

**TOXICOLOGICAL PROFILE FOR  
DIISOPROPYL METHYLPHOSPHONATE**

**U.S. DEPARTMENT OF HEALTH AND HUMAN SERVICES  
Public Health Service  
Agency for Toxic Substances and Disease Registry**

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### **UPDATE STATEMENT**

Toxicological profiles are revised and republished as necessary, but no less than once every three years. For information regarding the update status of previously released profiles, contact ATSDR at:

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Division of Toxicology/Toxicology Information Branch  
1600 Clifton Road NE, E-29  
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## FOREWORD

This toxicological profile is prepared in accordance with guidelines\* developed by the Agency for Toxic Substances and Disease Registry (ATSDR) and the Environmental Protection Agency (EPA). The original guidelines were published in the *Federal Register* on April 17, 1987. Each profile will be revised and republished as necessary.

The ATSDR toxicological profile succinctly characterizes the toxicologic and adverse health effects information for the hazardous substance described therein. Each peer-reviewed profile identifies and reviews the key literature that describes a hazardous substance's toxicologic properties. Other pertinent literature is also presented, but is described in less detail than the key studies. The profile is not intended to be an exhaustive document; however, more comprehensive sources of specialty information are referenced.

The focus of the profiles is on health and toxicologic information; therefore, each toxicological profile begins with a public health statement that describes, in nontechnical language, a substance's relevant toxicological properties. Following the public health statement is information concerning levels of significant human exposure and, where known, significant health effects. The adequacy of information to determine a substance's health effects is described in a health effects summary. Data needs that are of significance to protection of public health are identified by ATSDR and EPA.

Each profile includes the following:

- (A) The examination, summary, and interpretation of available toxicologic information and epidemiologic evaluations on a hazardous substance to ascertain the levels of significant human exposure for the substance and the associated acute, subacute, and chronic health effects;
- (B) A determination of whether adequate information on the health effects of each substance is available or in the process of development to determine levels of exposure that present a significant risk to human health of acute, subacute, and chronic health effects; and
- (C) Where appropriate, identification of toxicologic testing needed to identify the types or levels of exposure that may present significant risk of adverse health effects in humans.

The principal audiences for the toxicological profiles are health professionals at the Federal, State, and local levels; interested private sector organizations and groups; and members of the public.

This profile reflects ATSDR's assessment of all relevant toxicologic testing and information that has been peer-reviewed. Staff of the Centers for Disease Control and Prevention and other Federal scientists have also reviewed the profile. In addition, this profile has been peer-reviewed by a nongovernmental panel and was made available for public review. Final responsibility for the contents and views expressed in this toxicological profile resides with ATSDR.



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### \*Legislative Background

The toxicological profiles are developed in response to the Super-fund Amendments and Reauthorization Act (SARA) of 1986 (Public Law 99-499) which amended the Comprehensive Environmental Response, Compensation, and Liability Act of 1980 (CERCLA or Super-fund). Section 211 of SARA also amended Title 10 of the U. S. Code, creating the Defense Environmental Restoration Program. Section 2704(a) of Title 10 of the U. S. Code directs the Secretary of Defense to notify the Secretary of Health and Human Services of not less than 25 of the most commonly found unregulated hazardous substances at defense facilities. Section 2704(b) of Title 10 of the U. S. Code directs the Administrator of the Agency for Toxic Substances and Disease Registry (ATSDR) to prepare a toxicological profile for each substance on the list provided by the Secretary of Defense under subsection (b).

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**THE PROFILE HAS UNDERGONE THE FOLLOWING ATSDR INTERNAL REVIEWS:**

1. Green Border Review. Green Border review assures consistency with ATSDR policy.
2. Health Effects Review. The Health Effects Review Committee examines the health effects chapter of each profile for consistency and accuracy in interpreting health effects and classifying end points.
3. Minimal Risk Level Review. The Minimal Risk Level Workgroup considers issues relevant to substance-specific minimal risk levels (MRLs), reviews the health effects database of each profile, and makes recommendations for derivation of MRLs.





## PEER REVIEW

A peer review panel was assembled for diisopropyl methylphosphonate. The panel consisted of the following members:

1. Dr. Arthur Gregory, Private Consultant, Sterling, VA
2. Dr. Rudy Jaeger, President and Principal Scientist, Environmental Medicine, Inc., Westwood, NJ
3. Dr. Norbert Page, Private Consultant, Gaithersburg, MD

These experts collectively have knowledge of diisopropyl methylphosphonate's physical and chemical properties, toxicokinetics, key health end points, mechanisms of action, human and animal exposure, and quantification of risk to humans. All reviewers were selected in conformity with the conditions for peer review specified in Section 104(i)(13) of the Comprehensive Environmental Response, Compensation, and Liability Act, as amended.

Scientists from the Agency for Toxic Substances and Disease Registry (ATSDR) have reviewed the peer reviewers' comments and determined which comments will be included in the profile. A listing of the peer reviewers' comments not incorporated in the profile, with a brief explanation of the rationale for their exclusion, exists as part of the administrative record for this compound. A list of databases reviewed and a list of unpublished documents cited are also included in the administrative record.

The citation of the peer review panel should not be understood to imply its approval of the profile's final content. The responsibility for the content of this profile lies with the ATSDR.



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## 1. PUBLIC HEALTH STATEMENT

This public health statement tells you about diisopropyl methylphosphonate and the effects of exposure.

The Environmental Protection Agency (EPA) identifies the most serious hazardous waste sites in the nation. These sites make up the National Priorities List (NPL) and are the sites targeted for long-term federal clean-up. Diisopropyl methylphosphonate has been found in at least 2 of the 1,416 current or former NPL sites. However, it's unknown how many NPL sites have been evaluated for this substance. This is important because exposure to this substance may harm you and because these sites may be sources of exposure.

When a substance is released from a large area, such as an industrial plant, or from a container, such as a drum or bottle, it enters the environment. This release does not always lead to exposure. You are exposed to a substance only when you come in contact with it. You may be exposed by breathing, eating, or drinking the substance or by skin contact.

If you are exposed to diisopropyl methylphosphonate, many factors determine whether you'll be harmed. These factors include the dose (how much), the duration (how long), and how you come in contact with it. You must also consider the other chemicals you're exposed to and your age, sex, diet, family traits, lifestyle, and state of health.

### 1.1 WHAT IS DIISOPROPYL METHYLPHOSPHONATE?

Diisopropyl methylphosphonate is a colorless liquid at normal temperatures. It is also known as methyl-,bis-(1-methyl-ethyl)ester, phosphonic acid, and methyl-diisopropyl ester.

Diisopropyl methylphosphonate, or DIMP, is a chemical by-product resulting from the manufacture and detoxification of GB (also called Sarin), a nerve gas that the Army produced

from 1953 to 1957. A chemical by-product is a chemical that is formed while making another substance. You might find diisopropyl methylphosphonate in places where GB has been produced, stored, or used, for example, the Rocky Mountain Arsenal (RMA) outside of Denver, Colorado. The RMA is where GB was produced for loading into chemical warfare munitions. Production of GB was discontinued in 1957, and it is not likely that GB, or diisopropyl methylphosphonate, will be produced in the United States in the future because of signing of a chemical treaty that bans not only the use but also the production and stockpiling of poison gases. Diisopropyl methylphosphonate is a colorless liquid. A small amount (0.1%, or 1g of DIMP in 1,000 mL of water) of it dissolves in water, but some scientists have found it to be more soluble (8%). Once it is dissolved, little, if any, evaporates into the air. More information on the chemical and physical properties of diisopropyl methylphosphonate is found in Chapter 3. More information on the production and use of diisopropyl methylphosphonate is found in Chapter 4.

