1994). However, since arsenic is known to affect vasculature, and since altered placental and/or embryonal vasculature has been suggested as a mechanism leading to neural tube defects, the embryo may be sensitive to this manifestation of arsenic toxicity.

3.8 BIOMARKERS OF EXPOSURE AND EFFECT

Biomarkers are broadly defined as indicators signaling events in biologic systems or samples. They have been classified as markers of exposure, markers of effect, and markers of susceptibility (NAS/NRC 1989).

Due to a nascent understanding of the use and interpretation of biomarkers, implementation of biomarkers as tools of exposure in the general population is very limited. A biomarker of exposure is a xenobiotic

substance or its metabolite(s) or the product of an interaction between a xenobiotic agent and some target molecule(s) or cell(s) that is measured within a compartment of an organism (NAS/NRC 1989). The preferred biomarkers of exposure are generally the substance itself, substance-specific metabolites in readily obtainable body fluid(s), or excreta. However, several factors can confound the use and interpretation of biomarkers of exposure. The body burden of a substance may be the result of exposures from more than one source. The substance being measured may be a metabolite of another xenobiotic substance (e.g., high urinary levels of phenol can result from exposure to several different aromatic compounds). Depending on the properties of the substance (e.g., biologic half-life) and environmental conditions (e.g., duration and route of exposure), the substance and all of its metabolites may have left the body by the time samples can be taken. It may be difficult to identify individuals exposed to hazardous substances that are commonly found in body tissues and fluids (e.g., essential mineral nutrients such as copper, zinc, and selenium). Biomarkers of exposure to arsenic are discussed in Section 3.8.1.

Biomarkers of effect are defined as any measurable biochemical, physiologic, or other alteration within an organism that, depending on magnitude, can be recognized as an established or potential health impairment or disease (NAS/NRC 1989). This definition encompasses biochemical or cellular signals of tissue dysfunction (e.g., increased liver enzyme activity or pathologic changes in female genital epithelial cells), as well as physiologic signs of dysfunction such as increased blood pressure or decreased lung capacity. Note that these markers are not often substance specific. They also may not be directly adverse, but can indicate potential health impairment (e.g., DNA adducts). Biomarkers of effects caused by arsenic are discussed in Section 3.8.2.

A biomarker of susceptibility is an indicator of an inherent or acquired limitation of an organism's ability to respond to the challenge of exposure to a specific xenobiotic substance. It can be an intrinsic genetic or other characteristic or a preexisting disease that results in an increase in absorbed dose, a decrease in the biologically effective dose, or a target tissue response. If biomarkers of susceptibility exist, they are discussed in Section 3.10, Populations That Are Unusually Susceptible.

3.8.1 Biomarkers Used to Identify or Quantify Exposure to Arsenic

Arsenic levels in blood, urine, hair, and nails have all been investigated and used as biological indicators of exposure to arsenic. Since arsenic is cleared from blood within a few hours (Tam et al. 1979b; Vahter 1983), measurements of blood arsenic reflect exposures only within the very recent past. Typical values in nonexposed individuals are <1 μg/L (Heydorn 1970; Hindmarsh and McCurdy 1986; Valentine et al.

1979). Consumption of medicines containing arsenic is associated with blood values of 100–250 µg/L, while blood levels in acutely toxic and fatal cases may be 1,000 µg/L or higher (Driesback 1980).

However, blood levels do not appear to be reliable indicators of chronic exposure to low levels of arsenic. For example, there was no correlation between the level of arsenic in blood of residents and the level of arsenic in drinking water in several U.S. communities where water levels ranged from about 6 to 125 µg/L (Valentine et al. 1979, 1981). Consequently, measurement of blood arsenic is not generally considered to be a reliable means of monitoring human populations for arsenic exposure.

As discussed in Section 3.4.4, most arsenic that is absorbed from the lungs or the gastrointestinal tract is excreted in the urine, mainly within 1–2 days. For this reason, measurement of urinary arsenic levels is generally accepted as the most reliable indicator of recent arsenic exposure, and this approach has proved useful in identifying above-average exposures in populations living near industrial point sources of arsenic (e.g., Milham and Strong 1974; Polissar et al. 1990). By the inhalation route, several researchers have found that there is a good quantitative correlation between the concentration of arsenic in workplace air (C_{air}, µg/m^3) and the concentration in the urine (C_{urine}, µg/L) of exposed workers. For example, Pinto et al. (1976) found a linear relationship for exposures ranging up to 150 µg/m^3, given by the following equation:

$$C_{air}=0.3\ C_{urine}$$

Enterline et al. (1987a) reinvestigated this relationship over a wider range of exposures (up to 3,500 µg/m^3), and found that the curve tended to be concave upward, as given by the following equation:

$$C_{air}=0.0064\ (C_{urine})^{1.94}$$

This indicates that at higher exposure levels, a higher fraction of the dose is excreted in urine, although the toxicokinetic basis for this is not certain. Numerous studies have used above-average urinary levels (i.e., higher than about 100 µg/L) as evidence of recent arsenic ingestion (e.g., Borgoño et al. 1980; Fincher and Koerker 1987; Franzblau and Lilis 1989; Goldsmith and From 1986; Kyle and Pease 1965; Valentine et al. 1981). Calderon et al. (1999) found a quantitative correlation between the log of the mean total urinary arsenic concentration/creatinine (TAs/c, µg/mg) of people living in areas with arsenic-contaminated drinking water sources and the log of the inorganic arsenic concentration in the drinking water (InAs, µg/L). The equation for the regression line is:

$$TAs/c=10^{-2.57} \times (InAs)^{0.63}$$

where -2.57 and 0.63 are the intercept and slope, respectively, for the regression of the log10-transformed data. Mixed model regression analysis showed that the log of estimated arsenic intake from drinking water (µg/day) is also a good predictor of TAs/c excretion (Calderon et al. 1999).

There is some indication that speciation of urinary arsenic may indicate the extent of past cumulative exposure to arsenic. Hsueh et al. (1998a) reported higher levels of DMA and MMA in the urine of individuals with higher cumulative past exposure to inorganic arsenic. Speciated urinary arsenic is also a recommended biomarker for recent inorganic arsenic exposure. Walker and Griffin (1998) used the EPA Exposure Assessment Model and a number of site-specific data covering environmental and biological factors to predict total and speciated urinary arsenic concentrations for children living near high levels of arsenic-contaminated soil. There was reasonable agreement between the measured and predicted speciated urinary arsenic concentrations.

An important limitation to the use of total urinary arsenic as a biomarker of exposure is that arsenobetaine is excreted (unmetabolized) in urine after ingestion of certain seafoods (Brown et al. 1990; Kalman 1987; Tam et al. 1982). Since "fish arsenic" is essentially nontoxic, analytical methods based on total urinary arsenic content may overestimate exposures to arsenic species that are of health concern. As discussed in Section 7.1, there are adequate methods for distinguishing arsenobetaine from other forms of arsenic in urine (inorganic, MMA, DMA), although these are not convenient to use as a routine screening method.

Arsenic tends to accumulate in hair and nails, and measurement of arsenic levels in these tissues may be a useful indicator of past exposures. Normal levels in hair and nails are 1 ppm or less (Choucair and Ajax 1988; Franzblau and Lilis 1989). These values may increase from several-fold to over 100-fold following arsenic exposure (Agahian et al. 1990; Bencko 2005; Bencko et al. 1986; de Peyster and Silvers 1995; EPA 1977a, 1981b; Karagas et al. 1996; Milham and Strong 1974; Valentine et al. 1979; Yamauchi et al. 1989) and remain elevated for 6–12 months (Choucair and Ajax 1988). Minimum exposure levels that produce measurable increases in arsenic levels in hair and nails have not been precisely defined. For hair, ingestion of 50–120 ppb of arsenic in drinking water produced only a marginal effect, but a clear increase was noted at 393 ppb (Valentine et al. 1979). A study of children living in a region polluted with arsenic derived from a power plant burning coal with a high arsenic content found a significant correlation between arsenic levels in hair and distance from the source of emission (Bencko and Symon 1977).

Inhalation exposure of workers to about 0.6 μg/m^3 of arsenic in air significantly increased average levels in nails (Agahian et al. 1990), although there was wide variation between individuals.

Analysis of hair may yield misleading results due to the presence of arsenic adsorbed to the external surface, but this can be minimized by collecting samples from close to the scalp or from unexposed areas and by washing the hair before analysis (e.g., Paschal et al. 1989). Similarly, extensive washing of nails is required to remove exogenous contamination (Agahian et al. 1990). The relationship between consumption of food items and levels of arsenic in toenails has been evaluated by MacIntosh et al. (1997) using standard multivariate regression models. This approach does not appear to be highly reliable, but may be sufficient for exploring associations between diet and disease. Kurttio et al. (1998) used linear regression models to show that there is a good association between arsenic concentration in hair (mg/kg) and total arsenic concentration in urine (μg/L), arsenic concentration in drinking water (μg/L) or daily intake of arsenic (μg/day). A 10 μg/L increase in the drinking water concentration or a 10–20 μg/day increase in daily arsenic intake corresponded to a 0.1 mg/kg increase in the arsenic concentration in hair. It is also important to note that the measurement of arsenic in hair and fingernails is a process not readily accessible to many clinical offices.

3.8.2 Biomarkers Used to Characterize Effects Caused by Arsenic

As discussed in Section 3.2, the characteristic pattern of skin changes caused by arsenic (hyperkeratinization, hyperpigmentation) is probably the most sensitive and diagnostic clinical indicator of chronic exposure to arsenic. However, no means has been developed for detecting these effects except by routine dermatological examination.

Peripheral neuropathy is another characteristic effect of arsenic exposure, and several researchers have investigated decreased nerve conduction velocity or amplitude as a biomarker for peripheral neuropathy. While effects can usually be detected in individuals with clinical signs of neuropathy (e.g., Goebel et al. 1990; Jenkins 1966; Le Quesne and McLeod 1977; Morton and Caron 1989; Murphy et al. 1981), effects are only marginal (EPA 1977a; Hindmarsh et al. 1977; Valentine et al. 1981) or undetectable (EPA 1981b; Kreiss et al. 1983) in exposed populations without obvious clinical signs of toxicity. This indicates that this approach is probably not sufficiently sensitive to detect neurological effects earlier than by standard neurological examination (Hindmarsh and McCurdy 1986). Also, decreases in nerve conduction velocity or amplitude are not specific for arsenic-induced neuropathy.