## **1.2 WHAT HAPPENS TO DIISOPROPYL METHYLPHOSPHONATE WHEN IT ENTERS THE ENVIRONMENT?**

Diisopropyl methylphosphonate was released into the environment at the RMA between 1953 and 1957. While most discharged diisopropyl methylphosphonate entered the groundwater at the RMA, some also entered the surface water and soils. It is unlikely that diisopropyl methylphosphonate escaped to the air because it does not evaporate easily. Diisopropyl methylphosphonate does not break down in the environment very quickly and can remain in water and soil for years. The flow of water during irrigation can carry diisopropyl methylphosphonate through the soil. Plants can take up diisopropyl methylphosphonate and store or accumulate it in leaves and DIMP may enter the food chain when animals eat these plants. However, because DIMP is rapidly changed to isopropyl methylphosphonic acid (IMPA) by animals that eat it, it is unlikely that DIMP will be present further up the food chain. Muman exposure may also occur by consumption of home-grown fruits and vegetables irrigated by DIMP-contaminated water. For more information on what happens to diisopropyl methylphosphonate in the environment, see Chapters 4 and 5.

### **1.3 HOW MIGHT I BE EXPOSED TO DIISOPROPYL METHYLPHOSPHONATE?**

Exposure of the general population to diisopropyl methylphosphonate is expected to be rare and even then in small amounts. Only people living near a site where DIMP is found (such as RMA) may be exposed to this substance.

Drinking or showering with water containing diisopropyl methylphosphonate, or eating vegetables irrigated with it are the most likely ways that a person can be exposed to it. Water contaminated with DIMP has been found only at or near the RMA. In 1974 diisopropyl methylphosphonate was first measured in the groundwater at RMA. The reported amounts ranged from 0.5 parts per billion (ppb) to 44,000 ppb (1 ppb is equivalent to one part by weight of diisopropyl methylphosphonate in a billion parts by volume of water or soil). In 1989, soil samples from some areas in the RMA were reported to contain approximately 50 ppb to 240 ppb of diisopropyl methylphosphonate. During the most recent comprehensive groundwater sampling at RMA in 1995, diisopropyl methylphosphonate concentrations in groundwater were reported to range from less than 0.110 ppb to 965 ppb. It is possible that exposure to diisopropyl methylphosphonate might occur by eating plants that absorbed it from water or soil.

Please refer to Chapter 5 for more information on how you might be exposed to diisopropyl methylphosphonate.

### **1.4 HOW CAN DIISOPROPYL METHYLPHOSPHONATE ENTER AND LEAVE MY BODY?**

Diisopropyl methylphosphonate may enter your body after drinking water contaminated with it or it may enter through your skin. You could be exposed to diisopropyl methylphosphonate if you drink water that contains the chemical near the RMA. Exposure of the general population to diisopropyl methylphosphonate is not likely to occur. Studies in animals show that diisopropyl methylphosphonate enters the bloodstream relatively quickly following oral exposure. Animal

studies also show that once diisopropyl methylphosphonate is inside the body, it is changed almost completely into isopropyl methylphosphonic acid (IMPA) and leaves the body in the urine. Chapter 2 has more information on how diisopropyl methylphosphonate can enter and leave the body.

## **1.5 HOW CAN DIISOPROPYL METHYLPHOSPHONATE AFFECT MY HEALTH?**

Scientists use many tests to protect the public from the harmful effects of toxic chemicals and to find ways to treat people who have been harmed.

One way to see if a chemical will hurt people is to learn how the chemical is absorbed, changed, and released by the body; for some chemicals, animal testing may be necessary. Animal testing may also be used to identify health effects such as cancer or birth defects. Without laboratory animals, scientists would lose a basic method to get information needed to make wise decisions to protect public health. Scientists have the responsibility to treat research animals with care and compassion. Laws today protect the welfare of research animals, and scientists must comply with strict animal care guidelines.

Little is known about the human health effects of diisopropyl methylphosphonate. Skin rashes and other signs of irritation have been reported in some people who handled dead animals near a pond containing diisopropyl methylphosphonate and other chemicals, but it is not known which substances caused these effects.

Drinking large amounts of diisopropyl methylphosphonate kills animals. The amount needed to cause death in humans is not known for sure. Animal studies have shown no evidence that drinking or eating diisopropyl methylphosphonate causes fertility problems or birth defects. Animal studies have shown that eating diisopropyl methylphosphonate can affect some liver enzymes (indicating a response by the liver). However, test animals showed no liver disease. While most animal studies have shown only minimal toxic effects below a certain level of

exposure, there is some evidence that diisopropyl methylphosphonate can cause effects on the blood and nervous systems at high levels. Overall, however, it is not a very toxic chemical.

See Chapter 2 for more information about the health effects of diisopropyl methylphosphonate.

### **1.6 IS THERE A MEDICAL TEST TO DETERMINE WHETHER I HAVE BEEN EXPOSED TO DIISOPROPYL METHYLPHOSPHONATE?**

Once inside the body, diisopropyl methylphosphonate is rapidly converted to isopropyl methylphosphonic acid (IMPA), which is rapidly cleared from the blood. Laboratory tests can determine the amount of IMPA in the blood or urine. However, because IMPA leaves the body rapidly, these tests are useful only for recent exposure. It is helpful for your doctor to know whether there are other chemicals to which you have been exposed. See Chapters 2 and 6 for more information.

### **1.7 WHAT RECOMMENDATIONS HAS THE FEDERAL GOVERNMENT MADE TO PROTECT HUMAN HEALTH?**

The federal government develops regulations and recommendations to protect public health. Regulations can be enforced by law. Federal agencies that develop regulations for toxic substances include the Environmental Protection Agency (EPA), the Occupational Safety and Health Administration (OSHA), and the Food and Drug Administration (FDA). Recommendations provide valuable guidelines to protect public health but cannot be enforced by law. Federal organizations that develop recommendations for toxic substances include the Agency for Toxic Substances and Disease Registry (ATSDR) and the National Institute for Occupational Safety and Health (NIOSH).

Regulations and recommendations can be expressed in not-to-exceed levels in air, water, soil, or food that are usually based on levels that affect animals, then they are adjusted to help protect people. Sometimes these not-to-exceed levels differ among federal organizations because of

different exposure times (an 8-hour workday or a 24-hour day), the use of different animal studies, or other factors.

Recommendations and regulations are also periodically updated as more information becomes available. For the most current information, check with the federal agency or organization that provides it. Some regulations and recommendations for diisopropyl methylphosphonate include the following:

The EPA has issued guidelines concerning recommendations for levels of diisopropyl methylphosphonate that may be in drinking water, including Health Advisories. It has concluded that a lifetime of exposure to drinking water containing 0.6 mg/L (1 mg/L = 1 milligram of diisopropyl methylphosphonate in 1 liter of water) of diisopropyl methylphosphonate would not result in any noncancer harmful health effects. This amount, 0.6 mg/L, is the same as 600 parts per billion parts of water. The EPA has determined that diisopropyl methylphosphonate is unclassifiable as to its ability to cause cancer in humans, that is, there are not enough data to classify the ability of diisopropyl methylphosphonate to cause cancer in humans. For more information, see Chapter 7.