Arsenic is known to affect the activity of a number of enzymes, and some of these may have potential as biomarkers of effect. Most promising is the spectrum of effects caused by arsenic on the group of enzymes responsible for heme synthesis and degradation, including inhibition of coproporphyrinogen oxidase and heme synthetase (Woods and Fowler 1978; Woods and Southern 1989) and activation of heme oxygenase (Sardana et al. 1981). Menzel et al. (1998) has examined the *in vitro* induction of human lymphocyte heme oxygenase 1(HO1) as a biomarker of arsenite exposure. Arsenite did induce *de novo* synthesis of HO1 in human lymphoblastoid cells, but it has not been determined if the same response is induced *in vivo*. It has been shown in animals that these arsenic-induced enzymic changes result in increased urinary levels of uroporphyrin, coproporphyrin, and bilirubin (Albores et al. 1989; Woods and Fowler 1978), and it has been shown that these effects can be detected in the urine of arsenic-exposed humans (García-Vargas and Hernández-Zavala 1996). Therefore, altered urinary levels of these heme-related compounds could serve as a biomarker of effect. However, it is known that numerous other toxic metals also have similar effects on heme metabolism (Albores et al. 1989; Sardana et al. 1981; Woods and Southern 1989), so it is likely that these effects would not be specific for arsenic.

For more information on biomarkers for renal and hepatic effects of chemicals, see ATSDR/CDC Subcommittee Report on Biological Indicators of Organ Damage (Agency for Toxic Substances and Disease Registry 1990b) and for information on biomarkers for neurological effects, see OTA (1990).

3.9 INTERACTIONS WITH OTHER CHEMICALS

A number of researchers have found that arsenic compounds tend to reduce the effects of selenium (Hill 1975; Howell and Hill 1978; Kraus and Ganther 1989; Levander 1977; Miyazaki et al. 2003; Moxon et al. 1945; Schrauzer 1987; Schrauzer et al. 1978). Likewise, selenium can decrease the effects of arsenic, including clastogenicity (Beckman and Nordenson 1986; Biswas et al. 1999; Sweins 1983), delayed mutagenesis (Rossman and Uddin 2004), cocarcinogenesis (Uddin et al. 2005), cytotoxicity (Babich et al. 1989; Rössner et al. 1977; Styblo and Thomas 2001), and teratogenicity (Holmberg and Ferm 1969). The mechanism of this mutual inhibition of effects is not known, but may be related to the formation of a selenium-arsenic complex (seleno-bis [S-gluthionyl] arsinium ion; Gailer et al. 2002) that is excreted more rapidly than either arsenic or selenium alone (Cikrt et al. 1988; Hill 1975; Levander 1977; Levander and Baumann 1966) or due to selenium-induced changes in arsenic methylation (Styblo and Thomas 2001; Walton et al. 2003). There is little direct evidence that variations in selenium exposure in humans lead to significant increases or decreases in arsenic toxicity, although copper smelter workers who developed lung cancer had lower tissue levels of selenium than workers who did not develop lung tumors

(Gerhardsson et al. 1985, 1988). This suggests that selenium deficiency could significantly increase the risk of lung cancer following inhalation exposure to arsenic, but it is difficult to distinguish cause from effect in such a study. However, there is evidence that administration of selene can facilitate recovery from arsenic poisoning. In residents living in an area of Inner Mongolia with high levels of arsenic in drinking water, administration of 100–200 µg selenium/day in the form of selenium yeast and exposure to arsenic-free water for 14 months resulted in a greater improvement in clinical signs and symptoms, liver function, and EKG readings as compared to residents administered arsenic-free water only (Wuyi et al. 2001; Yang et al. 2002). An improvement in skin lesions was observed in 67 and 21% of the subjects in the selenium-supplemented and control groups (Yang et al. 2002). Additionally, the levels of arsenic in blood, hair, and urine were significantly lower after the 14-month period only in the selenium supplemented group.

The interaction between cigarette smoking, inhalation of arsenic, and the risk of lung cancer has not been extensively investigated. Smoking appeared to increase lung cancer risk synergistically (multiplicatively) in one study of smelter workers (Pershagen et al. 1981), although the data are not adequate to exclude a simple additive interaction (Thomas and Whittemore 1988). Cigarette smoking has been shown to increase the occurrence of lung cancer in people with high levels of arsenic in the drinking water (Chiou et al. 1995; Tsuda et al. 1995a). Suggestive evidence of a positive interaction between arsenic and benzo(a)pyrene has also been noted for induction of lung adenocarcinomas in hamsters (Pershagen et al. 1984).

Co-exposure to ethanol and arsenic may exacerbate the toxic effects of arsenic. Simultaneous exposure of rats to ethanol (10% in drinking water) and arsenic (dose not stated) for 6 weeks produced a significant increase in the concentration of arsenic in the kidney, a nonsignificant increase of arsenic in the liver and a significant increase in the concentration of glutathione in the liver, compared to rats treated with either ethanol or arsenic alone (Flora et al. 1997a, 1997b). Histological damage to the liver, but not the kidneys, was increased in rats treated with both ethanol and arsenic compared to those receiving only arsenic.

Studies of rats exposed to arsenic, lead, and cadmium, alone or in combination, have revealed mainly additive or subadditive effects on body weight, hematological parameters, and enzymes of heme synthesis (Mahaffey and Fowler 1977; Mahaffey et al. 1981). Similarly, studies of the tissue levels of arsenic in rats fed arsenic with or without lead or cadmium revealed only limited evidence of any toxicokinetic interactions (Mahaffey et al. 1981). Pretreatment of rats with a nontoxic dose of cadmium had no effect on the lethality of a high dose of arsenic and did not reduce arsenic-induced hepatotoxicity (Hochadel and

Waalkes 1997). These data do not suggest that arsenic toxicity is likely to be significantly influenced by concomitant exposure to these metals. However, supplementation with zinc or chromium may be useful in reducing chronic arsenism. Arsenic has been shown to cause an increase in total plasma cholesterol; co-administration of chromium(III) counteracts this effect (Aguilar et al. 1997). Pretreatment of mice with zinc, at least 24 hours before injection with arsenic-73, reduced arsenic retention compared to controls that did not receive the zinc pretreatment or received it only a short time before the administration of arsenic (Kreppel et al. 1994). Zinc is an inducer of metallothionein, but this induction does not appear to be the mechanism that reduces arsenic toxicity because other inducers of metallothionein did not reduce arsenic toxicity and arsenic elimination was increased by the zinc pretreatment.

Since methylation of arsenic is a detoxification mechanism, it is possible that chemicals that interfere with the methylation process could increase toxicity. This is supported by studies in animals in which reagents that inhibit methylation enzymes (e.g., periodate-oxidized adenosine) caused an increase in tissue levels of inorganic arsenic (Marafante and Vahter 1986; Marafante et al. 1985). Similarly, cellular glutathione levels appear to play a role in the methylation process, and treatment with reagents (e.g., phorone) that decrease glutathione levels increases arsenic toxicity (Buchet and Lauwerys 1987). Inadequate dietary intake of methionine, choline, or protein may also exacerbate arsenic toxicity. Rabbits pretreated with diets low in choline, methionine, or protein showed a significant increase in tissue retention of arsenic and a significant decrease in the excretion of dimethylarsinic acid (Vahter and Marafante 1987). The increased retention of arsenic in rabbits fed these deficient diets is likely to be due to a reduction in arsenic methylation. Thus, the toxic effects of chronic arsenic ingestion may be increased in populations that are also subject to malnutrition.

3.10 POPULATIONS THAT ARE UNUSUALLY SUSCEPTIBLE

A susceptible population will exhibit a different or enhanced response to arsenic than will most persons exposed to the same level of arsenic in the environment. Reasons may include genetic makeup, age, health and nutritional status, and exposure to other toxic substances (e.g., cigarette smoke). These parameters result in reduced detoxification or excretion of arsenic, or compromised function of organs affected by arsenic. Populations who are at greater risk due to their unusually high exposure to arsenic are discussed in Section 6.7, Populations with Potentially High Exposures.

No studies were located that identified an unusual susceptibility of any human subpopulation to arsenic. Several studies have evaluated possible sex-related differences in arsenic toxicity and carcinogenesis (Aposhian et al. 2000a, 2000b; Calderon et al. 1999; Loffredo et al. 2003; Mandal et al. 2001; Watanabe et al. 2001), but have not consistently identified differences. However, since the degree of arsenic toxicity may be influenced by the rate and extent of its methylation in the liver (see Section 3.4.3), it seems likely that some members of the population might be especially susceptible because of lower than normal methylating capacity. Studies of exposed humans in Taiwan suggested that subjects with lower secondary methylation indices have an increased risk of bladder cancer (Chen et al. 2003) and peripheral vascular disease (Tseng et al. 2005), particularly in subjects with high exposure levels. Reduced hepatic methylation could result from dietary deficiency of methyl donors such as choline or methionine (Buchet and Lauwerys 1987; Vahter and Marafante 1987), although this is unlikely to be a concern for most people in the United States. There is evidence that methylation capacity can vary greatly among individuals (e.g., Buchet et al. 1981a; Foà et al. 1984; Hopenhayn-Rich et al. 1996b; Tam et al. 1979b), but the basis of this variation and its impact on human susceptibility have not been fully established. There is some evidence that low dietary protein intake and possibly other nutritional deficiencies can decrease arsenic methylation (Steinmaus et al. 2005a). Recently, Heck et al. (2007) examined whether the capacity to methylate arsenic differs by nutrient intake in a cohort of 1,016 Bangladeshi adults exposed to arsenic in drinking water. The results showed that higher intakes of cysteine, methionine, calcium, protein, and vitamin B-12 were associated with lower percentages of inorganic arsenic and higher ratios of MMA to inorganic arsenic in urine. In addition, higher intakes of niacin and choline were associated with higher DMA/MMA ratios, after adjustment for sex, age, smoking, total urinary arsenic, and total energy intake. The issue of increased susceptibility to arsenic due to poor nutrition was discussed by NRC (2001), it was concluded that, with regard to skin effects, studies of cohorts from India, Bangladesh, and Taiwan suggest that nutrition plays an important role in arsenic toxicity. On the other hand, studies in other regions of the world (i.e., Chile) involving populations with much better nutrition argue against poor nutrition having a major impact on arsenic toxicity.

Various genetic polymorphisms also seem to play a role in arsenic-induced toxicity. For example, a study of 85 lung cancer patients and 108 healthy controls in northern Chile reported that there was a nonstatistically significant difference for the frequency of the GSTM1 null genotype between the healthy and lung cancer patients stratified by gender and smoking status. The same results were observed for the MspI CYP450 1A1 polymorphism (Adonis et al. 2005). Hsueh et al. (2005) examined the association of four polymorphisms: NAD(P)H oxidase, manganese superoxide dismutase (MnSOD), catalase, and endothelial nitric oxide synthase (eNOS) with arsenic related hypertension risk among 79 hypertensive

cases and 213 controls in an arseniasis-hyperendemic area in Taiwan. The results showed that MnSOD polymorphism significantly increased the risk of hypertension regardless of exposure to arsenic. NAD(P)H oxidase and eNOS polymorphisms were significantly associated with increased risk of hypertension in subjects with higher cumulative arsenic exposure (≥10.5 mg/L x year), whereas catalase polymorphism was not associated with hypertension. The results also showed that the association between MnSOD, NAD(P)H oxidase, and eNOS polymorphisms and risk of hypertension were more pronounced in subjects with high triglyceride level. A study of a population of West Bengal, India, exposed to arsenic via drinking water reported that the frequencies of null genotype in GSTT1 were 13.52 and 12.92% in skin-symptomatic and skin-asymptomatic individuals, and GSTM1 null genotype were 13.90 and 22.47% in skin-symptomatic and skin-asymptomatic individuals, respectively (Ghosh et al. 2006). Compared to those with GSTM1 null genotype, subjects with GST1-positive (at least one allele) had significantly higher risk of arsenic-induced skin lesions. Recently, Steinmaus et al. (2007) investigated urinary arsenic methylation patterns and genetic polymorphisms in methylenetetrahydro-folate reductase (MTHFR) and GST in 170 subjects (139 males) from an arsenic-exposed region in Argentina. MTHFR is a key enzyme in the metabolism of folate and has been linked to arsenic metabolism and toxicity (NRC 1999). Steinmaus et al. (2007) found that subjects with the TT/AA variant of MTHFR 677/1298 (associated with lower MTHFR activity) excreted a significantly higher proportion on ingested arsenic as inorganic arsenic and a smaller proportion as DMA(V). The study also reported that women with null genotype of GSTM1 excreted a significantly higher proportion of arsenic as monomethylarsenate than women with the active genotype. The study also found no association between polymorphisms in GSTT1 and arsenic methylation.

There is a report that described severe arsenic-induced neuropathy that developed only in a 5,10-methylenetetrahydrofolate-reductase (MTHFR) deficient member of a family that had been exposed to arsenic (Brouwer et al. 1992). The authors suggest that the MTHFR deficiency in this girl might explain the fact that of all the family members exposed to arsenic, only she developed severe clinical signs of arsenic poisoning. Liver disease does not appear to decrease methylation capacity in humans, at least at low levels of arsenic exposure (Buchet et al. 1982; Geubel et al. 1988).

3.11 METHODS FOR REDUCING TOXIC EFFECTS

This section will describe clinical practice and research concerning methods for reducing toxic effects of exposure to arsenic. However, because some of the treatments discussed may be experimental and unproven, this section should not be used as a guide for treatment of exposures to arsenic. When specific

exposures have occurred, poison control centers and medical toxicologists should be consulted for medical advice. The following texts provide specific information about treatment following exposures to arsenic:

Tintinalli JE, Ruiz E, Krone RL, eds. 1996. Emergency medicine. A comprehensive study. American College of Emergency Physicians. 4th ed. New York, NY: The McGraw-Hill Companies, Inc.

Goldfrank RL, Flomenbaum NE, Lewin NA, et al., eds. 1998. Goldfrank's toxicologic emergencies. 6th ed. Stamford, CT: Appleton and Lange.

Ellenhorn MJ. 1997. Ellenhorn's medical toxicology. Diagnosis and treatment of human poisoning. Baltimore, MD: Williams & Wilkins.

3.11.1 Reducing Peak Absorption Following Exposure

No data were located regarding the reduction of absorption after inhalation exposure to arsenic.

There are a number of methods for reducing absorption of arsenic following oral exposure. In cases of acute high-dose exposure, the removal of arsenic from the gastrointestinal tract may be facilitated by gastric lavage, stomach intubation, induced emesis, or use of cathartics (saline, sorbitol) within a few hours after ingestion (Agency for Toxic Substances and Disease Registry 1990a; Aposhian and Aposhian 1989; Campbell and Alvarez 1989; Driesback 1980; Ellenhorn and Barceloux 1988; EPA 1989e; Haddad and Winchester 1990; Kamijo et al. 1998; Stutz and Janusz 1988). However, the efficacy of several of these methods has been questioned by some authors, and in some cases, the treatments may be contraindicated. For example, vomiting and diarrhea often occur soon after ingesting arsenic, and therefore, use of an emetic or cathartic may not be necessary. Also, emesis should not be induced in obtunded, comatose, or convulsing patients (Campbell and Alvarez 1989; Ellenhorn and Barceloux 1988; EPA 1989e), and saline cathartics should be used with caution in patients with impaired renal function (Campbell and Alvarez 1989). Vantroyen et al. (2004) described a case of a massive arsenic trioxide overdose that was successfully treated by continuous gastric irrigation with sodium bicarbonate, forced diuresis, and administration of BAL and DMSA. Treatments of this sort are unlikely to be required following low-level exposures.

Another possible approach for reducing absorption following oral exposure is to administer substances that bind the arsenic in the gastrointestinal tract. For example, activated charcoal is sometimes used for this purpose (Campbell and Alvarez 1989; EPA 1989e; Stutz and Janusz 1988), although the effectiveness of this treatment is not well established. Because pentavalent arsenic is a phosphate analogue,

administration of phosphate-binding substance such as aluminum hydroxide might possibly be useful, but this has not been investigated. Sulfhydryl compounds might be given to bind trivalent arsenic, but it seems unlikely that these would be effective under the acid conditions in the stomach, and it is not clear that such complexes would have reduced gastrointestinal absorption.

Following dermal or ocular exposure to arsenic, several measures can be taken to minimize absorption. All contaminated clothing should be removed, and contacted skin should be immediately washed with soap and water. Eyes that have come in contact with arsenic should be flushed with copious amounts of clean water (EPA 1989e; Stutz and Janusz 1988).

3.11.2 Reducing Body Burden

Acute arsenic intoxication may require treatment with chelating agents such as dimercaprol (BAL) and D-penicillamine. Although body burden is not necessarily reduced, these chelators bind free arsenic and serve to reduce the body's pool of biologically active arsenic. Chelation therapy is most effective when instituted within a few hours after exposure, and efficacy decreases as time after exposure increases (Agency for Toxic Substances and Disease Registry 1990a; Kamijo et al. 1998; McFall et al. 1998; Peterson and Rumack 1977).

In general, chelating agents should be used with caution, since they may have serious side effects such as pain, fever, hypotension, and nephrotoxicity (Ellenhorn and Barceloux 1988). Some water-soluble and less toxic analogues of BAL such as dimercaptosuccinic acid (DMSA), dimercaptopropyl phthalamadic acid (DMPA), and dimercaptopropane sulfonic acid (DMPS) are currently under investigation and may prove to be promising treatments for arsenic poisoning (Agency for Toxic Substances and Disease Registry 1990a; Aposhian and Aposhian 1989; Aposhian et al. 1997; Guha Mazumder 1996; Kreppel et al. 1995). However, a randomized placebo trial of 2,3-dimercaptosuccinic acid as a therapy for chronic arsenosis due to drinking contaminated water found no significant difference between patients treated with 2,3-dimercaptosuccinic acid and those treated with a placebo (Guha Mazumder et al. 1998a). N-acetylcysteine has been used in animals to chelate arsenic (Haddad and Winchester 1990), and a human case study reported N-acetylcysteine to be successful in treating a case of arsenic poisoning that was not responding well to BAL treatment (Martin et al. 1990). Vantroyen et al. (2004) described a case of a massive arsenic trioxide overdose that was successfully treated by continuous gastric irrigation with sodium bicarbonate, forced diuresis, and administration of BAL and DMSA.

As discussed in Section 3.4.3, once arsenic has been absorbed into the blood stream, it undergoes methylation to yield MMA and DMA. These forms of arsenic are less toxic than inorganic arsenic and are cleared from the body by excretion in the urine. Therefore, if it were possible to enhance arsenic methylation, both body burden and toxicity of arsenic might be reduced. However, experimental evidence in animals and humans suggests that arsenic methylation is not enhanced to any significant degree by supplementation with methylation cofactors (Buchet and Lauwerys 1987; Buchet et al. 1982), presumably because it is enzyme level and not cofactor availability that is rate limiting in arsenic methylation.

3.11.3 Interfering with the Mechanism of Action for Toxic Effects

It is generally thought that trivalent arsenic exerts its toxic effects mainly by complexing with sulfhydryl groups in key enzymes within the body, thereby inhibiting critical functions such as gluconeogenesis and DNA repair (Aposhian and Aposhian 1989; Li and Rossman 1989). Therefore, administration of sulfhydryl-containing compounds soon after exposure could provide alternative target molecules for arsenic, and prevent inhibition of enzyme functions. In fact, many of the chelating agents discussed above (BAL, DMSA, DMPA, DMPS, N-acetylcysteine) contain sulfhydryl groups, and this may account for their efficacy.

The mechanism by which pentavalent arsenic acts is less certain. Since pentavalent arsenic is reduced in the body to the trivalent state, pentavalent arsenic may act in a similar manner as described above for trivalent arsenic. If this is the case, efforts to inhibit the reduction of pentavalent arsenic would decrease its toxicity. However, no methods are currently recognized for blocking this reduction. Pentavalent arsenic may also exert effects by acting as a phosphate analogue. As a phosphate analogue, pentavalent arsenic could potentially affect a number of biological processes, including ATP production, bone formation, and DNA synthesis. However, any effort to interfere in normal phosphate metabolism could produce serious side effects, and no method is known for selectively interfering with arsenate metabolism.

3.12 ADEQUACY OF THE DATABASE

Section 104(I)(5) of CERCLA, as amended, directs the Administrator of ATSDR (in consultation with the Administrator of EPA and agencies and programs of the Public Health Service) to assess whether adequate information on the health effects of arsenic is available. Where adequate information is not available, ATSDR, in conjunction with the National Toxicology Program (NTP), is required to assure the

initiation of a program of research designed to determine the health effects (and techniques for developing methods to determine such health effects) of arsenic.