## **1.8 WHERE CAN I GET MORE INFORMATION?**

If you have any more questions or concerns, please contact your community or state health or environmental quality department or

Agency for Toxic Substances and Disease Registry  
Division of Toxicology  
1600 Clifton Road NE, Mailstop E-29  
Atlanta, Georgia 30333



\*Information line and technical assistance

Phone: 1-800-447-1544

Fax: (404) 639-6315 or 6324

ATSDR can also tell you the location of occupational and environmental health clinics. These clinics specialize in recognizing, evaluating, and treating illnesses resulting from exposure to hazardous substances.

\*To order toxicological profiles, contact

National Technical Information Service

5285 Port Royal Road

Springfield, VA 22161

Phone (800) 553-6847 or (703) 487-4650



## 2. HEALTH EFFECTS

### 2.1 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 diisopropyl methylphosphonate. 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.

### 2.2 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”

## 2. HEALTH EFFECTS

effects and “serious” effects is considered to be important because it helps 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 or exposure levels below which no adverse effects 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.

Estimates of exposure levels posing minimal risk to humans (Minimal Risk Levels or MRLs) have been made for diisopropyl methylphosphonate. An MRL is defined as an estimate of daily human exposure to a substance that is likely to be without an appreciable risk of adverse effects (noncarcinogenic) over a specified duration of exposure. MRLs are derived when reliable and sufficient data exist to identify the target organ(s) of effect or the most sensitive health effect(s) for a specific duration within a given route of exposure. MRLs are based on noncancer health effects only and do not reflect a consideration of carcinogenic effects. MRLs can be derived for acute, intermediate, and chronic duration exposures for inhalation and oral routes. Appropriate methodology does not exist to develop MRLs for dermal exposure.

Although methods have been established to derive these levels (Barnes and Dourson 1988; EPA 1990), uncertainties are associated with these techniques. Furthermore, ATSDR acknowledges additional uncertainties inherent in the application of the procedures to derive less than lifetime MRLs. As an example, acute inhalation MRLs may not be protective for health effects that are delayed in development or result from repeated acute insults, such as hypersensitivity reactions, asthma, or chronic bronchitis. As these kinds of health effects data become available and methods to assess levels of significant human exposure improve, these MRLs will be revised.

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.

## 2. HEALTH EFFECTS

**2.2.1 Inhalation Exposure**

No studies were located regarding the following health effects in humans or animals after inhalation exposure to diisopropyl methylphosphonate.

**2.2.1.1 Death****2.2.1.2 Systemic Effects****2.2.1.3 Immunological and Lymphoreticular Effects****2.2.1.4 Neurological Effects****2.2.1.5 Reproductive Effects****2.2.1.6 Developmental Effects****2.2.1.7 Genotoxic Effects**

No studies were located regarding genotoxic effects in humans or animals after inhalation exposure to diisopropyl methylphosphonate.

Genotoxicity studies are discussed in Section 2.5.

**2.2.1.8 Cancer**

No studies were located regarding carcinogenic effects in humans or animals after inhalation exposure to diisopropyl methylphosphonate.

**2.2.2 Oral Exposure**

No data are available regarding health effects in humans after oral exposure to diisopropyl methylphosphonate. However, data are available regarding animals.

Results of these studies are discussed below and presented in Table 2-1 and Figure 2- 1.

TABLE 2-1. Levels of Significant Exposure to Diisopropyl Methylphosphonate - Oral

Key to figure <sup>a</sup>	Species (strain)	Exposure duration/ frequency (specific route)	System	NOAEL (mg/kg/day)	LOAEL (effect)		Reference
					Less serious (mg/kg/day)	Serious (mg/kg/day)	
<b>ACUTE EXPOSURE</b>							
<b>Death</b>							
1	Rat (Sprague- Dawley)	once (GW)				1125 M (LD <sub>50</sub> )	Hart 1976
						826 F (LD <sub>50</sub> )	
2	Rat (Fischer- 344)	once (G)				1000 M (3/5 died)	DOD 1991b
3	Rat (Fischer- 344)	3 d 1x/d (G)				800 M (3/7 died)	DOD 1991b
4	Mouse (Swiss- Webster)	once (GW)				1041 M (LD <sub>50</sub> )	Hart 1976
						1363 F (LD <sub>50</sub> )	
5	Mouse (B6C3F1)	3 d 1x/d (G)				2000 M (5/5 died)	DOD 1991a
6	Mink	once (G)				503 F (LD <sub>50</sub> )	Aulerich et al. 1979
7	Cow	once (C)				1000 (2/2 died)	Cysewski et al. 1981; Palmer et al. 1979
8	Duck (Mallard)	once (G)				1500 (3/12 died)	Jones et al. 1992
9	Duck (Mallard)	once (G)				1490 (LD <sub>50</sub> )	Aulerich et al. 1979

TABLE 2-1. Levels of Significant Exposure to Diisopropyl Methylphosphonate - Oral(continued)

Key to figure <sup>a</sup>	Species (strain)	Exposure duration/frequency (specific route)	System	NOAEL (mg/kg/day)	LOAEL (effect)		Reference
					Less serious (mg/kg/day)	Serious (mg/kg/day)	
<b>Systemic</b>							
10	Rat (Charles River COBS CD [SD] BR)	4 d (F)	Hepatic		150	(induction of microsomal enzymes leading to decreased hexobarbital sleeping time)	Hart 1976
			Bd Wt	150			
11	Rat (Sprague-Dawley)	10 d Gd 6-15 (F)	Bd Wt	300			Hart 1980
12	Rat (Sprague-Dawley)	once (GW)	Resp	632		928 (hyperemia of the lungs)	Hart 1976
13	Mouse (Swiss-Webster)	once (GW)	Resp	2000			Hart 1976
14	Dog (Beagle)	14 d (F)	Resp	38			Hart 1976
			Cardio	38			
			Hemato	38			
			Hepatic	38			
			Renal	38			
			Bd Wt	38			

TABLE 2-1. Levels of Significant Exposure to Diisopropyl Methylphosphonate - Oral (continued)

Key to <sup>a</sup> figure	Species (strain)	Exposure duration/ frequency (specific route)	System	NOAEL (mg/kg/day)	LOAEL (effect)		Reference
					Less serious (mg/kg/day)	Serious (mg/kg/day)	
15	Cow	once (C)	Resp	500		1000 (pulmonary emphysema)	Cysewski et al. 1981; Palmer et al. 1979
			Cardio	500		1000 (petechiae in the coronary grooves)	
			Gastro	500		1000 (acute gastroenteritis; ecchymotic hemorrhaging)	
			Renal	63	500 (mild congestion of the renal cortex)		
			Bd Wt	1000			
16	Duck (Mallard)	once (G)	Cardio		1500 (decrease in blood pressure)		Jones et al. 1992
17	Duck (Mallard)	once (G)	Bd Wt	1700	1800 (weight loss of 14.8%)		Aulerich et al. 1979
18	Duck (Mallard)	8 d (F)	Bd Wt	1007		1796 (57% decrease in body weight gain)	Aulerich et al. 1979
<b>Neurological</b>							
19	Rat (Sprague- Dawley)	once (GW)				430 (ataxia; decreased activity; prostration)	Hart 1976
20	Mouse (Swiss- Webster)	once (GW)				430 (decreased activity; prostration)	Hart 1976
21	Mouse (B6C3F1)	3 d 1x/d (G)				1000 M (coma)	DOD 1991a
22	Mink	once (G)		150 F		300 F (salivation; lethargy; immobilization)	Aulerich et al. 1979