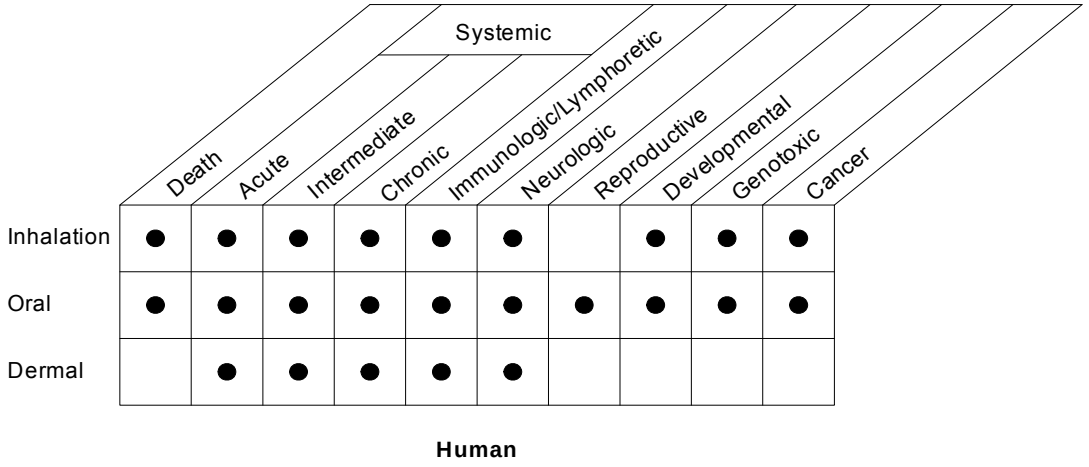
The following categories of possible data needs have been identified by a joint team of scientists from ATSDR, NTP, and EPA. They are defined as substance-specific informational needs that if met would reduce the uncertainties of human health assessment. This definition should not be interpreted to mean that all data needs discussed in this section must be filled. In the future, the identified data needs will be evaluated and prioritized, and a substance-specific research agenda will be proposed.

3.12.1 Existing Information on Health Effects of Arsenic

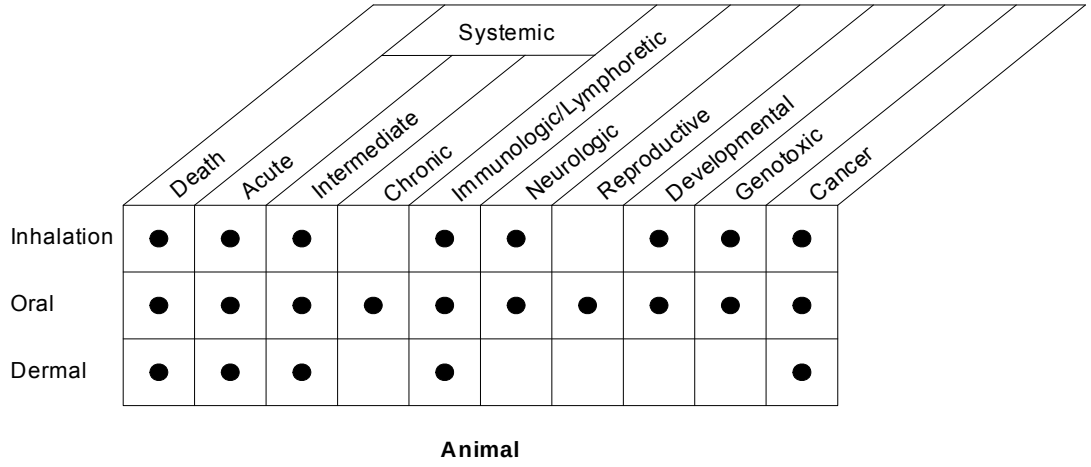
The existing data on health effects of inhalation, oral, and dermal exposure of humans and animals to inorganic and organic arsenic are summarized in Figures 3-13 and 3-14, respectively. The purpose of this figure is to illustrate the existing information concerning the health effects of arsenic. Each dot in the figure indicates that one or more studies provide information associated with that particular effect. The dot does not necessarily imply anything about the quality of the study or studies, nor should missing information in this figure be interpreted as a "data need". A data need, as defined in ATSDR's *Decision Guide for Identifying Substance-Specific Data Needs Related to Toxicological Profiles* (Agency for Toxic Substances and Disease Registry 1989), is substance-specific information necessary to conduct comprehensive public health assessments. Generally, ATSDR defines a data gap more broadly as any substance-specific information missing from the scientific literature.

As shown in Figure 3-13, there is a substantial database on the toxicity of inorganic arsenicals, both in humans and in animals. The oral route has been most thoroughly investigated, and reports are available on most end points of concern following acute, intermediate, and chronic exposure. The inhalation route has also been studied extensively, mainly in humans, with special emphasis on lung cancer. A number of noncancer end points have also been studied following inhalation exposure, but information on these effects is less extensive. Limited information on the effects of dermal exposure is also available in both humans and animals, focusing mainly on direct irritancy and dermal sensitization reactions. The absence of studies on other effects of inorganic arsenic following dermal exposure is probably not a critical data need, since dermal uptake of inorganic arsenic appears to be sufficiently limited that other routes of exposure (oral or inhalation) would almost always be expected to be of greater concern.

Figure 3-13. Existing Information on Health Effects of Inorganic Arsenic



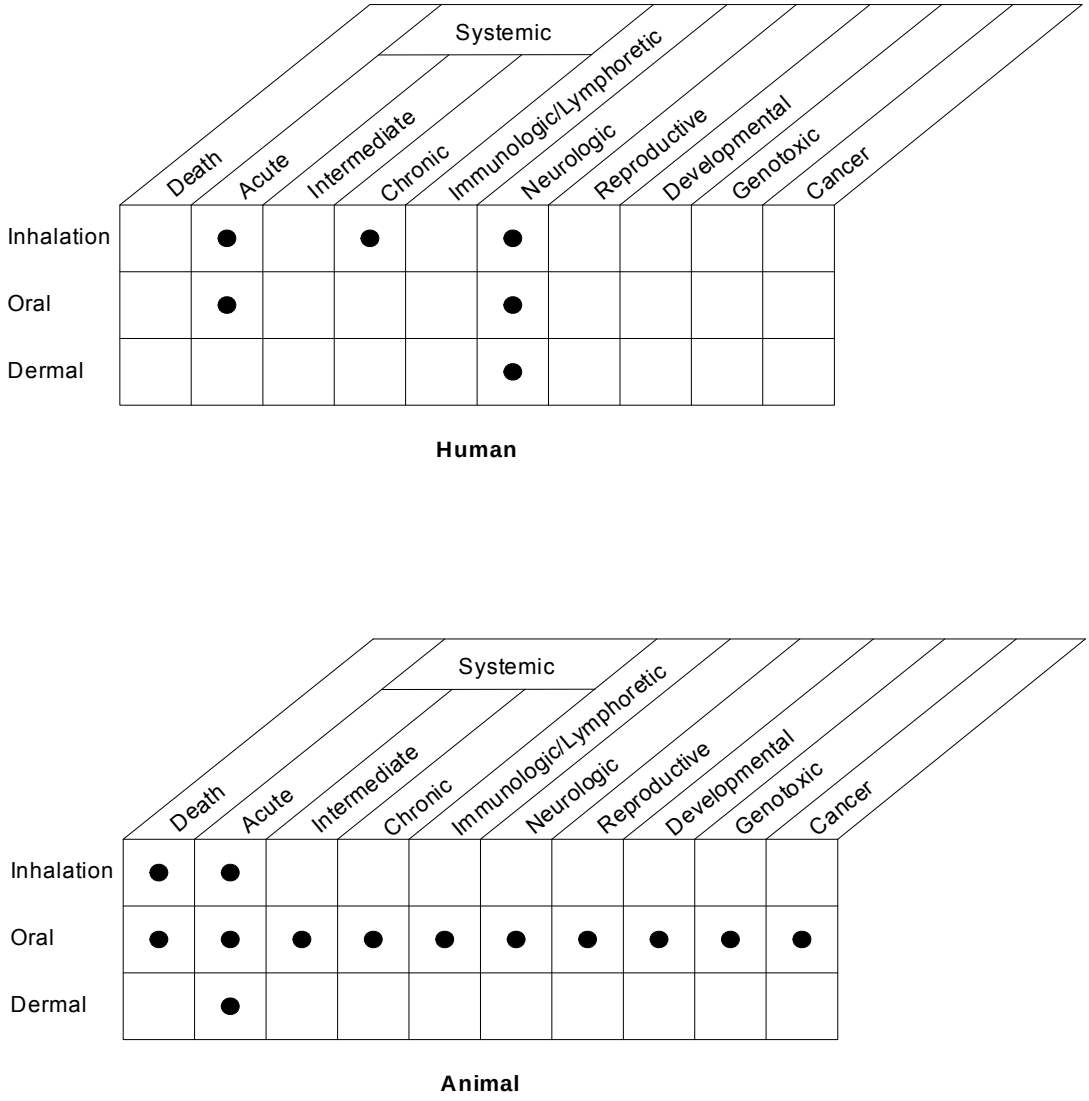
Human



Animal

● Existing Studies

Figure 3-14. Existing Information on Health Effects of Organic Arsenic



As shown in Figure 3-14, very little information is available on the effects of organic arsenic compounds in humans, although there are a number of studies in animals. These studies mainly involve the oral route, since all of these compounds are nonvolatile solids, although a few acute inhalation studies have been performed. Limited information is available on acute dermal lethality and dermal irritancy of some organic arsenicals, but data are lacking on other effects of organic arsenicals following dermal exposure. As discussed previously, in evaluating the adequacy of the database on arsenic, it is important to keep in mind that most studies in animals indicate that they are quantitatively less sensitive to arsenic than humans. For this reason, data from animal studies should be used to draw inferences about effects in humans only with caution.

3.12.2 Identification of Data Needs

Acute-Duration Exposure.