TABLE 2-1. Levels of Significant Exposure to Diisopropyl Methylphosphonate - Oral (continued)

Key to figure <sup>a</sup>	Species (strain)	Exposure duration/frequency (specific route)	System	NOAEL (mg/kg/day)	LOAEL (effect)		Reference
					Less serious (mg/kg/day)	Serious (mg/kg/day)	
23	Cow	once (C)		500		1000 (tympanitis; ataxia; depression; prostration; engorgement of meningeal vessels; excess fluid in cerebral ventricles)	Cysewski et al. 1981; Palmer et al. 1979
<b>INTERMEDIATE EXPOSURE</b>							
<b>Systemic</b>							
24	Rat (Sprague-Dawley)	90 d (F)	Cardio	300			Hart 1976
			Hemato	300			
			Hepatic	300			
			Renal	300			
			Ocular	300			
			Bd Wt	300			
25	Rat (Sprague-Dawley)	30 wk (3 generations) (F)	Bd Wt	300			Hart 1980
26	Mouse (Swiss-Albino)	90 d (F)	Resp	273			Hart 1976
			Hepatic	273			
			Renal	273			
			Bd Wt	273			
27	Dog (Beagle)	90 d (F)	Cardio	75			Hart 1980
			Hemato	75 <sup>b</sup>			
			Hepatic	75			
			Renal	75			
			Ocular	75			
			Bd Wt	75			

TABLE 2-1. Levels of Significant Exposure to Diisopropyl Methylphosphonate - Oral (continued)

Key to figure <sup>a</sup>	Species (strain)	Exposure duration/frequency (specific route)	System	NOAEL (mg/kg/day)	LOAEL (effect)		Reference
					Less serious (mg/kg/day)	Serious (mg/kg/day)	
28	Mink	21 d (F)	Resp	17	201 M (decreased lung weights)		Aulerich et al. 1979
			Cardio	201	1852 M (decreased heart weights)		
			Hemato	17	201 (decreased lymphocytes)		
			Hepatic	201	1852 M (decreased liver weights)		
			Renal	201	1852 M (decreased kidney weights)		
			Bd Wt	1852			
29	Mink	49 wk (F)	Resp	95			Aulerich et al. 1979
			Cardio	95			
			Gastro	95			
			Hemato	95			
			Musc/skel	95			
			Hepatic	95			
			Renal	95			
			Bd Wt	95			
30	Mink (Ranch Wild)	90 d (F)	Hemato	345 M	747 M (shortened RBC survival; increased Heinz bodies; increased reticulocytes; reduced RBC counts)		Bucci et al. 1992, 1994
				455 F		907 F (shortened RBC survival; increased Heinz bodies; increased reticulocytes; reduced RBC counts)	
			Hepatic	1009 M			
				1264 F			
			Ocular	1009 M			
				1264 F			
			Bd Wt	1009 M			
				1264 F			

TABLE 2-1. Levels of Significant Exposure to Diisopropyl Methylphosphonate - Oral (continued)

Key to <sup>a</sup> figure	Species (strain)	Exposure duration/ frequency (specific route)	System	NOAEL (mg/kg/day)		LOAEL (effect) (mg/kg/day)		Reference
						Less serious	Serious	
31	Mink (Ranch Wild)	1-11 mo ad lib (F)	Hemato	45	M	262M (increased Heinz body counts)		Bucci et al. 1997
				57	F			
					Bd Wt	262	M	
				330	F			
<b>Neurological</b>								
32	Rat (Sprague- Dawley)	90 d (F)		300				Hart 1976
33	Dog (Beagle)	13 wk 7d/wk (F)		75				Hart 1980
34	Mink	21 d (F)		1852				Aulerich et al. 1979
35	Mink	49 wk (F)		95				Aulerich et al. 1979
36	Mink (Ranch Wild)	1-11 mo ad lib (F)		262	M			Bucci et al. 1997
				330	F			
<b>Reproductive</b>								
37	Rat (Sprague- Dawley)	30 wk (3 gener- ations) (F)		300				Hart 1980
38	Mink	49 wk (F)		95				Aulerich et al. 1979
39	Mink (Ranch Wild)	1-11 mo ad lib (F)		262	M			Bucci et al. 1997
				330	F			

TABLE 2-1. Levels of Significant Exposure to Diisopropyl Methylphosphonate - Oral (continued)

Key to <sup>a</sup> figure	Species (strain)	Exposure duration/ frequency (specific route)	System	NOAEL (mg/kg/day)	LOAEL (effect)		Reference
					Less serious (mg/kg/day)	Serious (mg/kg/day)	
<b>Developmental</b>							
40	Rat (Sprague- Dawley)	Gd 6-15 (F)		300			Hart 1980
41	Mink (Ranch Wild)	1-11 mo ad lib (F)		45 M 330 F	262M (decreased RBC in F2 male kits at 6 weeks)		Bucci et al. 1997
<b>Immuno/Lymphor</b>							
42	Mink	21 d (F)		1852			Aulerich et al. 1979
43	Mink	49 wk (F)		95			Aulerich et al. 1979
44	Mink (Ranch Wild)	1-11 mo ad lib (F)		262 M 330 F			Bucci et al. 1997
<b>CHRONIC EXPOSURE</b>							
<b>Systemic</b>							
45	Mink (Ranch Wild)	13 mo ad lib (F)	Hemato  Bd Wt	57 <sup>c</sup> F 330 F	330 F (increased Heinz bodies)		Bucci et al. 1997
<b>Neurological</b>							
46	Mink (Ranch Wild)	13 mo ad lib (F)		330 F			Bucci et al. 1997
<b>Reproductive</b>							
47	Mink (Ranch Wild)	1-11 mo ad lib (F)		330 F			Bucci et al. 1997

TABLE 2-1. Levels of Significant Exposure to Diisopropyl Methylphosphonate - Oral (continued)

Key to <sup>a</sup> figure	Species (strain)	Exposure duration/ frequency (specific route)	System	NOAEL (mg/kg/day)	LOAEL (effect)		Reference
					Less serious (mg/kg/day)	Serious (mg/kg/day)	
<b>Immuno/Lymphor</b>							
48	Mink (Ranch Wild)	13 mo ad lib (F)		330 F			Bucci et al. 1997

<sup>a</sup>The number corresponds to entries in Figure 2-1.

<sup>b</sup>An intermediate-duration Minimal Risk Level (MRL) of 0.8 mg/kg/day was derived based on this end point; dose of 75 mg/kg/day divided by an uncertainty factor of 100 (10 for extrapolation from animals to humans and 10 for human variability).

<sup>c</sup>A chronic-duration Minimal Risk Level (MRL) of 0.6 mg/kg/day based on this end point; dose of 57 mg/kg/day divided by an uncertainty factor of 100 (10 for extrapolation from animals to humans and 10 for human variability).

ad lib = ad libitum; Bd Wt = body weight; (C) = gelatin capsule; Cardio = cardiovascular; d = day(s); F = female; (F) = feed; (G) = gavage (type unspecified); Gastro = gastrointestinal; Gd = gestation day(s); (GW) = gavage in water; Hemato = hematological; LD<sub>50</sub> = lethal dose, 50% kill; LOAEL = lowest-observed-adverse-effect level; M = male; MCV = mean cell volume; mo = month(s); Musc/skel = musculoskeletal; NOAEL = no-observed-adverse-effect level; RBC = red blood cells; Resp = respiratory; wk = week(s); x = times(s)

**Figure 2-1. Levels of Significant Exposure to Diisopryl Methylphosphonate - Oral**  
**Acute (≤14 days)**

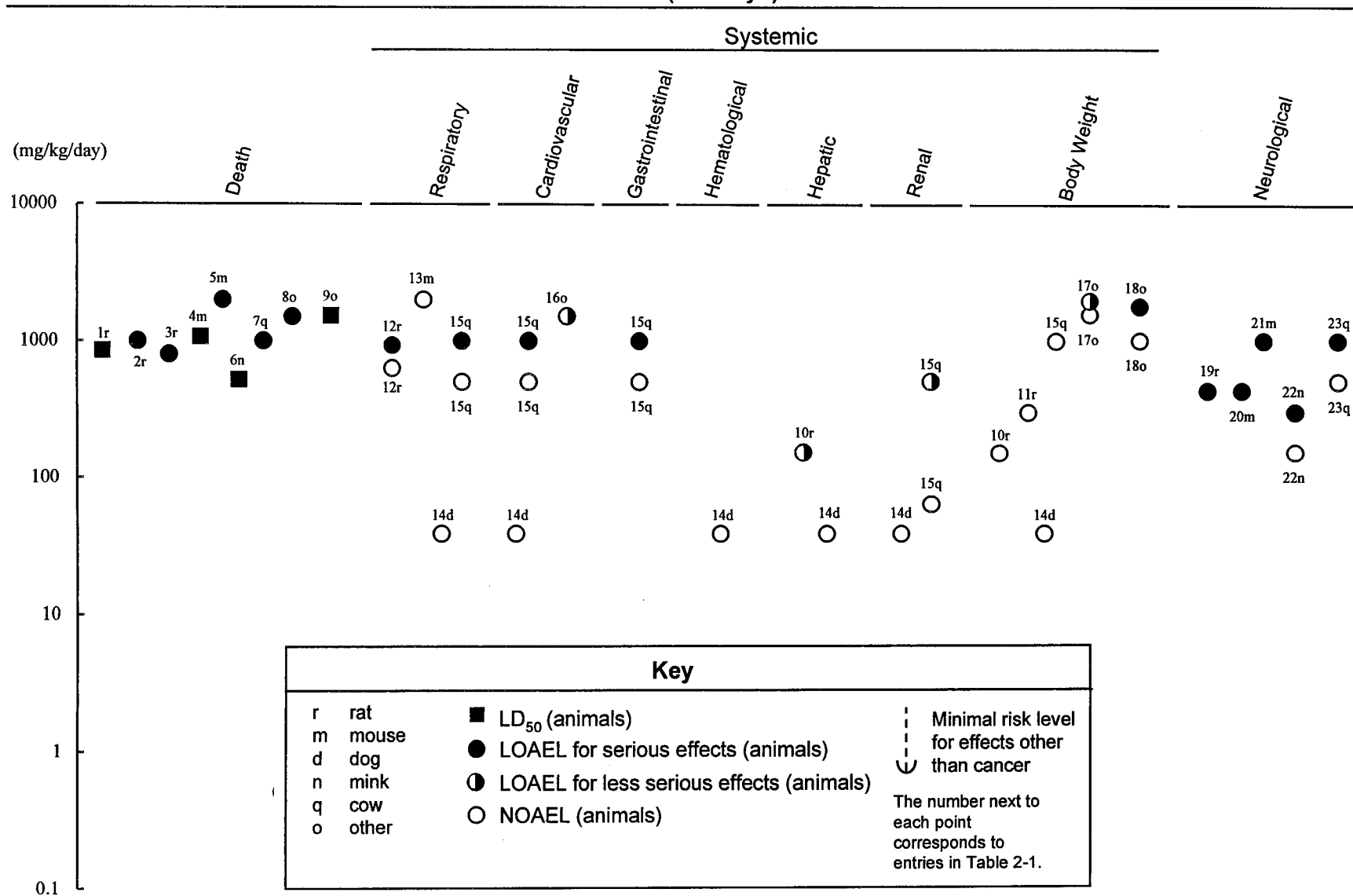
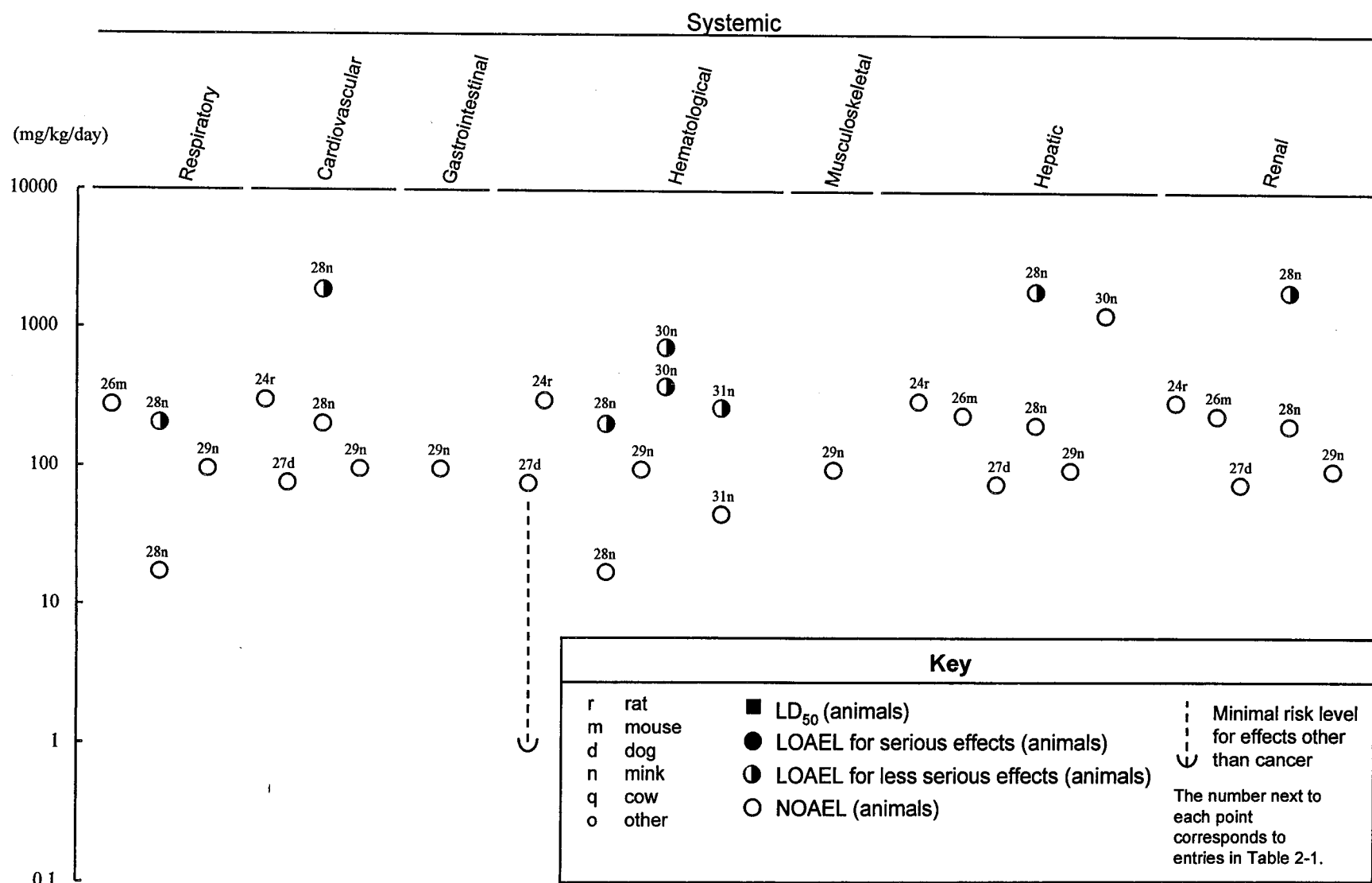