Inorganic Arsenicals. There is only limited information on the effects of acute inhalation exposure to arsenic in humans, but the chief symptoms appear to be irritation of the respiratory and gastrointestinal tracts (Beckett et al. 1986; Bolla-Wilson and Bleecker 1987; Dunlap 1921; Ide and Bullough 1988; Morton and Caron 1989; Pinto and McGill 1953). Quantitative data are lacking, but effects generally appear to be mild even at high-exposure levels. On this basis, it seems that risks of acute effects are probably low for inhalation exposures in the environment or near waste sites. Research to obtain a quantitative acute inhalation NOAEL value that could be used to derive an acute inhalation MRL would, therefore, be useful but not critical. There are numerous case studies in humans on the acute oral toxicity of arsenic, and the main end points (gastrointestinal irritation, pancytopenia, hepatic injury, neuropathy) are well characterized (Armstrong et al. 1984; Fincher and Koerker 1987). An acute oral MRL of 0.005 mg As/kg/day was derived for inorganic arsenic based on a LOAEL for gastrointestinal symptoms and facial edema reported by Mizuta et al. (1956). Additional studies to define an acute oral NOAEL would be useful to reduce uncertainty in the MRL derivation. Acute dermal exposure is unlikely to cause serious systemic injury, but it can lead to contact dermatitis and skin sensitization (Holmqvist 1951; Pinto and McGill 1953). However, available data do not permit a quantitative estimate of the concentration of arsenic on the skin or in air, dust, soil, or water that causes these effects. Further research would be valuable to obtain a quantitative NOAEL for direct dermal effects, since humans may have dermal contact with contaminated soil or water near hazardous waste sites.

Organic Arsenicals. Information on the acute toxicity of organic arsenicals in humans is limited to reports of gastrointestinal irritation in individuals ingesting pesticides containing organic arsenicals (Lee et al. 1995; Shum et al. 1995); these case reports provide limited dosing information. Acute lethality and systemic toxicity data exist for several compounds by inhalation, oral, and dermal exposure of animals. Inhalation data are limited to a lethality study of rats and mice exposed to MMA or DMA that reported respiratory and ocular irritation (Stevens et al. 1979). The oral acute studies consist of lethality studies for MMA (Gur and Nyska 1990; Jaghabir et al. 1988; Kaise et al. 1989), DMA (Kaise et al. 1989), and roxarsone (Kerr et al. 1963; NTP 1989b), systemic toxicity studies (or longer-term studies reporting effects within the first 2 weeks of exposure) for MMA (Irvine et al. 2006), DMA (Ahmad et al. 1999a; Chernoff et al. 1990; Cohen et al. 2001; Crown et al. 1987; Irvine et al. 2006; Kavlock et al. 1985; Rogers et al. 1981; Zomber et al. 1989), or roxarsone (NTP 1989b). For MMA, the available data suggest that the gastrointestinal tract may be the most sensitive target of toxicity; however, the study identifying the lowest LOAEL (Irvine et al. 2006) involved bolus administration and this is not an appropriate exposure route to estimate human risk for gastrointestinal effects following environmental exposure to MMA. The available animal studies for DMA have examined urinary bladder (Cohen et al. 2001) and developmental toxicity (Chernoff et al. 1990, Irvine et al. 2006; Kavlock et al. 1985; Rogers et al. 1981). For DMA, acute-duration studies in rats suggest that the urinary bladder is the most sensitive target of toxicity in rats (Cohen et al. 2001); however, there is evidence from longer-term studies that rats may be more sensitive than humans and other species for bladder effects. Thus, rat data were not considered as the basis of an acute-duration oral MRL for DMA. Other effects observed following acute exposure to DMA include developmental and maternal effects in mice (Kavlock et al. 1985; Rogers et al. 1981) and rabbits (Irvine et al. 2006) and diarrhea and vomiting in dogs receiving a bolus dose of DMA (Zomber et al. 1989). An acute-duration oral MRL was not derived for DMA because it is not known if systemic effects would occur at lower doses than the developmental effects. For roxarsone, the available data suggest that the most sensitive effect following acute oral exposure is neuropathy observed in pigs (Kennedy et al. 1986; Rice et al. 1985). At the only dose tested in this study, tremors, clonic convulsions, and equivocal evidence of myelin degeneration were observed; these were considered serious effects and not suitable for the derivation of an acute-duration oral MRL for roxarsone. Additional studies are needed for MMA, DMA, and roxarsone that examine a variety of end points in several species; studies for roxarsone should also include examination of neurological end points, which would be useful for identifying the critical targets of toxicity and establishing dose-response relationships.

Intermediate-Duration Exposure.

Inorganic Arsenicals. Intermediate-duration inhalation exposure of humans to arsenic appears to result in respiratory tract irritation (occasionally including perforation of the nasal septum) and mild gastrointestinal tract irritation (Ide and Bullough 1988). Quantitative data are too limited (only one study, of one individual) to derive an intermediate-duration inhalation MRL. Further studies to define the NOAEL for intermediate-duration inhalation exposure of humans would be valuable, since humans could be exposed to arsenic-containing airborne dusts near smelters, chemical plants, or waste sites. Effects of intermediate-duration oral exposure are similar to those of acute oral exposure, but may also include development of vascular injury and a characteristic group of skin changes (Franzblau and Lilis 1989; Holland 1904; Wagner et al. 1979). Most studies indicate that these effects occur at doses of about 0.05 mg As/kg/day or higher, but the data do not provide a firm basis for identifying the intermediate-duration NOAEL. For this reason, no intermediate-duration oral MRL has been derived. Further studies to establish the NOAEL would be valuable, since humans could have intermediate-duration oral exposures to arsenic through ingestion of contaminated soil or water near smelters, chemical factories, or waste sites. Since dermal effects appear to be restricted to acute irritancy, intermediate-duration dermal studies are probably not essential.

Organic Arsenicals. No information was located on the intermediate-duration toxicity of organic arsenicals in humans. Several studies have examined the intermediate-duration oral toxicity of MMA; dietary exposure studies in rats and mice (Arnold et al. 2003) identify the gastrointestinal tract as the most sensitive target. Diarrhea and lesions in the cecum, colon, and rectum have been observed. The rat 13-week study (Arnold et al. 2003) was used as the basis of the MRL. Because rats appear to be more sensitive to the toxicity of DMA, rat studies were not considered for MRL derivation. The only non-rat study was a chronic-duration dog study reporting effects during the first 51 weeks of exposure (Zomber et al. 1989); these effects included diarrhea and vomiting. However, because DMA was administered via capsule, this study was not considered adequate for derivation of an MRL. Additional studies are needed for DMA to identify critical targets of toxicity and establish dose-response relationships in non-rat species. The available data for roxarsone suggest that neurotoxicity in pigs is the most sensitive end point. One of the two available neurotoxicity studies in pigs (Edmonds and Baker 1986) did not include sensitive tests of toxicity and was not considered for MRL derivation; the other study identified a serious LOAEL at the only dose tested and thus, was not suitable for MRL derivation. Several comprehensive studies examined the toxicity of roxarsone in rats and mice (NTP 1989b). Renal tubular damage in rats was the most sensitive end point (NTP 1989b); however, the LOAEL for this effect was 9 times higher

than the dose associated with neurotoxicity in pigs. Additional studies are needed to establish a no effect level for neurotoxicity in pigs, which could be used to derive an intermediate duration MRL for roxarsone. Further studies on the intermediate-duration inhalation and dermal toxicity of these compounds would be valuable, especially in humans, since people may be exposed to organic arsenicals during their manufacture or use, or from materials deposited in waste sites.

Chronic-Duration Exposure and Cancer.

Inorganic Arsenicals. The target tissues of chronic-duration exposure of humans to inorganic arsenic are the same as for intermediate-duration exposure for both the oral and inhalation routes. Effects of dermal exposure appear to be restricted to direct irritation of exposed surfaces. Therefore, chronic-duration studies are probably not essential for the dermal route. Quantitative data from one study identify an inhalation exposure level of about 0.1 mg As/m^3 as the LOAEL for skin changes (Perry et al. 1948), but because there are no additional supporting studies and a NOAEL is not clearly established, a chronic-duration inhalation MRL has not been derived. Additional studies in humans to define the chronic inhalation NOAEL for dermal or other effects would be valuable, since humans may be chronically exposed to arsenic dusts in air near smelters, chemical factories, or waste sites. Chronic oral exposure data from studies in humans indicate that the LOAEL for skin lesions and other effects is probably about 0.01–0.02 mg As/kg/day (10–20 µg As/kg/day), and that the NOAEL is probably between 0.0004 and 0.0009 mg As/kg/day (0.4–0.9 µg As/kg/day) (Cebrián et al. 1983; EPA 1981b; Hindmarsh et al. 1977; Tseng 1977; Tseng et al. 1968). The NOAEL of 0.0008 mg As/kg/day from the study by Tseng et al. (1968) is appropriate for derivation of a chronic-duration oral MRL, but an uncertainty factor of 3 was required to account for the fact that the population that constituted the no-effect group were relatively young (possibly decreasing the ability to detect dermal or other effects that increase in prevalence with age). Another issue that needs to be acknowledged, which is common to ecological studies and contributes to uncertainty, is the fact that individual doses were not available and were calculated from group mean arsenic concentrations in well water using estimated water intake parameters. For this reason, further epidemiological studies that do not rely on an ecological-based exposure assessment that would provide additional support for the threshold dose for arsenic in humans would be valuable.

There are numerous studies in humans that support the carcinogenic effects of inorganic arsenic from inhalation exposure (Enterline et al. 1987a, 1987b, 1995; Järup and Pershagen 1991; Järup et al. 1989; Lee-Feldstein 1986; Welch et al. 1982) and oral exposure (Chen et al. 1986, 1988b, 1992; Chiou et al. 1995; Ferreccio et al. 1996; Hsueh et al. 1995; Lander et al. 1975; Liu and Chen 1996; Lüchtrath 1983;

Smith et al. 1992; Tseng 1977; Tseng et al. 1968; Yu et al. 1992; Zaldívar 1974; Zaldívar et al. 1981). Quantitative slope factors have been derived for both routes. There is a noticeable absence, however, of 2-year animal carcinogenicity studies for either the inhalation or oral route of exposure (Chan and Huff 1997). In light of the ongoing controversy over the reasons for the absence of a carcinogenic effect in animals, it seems prudent to firmly establish a negative effect in a 2-year study. The carcinogenic effects of chronic dermal exposure to inorganic arsenicals have not been studied, but dermal exposure is a relatively minor route of exposure, and these studies would not be a top priority.

The mechanism of arsenic carcinogenicity is not known, although the current view is that it functions mainly as a promoter or cocarcinogen. Further studies on the mechanism of arsenic toxicity would be particularly valuable to improve our ability to evaluate human cancer risks from inhalation or oral exposures that might occur near waste sites. Also, mechanistic studies could help in the evaluation of cancer risks from organic derivatives (see below).

Organic Arsenicals. There is very little information on the chronic toxicity of organic arsenicals in humans. One study of workers exposed to arsanilic acid did not identify any adverse effects, but no systematic, clinical, or toxicological examinations of exposed people were performed (Watrous and McCaughey 1945). Chronic toxicity studies are available for rats, mice, and dogs exposed to MMA (Arnold et al. 2003; Waner and Nyska 1988), DMA (Arnold et al. 2006; Zomber et al. 1989), and roxarsone (NTP 1989b; Prier et al. 1963). Chronic exposure to MMA results in diarrhea in rats, mice, and dogs (Arnold et al. 2003; Waner and Nyska 1988) and an increase in progressive nephropathy in rats and mice (Arnold et al. 2003). The increased incidence of progressive nephropathy was used as the basis of the chronic-duration oral MRL for MMA. For DMA, chronic exposure also resulted in an increased incidence of diarrhea and vomiting in dogs (Zomber et al. 1989) and an increased incidence of vacuolization in the urinary bladder and progressive nephropathy in mice (Arnold et al. 2006). The vacuolization in the urinary bladder was used as the basis of a chronic-duration oral MRL for DMA. The available data for chronic-exposure to roxarsone were considered inadequate for derivation of an MRL. The highest doses tested in the rat, mouse, and dog studies (NTP 1989b; Prier et al. 1963) were NOAELs. Intermediate-duration studies identify neurotoxicity in pigs as the most sensitive end point; this has not been adequately examined following chronic exposure and studies are needed.

No information was located on carcinogenic effects of organic arsenicals in humans. The carcinogenic potential of MMA (Arnold et al. 2003), DMA (Arnold et al. 2006), and roxarsone (NTP 1989b) following oral exposure has been investigated in rats and mice. No evidence of carcinogenicity was observed

following oral exposure to MMA (Arnold et al. 2003) and equivocal evidence of carcinogenicity was found in male rats, with no evidence of carcinogenicity in female rats or in male or female mice orally exposed to roxarsone (NTP 1989b). Oral exposure to DMA resulted in an increased incidence of urinary bladder tumors in rats and no evidence of carcinogenicity in mice (Arnold et al. 2006). However, there is concern that the rat is not a good model to assess the carcinogenic potential of DMA in humans due to species differences in the toxicokinetic properties of DMA. No information was located on the carcinogenicity of organic arsenicals following inhalation or dermal exposure. Studies of humans exposed in the workplace would provide valuable information on the carcinogenic potential of organic arsenicals, particularly DMA. Studies on cancer risk following inhalation and dermal exposure to organic arsenicals are would be useful since these are possible routes of exposure for humans.

Genotoxicity.

Inorganic Arsenicals. There are several studies that suggest that inorganic arsenic may cause genotoxicity (mainly chromosomal effects) in exposed humans (Burgdorf et al. 1977; Nordenson et al. 1978), and this is supported by numerous studies in animals (Datta et al. 1986; DeKnudt et al. 1986; Nagymajtényi et al. 1985) and cultured cells (Beckman and Nordenson 1986; Casto et al. 1979; DiPaolo and Casto 1979; Lee et al. 1985; Nakamuro and Sayato 1981; Nishioka 1975; Oberly et al. 1982; Okui and Fujiwara 1986; Sweins 1983; Ulitzur and Barak 1988; Zanzoni and Jung 1980). The mechanism of genotoxicity is not known, but may be due to the ability of arsenite to interfere with DNA repair (Li and Rossman 1989) or to alter apoptosis (Pi et al. 2005) or the ability of arsenate to act as a phosphate analog. Further studies to improve our understanding of the mechanism of genotoxicity would be valuable, since this could aid in the understanding of arsenic-induced cancer risk.

Organic Arsenicals. For organic arsenicals, *in vitro* genotoxicity studies are available for arsenobetaine (Eguchi et al. 1997; Oya-Ohta et al. 1996), MMA (Chun and Killeen 1989a, 1989b, 1989c, 1989d; Eguchi et al. 1997; Oya-Ohta et al. 1996), DMA (Eguchi et al. 1997; Endo et al. 1992; Kato et al. 1994; Kawaguchi et al. 1996; Kitamura et al. 2002; Kuroda et al. 2004; Moore et al. 1997a; Oya-Ohta et al. 1996; Rasmussen and Menzel 1997; Rin et al. 1995; Tezuka et al. 1993; Ueda et al. 1997; Yamanaka et al. 1989b, 1993, 1995, 1997), and roxarsone (Matthews et al. 1993; NTP 1989b; Storer et al. 1996) and *in vivo* studies are available for DMA (Kashiwada et al. 1998; Yamanaka and Okada 1994; Yamanaka et al. 1989a, 1989b, 1993, 2001). The results of these studies suggest that DMA and roxarsone are clastogenic and can cause DNA strand breaks. Additional *in vivo* studies are needed to evaluate the genotoxic potential of MMA and roxarsone.

Reproductive Toxicity.

Inorganic Arsenicals. Several studies have examined reproductive function in populations living in Bangladesh or India exposed to high levels of arsenic in drinking water and found increases in spontaneous abortions/stillbirths or preterm births (Ahmad et al. 2001; von Ehrenstein et al. 2006); another study in U.S. women did not find an increase in adverse reproductive outcomes (Aschengrau et al. 1989). Available animal studies did not find evidence for reproductive effects following inhalation or oral exposure (Holson et al. 1999, 2000), except for a trend toward decreased pups per litter in mice in a 3-generation study (Schroeder and Mitchener 1971) that is consistent with embryolethality observed in developmental studies of inorganic arsenic. Studies on spermatogenesis and reproductive success in arsenic-exposed workers would be valuable in evaluating whether there are significant reproductive risks of arsenic in humans, and this could be further strengthened by studies including histopathological examination of reproductive tissues (which was not done in the existing studies) in animals.

Organic Arsenicals. No information was located on reproductive effects of organic arsenicals in humans and no inhalation or dermal exposure animal studies were located. Intermediate- and chronic-duration oral studies for MMA (Arnold et al. 2003), DMA (Arnold et al. 2006), and roxarsone (NTP 1989b) have not reported histological damage to reproductive tissues. Decreases in pregnancy rate and male fertility index were observed in a two-generation study in rats (Schroeder 1994) and a single generation study in mice (Prukop and Savage 1986) exposed to MMA; the poor reporting in the Prukop and Savage (1986) study limits its usefulness in assessing reproductive toxicity. However, in the two-generation study, the differences between control and exposed rats were not statistically different; the effect was considered biologically significant because effects observed in the exposed rats were outside the range found in historical controls. Another reproductive performance study to confirm these results would be useful. No reproductive effects were observed in a two generation study in rats exposed to DMA (Rubin et al. 1989).

Developmental Toxicity.

Inorganic Arsenicals. There are several epidemiological studies that suggest that inhalation (Ihrig et al. 1998; Nordström et al. 1978a, 1978b, 1979a, 1979b) or oral (Hopenhayn et al. 2003a; Yang et al. 2003) exposure to inorganic arsenic might increase the risk of low birth weight, congenital defects, or abortion in exposed women. These studies do not establish that arsenic was responsible, since all involved exposures to other chemicals or risk factors, but do suggest that additional studies on developmental

parameters in humans exposed to arsenic would be valuable in determining whether this is an effect of concern. Other human studies have not found significant associations between arsenic levels in drinking water and increased neonatal deaths or infant mortality (von Ehrenstein et al. 2006) or the increase in congenital heart defects (Zierler et al. 1988) or neural tube defects (Brender et al. 2006). Studies in animals support the view that oral, inhalation, and parenteral exposure to inorganic arsenic can all increase the incidence of fetotoxicity and teratogenicity, although this appears to occur only at doses that are toxic or even lethal to the dams (Baxley et al. 1981; Beaudoin 1974; Carpenter 1987; Ferm and Carpenter 1968; Ferm et al. 1971; Hanlon and Ferm 1986; Holson et al. 1999, 2000; Hood and Bishop 1972; Hood and Harrison 1982; Hood et al. 1978; Mason et al. 1989; Nagymajtényi et al. 1985; Nemec et al. 1998; Stump et al. 1999; Willhite 1981). There are also some data to suggest that it may increase the risk of transplacental cancer in humans (Smith et al. 2006) and animals (Waalkes et al. 2003). Thus, additional studies in animals may be useful in defining the mechanisms of these developmental effects and in identifying the time of maximum susceptibility of the fetus, but such studies probably will not help identify a safe exposure level for humans.

Organic Arsenicals. No information was located regarding developmental effects in humans after oral or inhalation exposure to organic arsenicals. Animal studies conducted in rats (Chernoff et al. 1990; Irvine et al. 2006; Rogers et al. 1981), mice (Kavlock et al. 1985; Rogers et al. 1981), and rabbits (Irvine et al. 2006) have examined the developmental toxicity of organic arsenicals. Decreases in fetal body weights and delays in ossification were commonly reported at maternally toxic (decreases in body weight gain) doses of DMA (Irvine et al. 2006; Kavlock et al. 1985; Rogers et al. 1981). However, one study found increases in the percentage of fetuses with irregular palatine rugae at DMA doses not associated with maternal toxicity (Rogers et al. 1981). This effect has not been reported in other studies and additional developmental studies are needed to confirm the finding. In view of the apparent differences in susceptibility between animals and humans, it would be valuable to investigate whether there are any measurable effects on development in humans exposed to organic arsenicals in the workplace or the environment.

Immunotoxicity.

Inorganic Arsenicals. No studies were located on immunotoxic effects in humans after oral exposure to inorganic arsenic. One inhalation study in humans (Bencko et al. 1988), an inhalation study in animals (Aranyi et al. 1985), one oral study in animals (Kerkvliet et al. 1980), and one intratracheal instillation study in animals (Sikorski et al. 1989) suggest that arsenic causes little or no functional impairment of the

immune system, but one inhalation study in animals found decreased pulmonary bactericidal activity and increased susceptibility to streptococcal infection in exposed mice (Aranyi et al. 1985). Additional studies (both in humans and animals) would be valuable to investigate this end point further. Dermal exposure of humans to high levels of arsenic dusts may cause dermal sensitization (Holmqvist 1951), but the dose and time dependence of this phenomenon are not known. Studies to determine whether dermal sensitization occurs in people with low level dermal exposures to arsenic in dust or soil, such as might occur for residents near an arsenic-containing waste site, would be valuable in assessing the significance of this effect to nonoccupationally exposed populations.