Figure 2-1. Levels of Significant Exposure to Diisopryl Methylphosphonate - Oral (cont.)  
Intermediate (15-364 days)



**Figure 2-1. Levels of Significant Exposure to Diisopryl Methylphosphonate - Oral (cont.)  
Intermediate (15-364 days)**

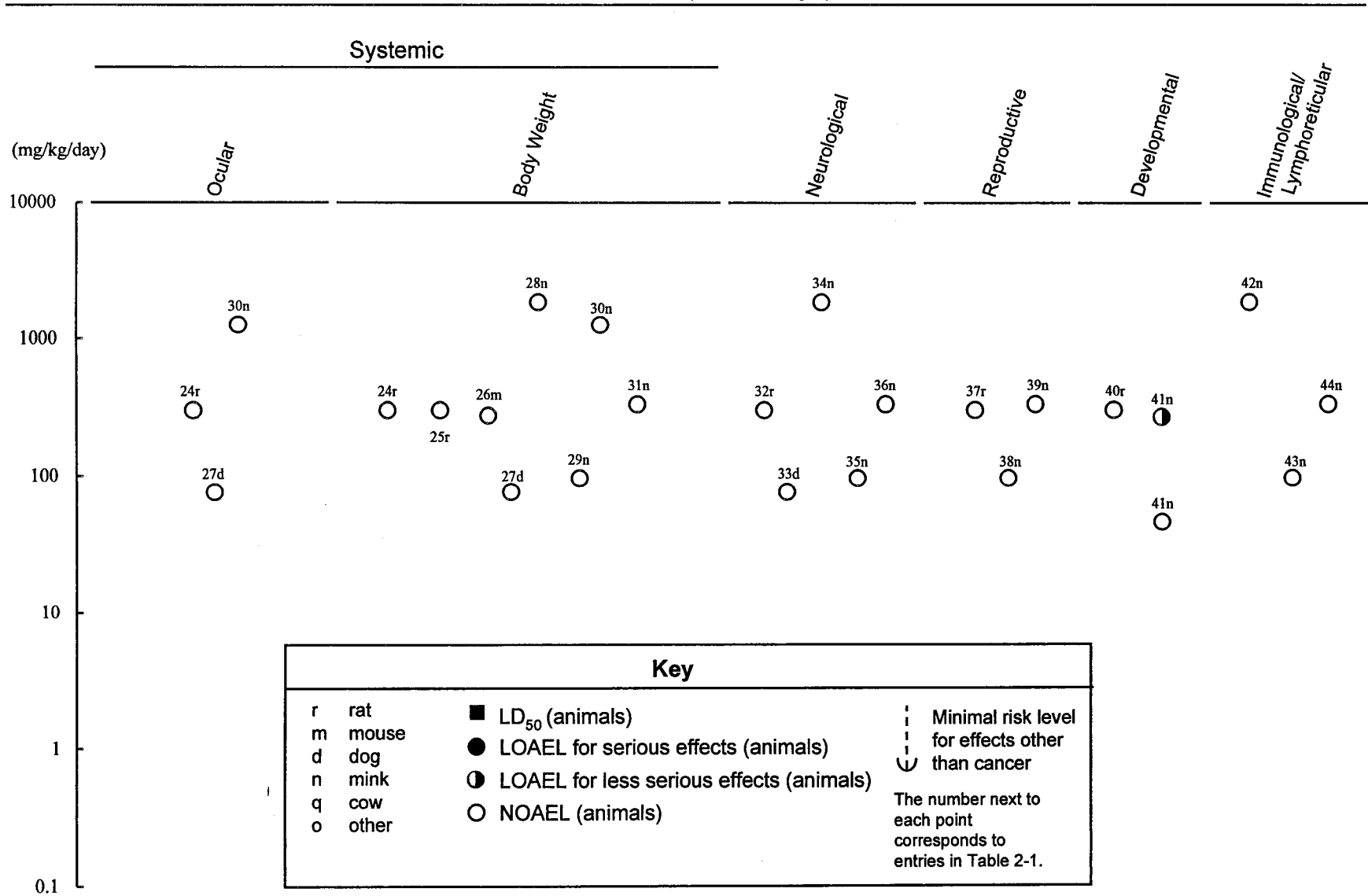
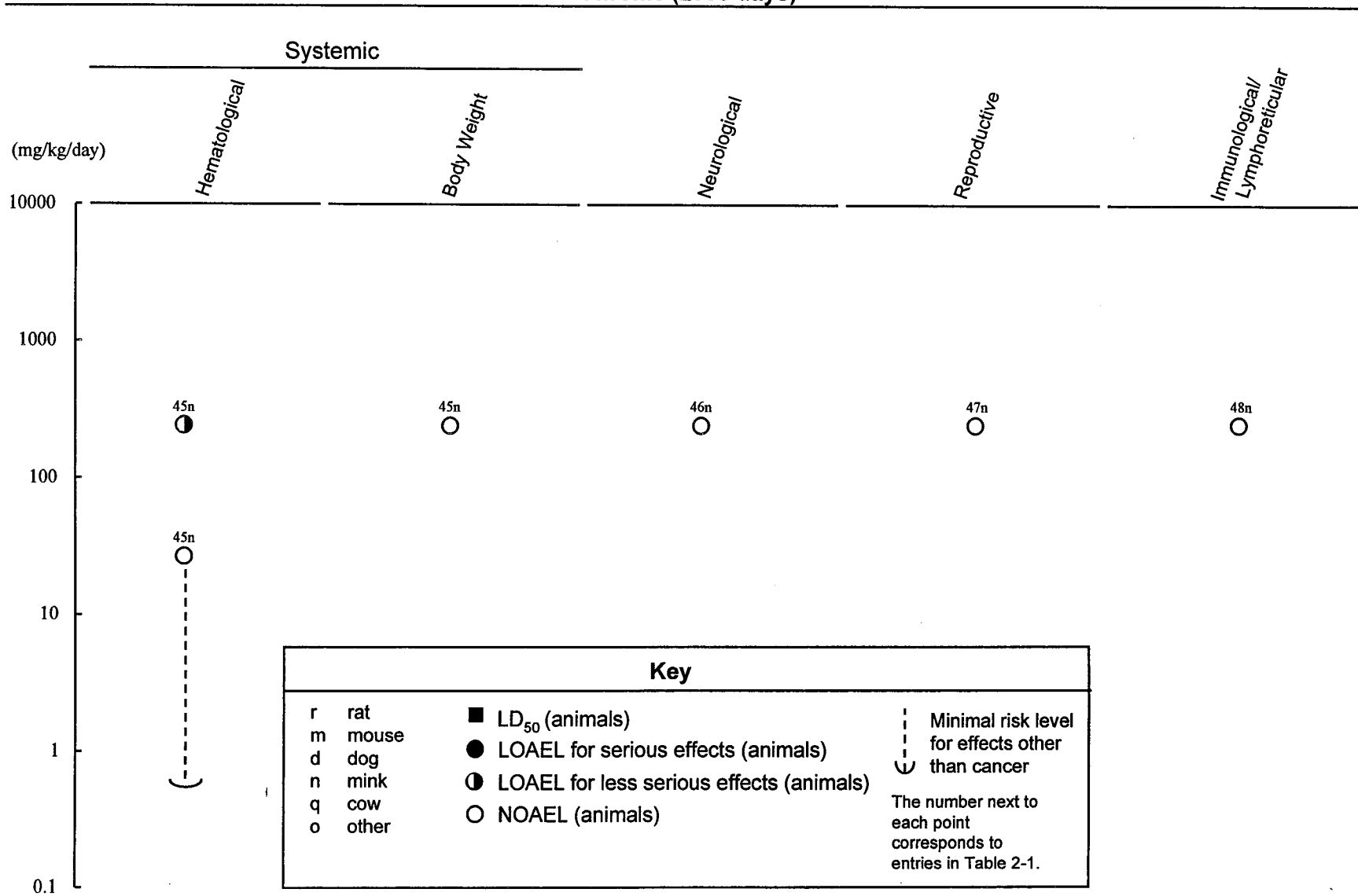