Organic Arsenicals. No information was located on the effect of organic arsenicals exposure in humans or animals on immune function. Since there are suggestions that inorganic arsenic may cause some changes in the immune system, studies on possible immune effects of the common organic arsenicals might be helpful.

Neurotoxicity.

Inorganic Arsenicals. There is convincing evidence from studies in humans that inorganic arsenic can cause serious neurological effects, both after inhalation (Beckett et al. 1986; Bencko et al. 1977; Blom et al. 1985; Buchancová et al. 1998; Calderón et al. 2001; Danan et al. 1984; Feldman et al. 1979; Gerr et al. 2000; Lagerkvist and Zetterlund 1994; Morton and Caron 1989) and oral exposure (Armstrong et al. 1984; Bartolome et al. 1999; Chakraborti et al. 2003a, 2003b; Civantos et al. 1995; Cullen et al. 1995; Danan et al. 1984; EPA 1977a; Feldman et al. 1979; Fincher and Foy et al. 1992; Franzblau and Lilis 1989; Guha Mazumder et al. 1988; Hindmarsh et al. 1977; Huang et al. 1985; Fincher and Koerker 1987; Levin-Scherz et al. 1987; Lewis et al. 1999; Mizuta et al. 1956; Muzi et al. 2001; Quatrehomme et al. 1992; Silver and Wainman 1952; Szuler et al. 1979; Tsai et al. 2003; Uede and Furukawa 2003; Vantroyen et al. 2004; Wagner et al. 1979). This is based mainly on clinical observations and neurological examinations of exposed persons. Available studies provide a reasonable estimate of LOAEL and NOAEL values by the oral route, but similar data are lacking for the inhalation route. Further studies designed to identify the threshold for neurological effects in humans exposed by the inhalation route would be valuable, since humans may be exposed to arsenic dusts in air from smelters, chemical factories, or waste sites. Adult animals appear to be much less susceptible than humans to the neurological effects of inorganic arsenic, so studies in adult animals would probably not help in estimation of a safe exposure limit. However, in light of recent findings of possible associations between arsenic in drinking water and neurobehavioral alterations in children (Tsai et al. 2003; von Ehrenstein et

al. 2007; Wang et al. 2007; Wasserman et al. 2004, 2007), studies in animals, in which confounding can be eliminated, may be warranted.

Organic Arsenicals. Information on neurological effects of organic arsenicals in humans is limited to an occupational study that did not find increases in the frequency of central or peripheral nervous system complaints (Watrous and McCaughey 1945) and a case report of a women of a women reporting numbness and tingling of the fingertips, toes, and circomoral region who was exposed to organic arsenic in soup (Luong and Nguyen 1999). Neurological effects have also been observed in some animal studies. Decreases spontaneous motility, ataxia, and increased startle response were observed in mice exposed to a single high dose of DMA (Kaise et al. 1989). Degeneration of myelin and axons were observed in several studies involving oral exposure of pigs to roxarsone (Edmonds and Baker 1986; Kennedy et al. 1986; Rice et al. 1985). Hyperexcitability, ataxia, and trembling have also been observed in rats and mice orally exposed to roxarsone (Kerr et al. 1963; NTP 1989b). These findings suggest that more extensive investigations of the neurotoxic potential of roxarsone and other organic arsenicals would be valuable to determine the potential human health risk from these compounds, since humans could be exposed during the manufacture or use of these compounds, or near waste sites where they have been deposited.

Epidemiological and Human Dosimetry Studies. Numerous epidemiologic studies of humans exposed to inorganic arsenic by the oral and inhalation routes constitute the database on arsenic-related cancer and noncancer human health effects. As with virtually all epidemiologic investigations, these studies are limited by possible confounding from factors such as smoking, exposure to other chemicals, and differences in population characteristics (e.g., nutritional state, metabolism, and toxicokinetics) that inhibit extrapolation of study results to a wider population. Moreover, many of these studies lack good dose estimates for study participants. Some studies lack quantitative data altogether. For this reason, improved data on confounding factors and improved methods of human dosimetry would be valuable in any further human epidemiologic studies of arsenic, either in the workplace or in the general environment. Recent work has broadened the qualitative dose-response information beyond the highly exposed Taiwanese population, but additional studies of persons with lower exposure levels would be especially valuable for risk assessments for the U.S. population. From a public health standpoint, well designed studies of common noncancer health outcomes (e.g., cardiovascular disease and diabetes) could be more important than additional studies of cancer. Availability of methods for biomonitoring of exposure are discussed below.

Biomarkers of Exposure and Effect.

Exposure. There are sensitive and specific methods for measuring arsenic in blood, urine, hair, nails, and other tissues, and this is the approach normally employed for measuring arsenic exposure in humans. Usually total arsenic is measured, but methods are available for measuring inorganic arsenic and each of the organic derivatives separately. Urinary levels are generally considered to be the most reliable indication of recent exposures (Enterline et al. 1987a; Milham and Strong 1974; Pinto et al. 1976; Polissar et al. 1990), but if a high urinary level is present, care must be taken to account for the presence of nontoxic forms of arsenic from the diet. Blood levels are sometimes used to evaluate the status of acutely poisoned individuals (Driesback 1980; Heydorn 1970; Hindmarsh and McCurdy 1986; Valentine et al. 1979, 1981), but this approach is not generally useful for biomonitoring of long-term exposure to low levels. Hair and nails provide a valuable indication of exposures that occurred 1–10 months earlier (Agahian et al. 1990; Bencko et al. 1986; Choucair and Ajax 1988; EPA 1977a, 1981b; Milham and Strong 1974; Valentine et al. 1979; Yamauchi et al. 1989), although care must be taken to exclude external contamination of these samples. Cumulative urinary arsenic levels may be used to derive a quantitative estimate of exposure (Enterline et al. 1987a; Pinto et al. 1976), but data on the quantitative relation between exposure and arsenic levels in nails and hair were not located. Efforts to establish an algorithm for estimating past exposure levels from hair or nail levels would be valuable in quantifying average long-term exposure levels in people where repeated urinary monitoring is not feasible.

Effect. The effects of arsenic are mainly nonspecific, but the combined presence of several of the most characteristic clinical signs (e.g., nausea, diarrhea, peripheral neuropathy, anemia, vascular lesions, hyperkeratinization, hyperpigmentation) is usually adequate to suggest arsenic intoxication. Although there are standard clinical methods for detecting and evaluating each of these effects, there are no recognized methods for identifying early (preclinical) effects in exposed persons. Neurophysiological measurements of nerve conduction velocity or amplitude have been investigated (Goebel et al. 1990; Jenkins 1966; Le Quesne and McLeod 1977; Morton and Caron 1989; Murphy et al. 1981), but at present, this approach does not seem to offer much advantage over a standard neurological examination. Changes in urinary excretion levels of several heme-related metabolites appear to be a good indication of preclinical effects of arsenic toxicity in animals (Albores et al. 1989; Sardana et al. 1981; Woods and Fowler 1978; Woods and Southern 1989), but this has not been established in humans and is not specific for arsenic-induced effects. Further efforts to develop these approaches and to identify other more specific biochemical or physiological indicators of arsenic-induced effects would be very valuable in monitoring the health of persons exposed to low levels of arsenic in the environment or near waste sites.

Absorption, Distribution, Metabolism, and Excretion. Available data from toxicokinetic studies in humans reveal that arsenates and arsenites are well absorbed following both oral and inhalation exposure. Data on distribution are limited, but it appears that arsenic is transported to nearly all tissues. Metabolism involves mainly reduction-oxidation reactions that interconvert As(+5) and As(+3) and methylation of As(+3) to yield MMA and DMA. Most arsenic is rapidly excreted in the urine as a mixture of inorganic arsenics, MMA, and DMA, although some may remain bound in tissues (especially skin, hair, and fingernails). These findings are strongly supported by numerous studies in animals. Because methylation represents a detoxification pathway, an area of special interest is the capacity of the human body to methylate inorganic arsenic. Limited data suggest that the methylation system might begin to become saturated at intakes of about 0.2–1 mg As/day (Buchet et al. 1981b; Marcus and Rispin 1988), but this is uncertain. Further studies to define the rate and saturation kinetics of whole-body methylation in humans would be especially helpful in evaluating human health risk from the low levels of arsenic intake that are usually encountered in the environment. Along the same line, further studies to determine the nature and magnitude of individual variations in methylation capacity and how this depends on diet, age, and other factors would be very useful in understanding and predicting which members of a population are likely to be most susceptible.

The toxicokinetics of dermal exposure have not been studied. It is usually considered that dermal uptake of arsenates and arsenites is sufficiently slow that this route is unlikely to be of health concern (except that due to direct irritation), but studies to test the validity of this assumption would be valuable. Also, dermal uptake of organic arsenicals could be of concern, and quantitative data on the rate and extent of this would be helpful in evaluating risks from application of arsenical pesticides or exposures to organic arsenicals in waste sites.

Comparative Toxicokinetics. Available toxicity data indicate that arsenic causes many of the same effects in animals that are observed in humans, but that animals are significantly less sensitive. The basis for this difference in susceptibility is not certain but is probably mainly a result of differences in absorption, distribution, metabolism, or excretion. For example, rats strongly retain arsenic in red blood cells (Lanz et al. 1950), while humans (and most other species) do not. Similarly, marmoset monkeys do not methylate inorganic arsenic (Vahter and Marafante 1985; Vahter et al. 1982), while humans and other animal species do. Because of these clear differences in toxicity and toxicokinetics between species, further comparative toxicokinetic studies that focus on the mechanistic basis for these differences would be very valuable. At a minimum, this would help clarify which laboratory species are the most useful

models for humans and could ultimately lead to development of a PBPK model that would permit reliable extrapolation of observations across species.