Figure 2-1. Levels of Significant Exposure to Diisopryl Methylphosphonate - Oral (cont.)

Chronic ( $\geq 365$  days)



## 2. HEALTH EFFECTS

**2.2.2.1 Death**

No studies were located regarding death in humans after oral exposure to diisopropyl methylphosphonate.

Three of five male Fisher-344 rats dosed once with 1,000 mg/kg diisopropyl methylphosphonate by gavage died (DOD 1991b). In a micronucleus assay using this species, 3 of 7 rats dosed on 3 successive days with 800 mg/kg died, but no deaths occurred at the next lower dose, 400 mg/kg (DOD 1991b). A repeat of this experiment in which rats were dosed with 400, 600, or 800 mg/kg for 3 days resulted in the deaths of 2 of 7 high-dose rats (DOD 1991 b). Oral LD<sub>50</sub>s of 1,125 and 826 mg/kg have been reported for male and female rats, respectively (Hart 1976). Single doses of diisopropyl methylphosphonate (430, 632, 928, 1,362, and 2,000 mg/kg) dissolved in polyethylene glycol 400 were administered by gastric intubation to 10 male and 10 female Sprague-Dawley rats at each dose level. The animals were examined for 14 days. By the day following the administration of the compound, all 10 males in the 1,362-mg/kg group and 9 of 10 males in the 2,000-mg/kg group had died (the remaining animal in the 2,000-mg/kg group died the following day). Similarly, all 10 females in the 1,362-mg/kg group and 9 of 10 females in the 2,000-mg/kg group had died (the remaining female died on day 2). Further, on the first 2 days subsequent to dosing 8 of 10 females but none of the 10 males in the 928-mg/kg group died. No deaths occurred in males or females administered 430 or 632 mg/kg. Signs of intoxication in both sexes included decreased activity, occasional ataxia, and prostration (Hart 1976).

Parallel experiments in Swiss Webster mice yielded LD<sub>50</sub>s of 1,041 and 1,363 mg/kg in males and females, respectively (Hart 1976). Mortalities included 2 of 10 females in the 430-mg/kg group, 2 of 10 females and 3 of 10 males in the 928-mg/kg group, 3 of 10 females and 9 of 10 males in the 1,362-mg/kg group, and all 10 males and all 10 females in the 2,000-mg/kg group. As is the case with rats, nearly all deaths occurred on the 1st day following gavage treatment with diisopropyl methylphosphonate. Signs of intoxication included decreased activity and prostration (Hart 1976). In a range-finding study for a bone marrow micronucleus assay, 5 of 5 male mice receiving 2,000 mg/kg for 3 days by gavage died, but no deaths occurred in mice receiving 1,000 mg/kg (DOD 1991a).

An LD<sub>50</sub> of 503 mg/kg was calculated from the results of a study in which adult female mink were administered single doses of diisopropyl methylphosphonate by gavage (doses of 75, 150, 300, 450, 500,

## 2. HEALTH EFFECTS

550, and 600 mg/kg) (Aulerich et al. 1979). Deaths occurred within a few hours after dosing. Animals that did not die recovered completely within several hours after administration of the compound.

In a 14-day lethality study of calves dosed with diisopropyl methylphosphonate at 62.5, 125, 250, 500, or 1,000 mg/kg via gelatin capsules gavaged using a balling gun, both of the two calves in the highest dose group died (Cysewski et al. 1981; Palmer et al. 1979). No animals in any of the other dose groups died. The study authors calculated the LD<sub>50</sub> to be approximately 750 mg/kg.

An LD<sub>50</sub> of 1,490 mg/kg was derived from a study using Mallard ducks in which groups of 10 of each sex were given a single dose of diisopropyl methylphosphonate by gavage at doses that ranged from 1,300 to 1,800 mg/kg and were observed for 14 days (Aulerich et al. 1979). The study authors attributed the deaths of many ducks to drowning on the copious amounts of saliva that were generated following exposure rather than true systemic toxicity. Necropsy showed no gross pathological changes.

No mortality occurred, nor were there judged to be any signs of toxicity, in beagle dogs receiving diisopropyl methylphosphonate in the diet for 2 weeks at 0, 4, 13, or 38 mg/kg/day (each dose group consisted of one male and one female) (Hart 1976). In a developmental toxicity study, no deaths occurred in female rats receiving diisopropyl methylphosphonate in the diet at 0, 10, 30, or 300 mg/kg/day on days 6-15 of gestation (Hart 1980).

No significant mortality related to diisopropyl methylphosphonate was noted in groups of 5 male and 5 female juvenile pastel mink receiving dietary doses of 0.2, 2, 17, 201, or 1,852 mg/kg/day for 21 days (Aulerich et al. 1979). A comparison of this study with the LD<sub>50</sub> study in mink suggests that diisopropyl methylphosphonate is more toxic following gavage administration than following administration in the diet.