Methods for Reducing Toxic Effects. There are a number of general methods for reducing the absorption of arsenic in the gastrointestinal tract and skin, but there are currently no methods for reducing the absorption of arsenic from the lungs. The removal of arsenic from the gastrointestinal tract is usually facilitated by the use of emetics, cathartics, lavages, or activated charcoal (Agency for Toxic Substances and Disease Registry 1990a; Aposhian and Aposhian 1989; Campbell and Alvarez 1989; Driesback 1980; Ellenhorn and Barceloux 1988; EPA 1989e; Haddad and Winchester 1990; Mitra et al. 2004; Stutz and Janusz 1988). Studies that investigate the effects of phosphate-binding chemicals (aluminum hydroxide) and nonabsorbable sulfhydryl compounds on the absorption of pentavalent and trivalent arsenic, respectively, may be useful in developing treatments that are more specific to arsenic intoxication. Once arsenic is in the body, treatment usually involves the use of one or more chelators, such as BAL or penicillamine. However, these agents often exhibit adverse side effects (Agency for Toxic Substances and Disease Registry 1990a; Ellenhorn and Barceloux 1988), and are generally only applied following high-dose acute exposure. Further studies investigating the efficacy of less toxic arsenic chelators, such as DMSA, DMPA, DMPS, and N-acetyl cysteine, may lead to the development of safer treatment methods. Studies on the efficacy of chelators and agents to enhance methylation and elimination in treatment of chronic arsenic exposure would also be helpful, as available treatment methods for chronic arsenic exposure are limited. Trivalent arsenic is generally believed to exert toxic effects by binding to the vicinal sulfhydryl group of key enzymes, thereby interfering with a number of biological processes, such as gluconeogenesis and DNA repair (Li and Rossman 1989; Szinicz and Forth 1988). Since pentavalent arsenic may need to be reduced in the body to the trivalent state before it can exert toxic effects, studies that investigate methods for blocking this conversion may lead to a method for interfering with the mechanism of action for pentavalent arsenic. The insufficient intake of calcium, animal protein, folate, selenium, and fiber may enhance the toxic effects of inorganic arsenic (Mitra et al. 2004), but it is not known if dietary supplementation will prove effective in patients who already show arsenic-induced symptoms.

Children's Susceptibility. Data needs relating to both prenatal and childhood exposures, and developmental effects expressed either prenatally or during childhood, are discussed in detail in the Developmental Toxicity subsection above.

A majority of the data on the effects of exposure of humans to arsenic has focused on adults. Although a few studies of acute poisoning and chronic exposure specifically describe children (Borgoño et al. 1980; Concha et al. 1998a, 1998b, 1999; Foy et al. 1992; Kersjes et al. 1987; Rosenberg 1974; Zaldívar 1974; Zaldívar and Guillier 1977), in general, data are lacking. Specifically, although there is a substantial database on the effect of arsenic on animal development, there are few data describing developmental effects in humans. Additional research in this area, using populations in areas of endemic arsenic exposure, would be useful.

Although there is no reason to suspect that the pharmacokinetics of arsenic differs in children and adults, there are few data available on this topic. Research on absorption, distribution, metabolism, and excretion in children would aid in determining if children are at an increased risk, especially in areas where chronic exposure to an environmental source occurs. With regard to exposure during development, additional research on maternal kinetics, and transfer via breast milk would be useful in obtaining a more complete picture of prenatal and neonatal development, especially with regard to neural development and the possible development of childhood cancer.

Child health data needs relating to exposure are discussed in Section 6.8.1, Identification of Data Needs: Exposures of Children.

3.12.3 Ongoing Studies

A number of researchers are continuing to investigate the toxicity and toxicokinetics of arsenic. Table 3-18 summarizes studies being sponsored by agencies of the U.S. federal government (FEDRIP 2007). Additional research is being sponsored by industry groups and other agencies, and research is also ongoing in a number of foreign countries.

Table 3-18. Ongoing Studies on Health Effects of Arsenic, Federally Funded

Investigator	Affiliation	Title	Sponsor
Ahsan, H	Columbia University, New York, New York	Chemoprevention of arsenic-induced skin cancer	NCI
Ahsan, H	Columbia University, New York, New York	Genetic susceptibility to arsenic-induced skin cancer	NCI
Andrew, A	Darmouth College, Hanover, New Hampshire	Bladder cancer prognostic indicators	NCI
Beckman, K	Children's Hospital and Research Center, Oakland, California	Fetal arsenic-nutrient interaction in adult-onset cancer	NIEHS
Bodwell, J	Darmouth College, Hanover, New Hampshire	Arsenic effects on glucocorticoid receptor action	NIEHS
Calderon, R	EPA, Research Triangle Park, North Carolina	Arsenic-induced skin conditions identified in Southwest United States	HEERL
Christiani, D	Harvard University, Boston, Massachusetts	Arsenic and health in Bangladesh	NIEHS
Dong, Z	University of Minnesota, Minneapolis, Minnesota	Molecular basis of arsenic-induced cell transformation	NCI
Finnell, R	Texas A & M University College Station, Texas	Sensitive genotypes to arsenic as a model environmental toxicant	NIEHS
Frenkel, K	New York University, New York, New York	Metal induced inflammatory factors, oxidative stress, and suppression	NIEHS
Futscher, B	University of Arizona, Tucson, Arizona	Epigenetic remodeling by environmental arsenicals	NCI
Gamble, M	Columbia University, New York, New York	Nutritional influences on arsenic toxicity	NIEHS
Germolec, D	NIH, Research Triangle Park, North Carolina	The role of growth factors and inflammatory mediators in arsenic-induced dermatotoxicity	NIEHS
Guallar, E	Johns Hopkins University, Baltimore, Maryland	Mercury, arsenic, and carotid atherosclerosis	NIEHS
He, K	Northwestern University, Chicago, Illinois	Trace elements and CVD risks factors among young adults	NHLBI
Hei, T	Columbia University, New York, New York	Mechanisms of arsenic carcinogenesis	NIEHS
Huang, C	New York University, New York, New York	Effects of arsenic on PI-3K signaling pathway	NCI
Hudgens, E	EPA, Research Triangle Park, North Carolina	Study of individuals chronically exposed to arsenic in drinking water	HEERL
Hughes, M	EPA, Research Triangle Park, North Carolina	Biomarkers of exposure: a case study with inorganic arsenic	HEERL
Hughes, M	EPA, Research Triangle Park, North Carolina	Tissue dosimetry, metabolism, and excretion of pentavalent arsenic	HEERL
Jing, Y	New York University, New York, New York	Arsenic trioxide and acute myeloid leukemia	NCI

Table 3-18. Ongoing Studies on Health Effects of Arsenic, Federally Funded

Investigator	Affiliation	Title	Sponsor
Jung, M	Georgetown University, Washington, DC	Epigenetic regulation by poly(ADP-ribose) in response to arsenite	NIEHS
Karin, M	University of California San Diego, La Jolla, California	Interaction of heavy metal ions with the human genome	NIEHS
Kelsey, K	Harvard University, Boston, Massachusetts	Arsenic mode of action in cancer—models of epigenetic mechanism	NIEHS
Liu, K	University of New Mexico, Albuquerque, New Mexico	Oxidative mechanisms of arsenic-induced carcinogenesis	NIEHS
Markowski, V	University of Southern Maine, Portland, Maine	Developmental arsenic exposure produces cognitive impairment	NIEHS
Martin, M	Georgetown University, Washington, DC	Arsenic and epigenetic regulation of gene expression	NIEHS
Muscarella, D	Cornell University Ithaca, Ithaca, New York	Arsenite effects on CD40 signaling and B-cell apoptosis	NIEHS
Nichols, R	Dartmouth College, Hanover, New Hampshire	Effect of arsenic on cytochrome P450	NIEHS
Nriagu, J	University of Michigan, Ann Harbor, Michigan	Arsenic exposure and bladder cancer in Michigan	NCI
Rosen, B	Wayne State University, Detroit, Michigan	Mechanisms of arsenical transport	NIGMS
Rosen, B	Wayne State University, Detroit, Michigan	Metal binding domains in metallo-regulatory proteins	NIAID
Rosenblatt, A	University of Miami, Coral Gables, Florida	Environmental arsenic and androgen receptor regulation	NIEHS
Rossman, T	New York University, New York, New York	Investigation and genetic analysis of the human arsenite efflux pump	NIEHS
Schwartz, J	Harvard University, Boston Massachusetts	Epigenetic effects of particles and metals on cardiac health of an aging cohort	NIEHS
Self, W	University of Central Florida, Orlando, Florida	Impact of arsenicals on selenoprotein synthesis	NIEHS
Sens, D	University of North Dakota, Grand Forks, North Dakota	Metallothionein isoform-3 urinary marker bladder cancer	NIEHS
Sheldon, L	Dartmouth College, Hanover, New Hampshire	Arsenic, histone modification, and transcription	NIEHS
Shi, X	University of Kentucky, Lexington, Kentucky	Mechanism of arsenic-induced carcinogenesis	NCI
Smith, A	University of California, Berkeley, California	Arsenic biomarker epidemiology	NIEHS
Spallholz, J	Texas Tech University, Lubbock, Texas	Selenium against arsenic toxicity and skin lesions	NCI
States, C	University of Louisville, Louisville, Kentucky	Arsenic induced miotic arrest associated apoptosis	NIEHS
Styblo, M	University of North Carolina, Chapel Hill, North Carolina	Metabolism and toxicity of arsenic in human liver	NIEHS

Table 3-18. Ongoing Studies on Health Effects of Arsenic, Federally Funded

Investigator	Affiliation	Title	Sponsor
Taylor, P	Division of Cancer Epidemiology and Genetics, NCI, Bethesda, Maryland	Biologic specimen bank for early lung cancer markers in Chinese tin miners	NCI
Taylor, B	University of Louisville, Louisville, Kentucky	Arsenite inhibition of mitotic progression	NIEHS
Vaillancourt, R	University of Arizona, Tucson, Arizona	Modulation of Prostaglandins by Arsenic	NIEHS
Willett, W	Harvard University, Boston, Massachusetts	Prospective studies of diet and cancer in men and women	NCI
Wright, R	Brigham and Women's Hospital, Boston, Massachusetts	Metal mixtures and neurondevelopment	NIEHS
Zhang, D	University of Arizona, Tucson, Arizona	The protective role of Nrf2 in arsenic-induced toxicity and carcinogenicity	NIEHS

EPA = Environmental Protection Agency; NHEERL = National Health and Environmental Effects Research Laboratory; NCI = National Cancer Institute; NHLBI = National Heart, Lung, and Blood Institute; NIEHS = National Institute of Environmental Health Sciences; NIAID= National Institute of Allergy and Infectious Diseases; NIGMS = National Institute of General Medical Sciences; NIH = National Institute of Health

Source: FEDRIP 2007