No mortality was noted in male or female rats that received diisopropyl methylphosphonate in drinking water at 0.0090 or 0.9 mg/kg/day for 13 weeks (males) or 19 weeks (females) (Hardisty et al. 1977). EPA (1989) indicated that actual doses could not be verified, considered the study inappropriate for human health risk assessment, and rejected it for use in the development of a hazard advisory. No mortality occurred and no toxic effects were noted in beagle dogs (4 per sex per dose group) that received diisopropyl methylphosphonate in the diet (0, 4, 38, or 75 mg/kg/day) for 13 weeks (Hart 1980).

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No deaths or evidence of toxicity were attributable to diisopropyl methylphosphonate administered for 26 weeks in the drinking water of rats at concentrations of 0.6 ppb, 6.0 ppb, 10 ppm, and 1,000 ppm ( $6.6 \times 10^{-7}$ ,  $6.6 \times 10^{-5}$ , 0.011, and 1.1 mg/kg/day, respectively) (Army 1978). It should be noted that there is some confusion concerning the concentration units used in this study (EPA 1989). EPA (1989) states that conversions between ppm and mg/L were incorrectly calculated using the air conversion factor. EPA (1989) also indicates that analysis of the diisopropyl methylphosphonate used in this study determined that it was only 65% pure. Therefore, results from the Army (1978) study are considered inappropriate for human health risk assessment. No deaths of adult rats were recorded in a three-generation study of reproductive effects in rats receiving diisopropyl methylphosphonate in the diet at 0, 30, or 300 mg/kg/day (Hart 1980).

In a 90-day toxicity study in which 179 mice received diisopropyl methylphosphonate in the diet at doses of 0, 27, 91, or 273 mg/kg/day, two deaths occurred in the 91 mg/kg/day male group. Since no deaths were observed in the 273-mg/kg/day group and no other signs of toxicity were observed, it was concluded that there was no evidence of toxicity in this 90-day study (Hart 1976). Four male rats died (1 control, 1 low-dose, and 2 high-dose animals) out of a total of 256 animals in a 90-day study, in which rats received diisopropyl methylphosphonate in the diet at doses of 0, 30, 100, or 300 mg/kg/day. The deaths were neither dose nor duration related and were not considered of toxicologic importance (Hart 1976).

In a reproductive study in which groups of 25 female and 6 male dark variety mink were fed diisopropyl methylphosphonate in the diet at 0, 11, 37, or 95 mg/kg/day for approximately 49 weeks, an increase in deaths occurred in females that was statistically significant at the high dose (Aulerich et al. 1979). No control females died, while 2 of 23, 3 of 24, and 5 of 24 died at the low, middle, and high doses, respectively. The first death that occurred was in the lowest dose group. No adverse symptoms were noted before the deaths occurred. Three of the five deaths at the high dose occurred between the time of mating and lactation. Among males, 1 of 6 controls and 1 of 6 treated at 11 mg/kg/day died. All male mink treated at the higher doses survived.

Although the increase in deaths in female mink was statistically significant at the high dose, it was not clear if the deaths were treatment related. In a concurrent study that was conducted to assess the toxicity of dicyclopentadiene, which used mink from the same lot, the mortality in the untreated female mink was 4 of 24, with 2 mink dying between the time of mating and lactation. It should also be noted that mink have a relatively high natural mortality (EPA 1989). The natural mortality for 1st-year mink in a commercial fur ranch operation approaches 6% annually, and up to 15% of lactating females may die in the late gestation

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period from “nursing sickness” (Schneider and Hunter 1993). Although it is not clear when the mink in the Aulerich et al. (1979) study died in relation to lactation, some female mink did die during the time period that included lactation. Further evidence that the deaths of mink may not have been related to diisopropyl methylphosphonate treatment comes from a second study in mink (Bucci et al. 1992, 1994). In this 90-day study, no mortality was observed in male or female mink receiving diisopropyl methylphosphonate in the diet at doses of 0, 7, 63, 345, 749, or 1,009 mg/kg/day for males and 0, 9, 82, 455, 908, or 1,264 mg/kg/day for females. Although the Bucci et al. (1992, 1994) study lacks a breeding period, it was a better controlled study than the Aulerich et al. (1979) study. In the Bucci et al. (1992, 1994) study, diisopropyl methylphosphonate was measured in the diet, food intake was measured weekly, and the mink were housed in a laboratory. In the Aulerich et al. (1979) study, diisopropyl methylphosphonate dietary levels were not measured, food intake was estimated only once every 2 weeks, and the animals were housed outside in pens.

Recently, a two-generation reproductive study in mink was performed by Bucci et al. (1997). In the parental ( $F_0$ ) generation, groups of 7 male and 35 female brown Ranch Wild mink were fed diisopropyl methylphosphonate in the diet at 0, 15, 47, or 285 mg/kg/day (males) or 0, 26, 85, or 460 mg/kg/day (females) for 4 weeks (males) or 4 months (females). Two groups of control animals were used. However, the study authors noted that the DIIMP consumption calculated for the  $F_1$  generation should be used for extrapolation to humans, instead of the values obtained for  $F_0$  animals, because body weight and food consumption were not monitored for  $F_0$  animals until they were brought inside from the farm at 8 months of age. Furthermore, the  $F_1$  data offers a more conservative estimate, because the exposure was over a longer period of time. Although 3.5% (6/175) of the  $F_0$  females (1 control, 2 low dose, 2 mid dose, and 1 high dose) died before the scheduled sacrifice, all of the animals except 1 mid dose animal showed signs of a stress syndrome characterized by stopping eating, lethargy, and weight loss that are associated with mink.

In the  $F_1$  generation, 13 males and 35 females were used per group. Concentrations of diisopropyl methylphosphonate in feed corresponded to 0, 16, 45, or 262 mg/kg/day (males) or 0, 20, 57, or 330 mg/kg/day (females) for either 8 months (males) or 13 months (females). Again, two groups of control animals were used. In the  $F_1$  generation, 4.6% (8/175) of the females died before the scheduled sacrifice. Six of these (1 in each control group, 1 each in the low- and mid-dose groups, and 2 in the high-dose group) died of a stress syndrome common in minks that was induced by a slight anesthesia overdose. One mid-dose male also died of the same syndrome. A 7th female from the low-dose group, which was prone to seizures after being stressed during procedures such as cage cleaning, died unexpectedly after such a seizure. The 8th animal, a high-dose female who had shown signs of stress syndrome, was found to have a ruptured uterus.

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Because the deaths either occurred early in the study, were randomly distributed among dose groups, or were associated with a stress syndrome common among mink, they are not believed to result from DIMP exposure. Thus, it appears that the deaths observed in the previous study by Aulerich et al. (1979) were not treatment related.

#### 2.2.2.2 Systemic Effects

The highest NOAEL and all LOAEL values from each reliable study for systemic effects in each species and duration category are recorded in Table 2-1 and plotted in Figure 2-1.

No studies were located regarding musculoskeletal or dermal effects in humans or animals after oral exposure to diisopropyl methylphosphonate.

**Respiratory Effects.** No studies were located regarding respiratory effects in humans after oral exposure to diisopropyl methylphosphonate.

The respiratory system does not appear to be a target of diisopropyl methylphosphonate. Exposure has only occasionally resulted in respiratory effects, and the effects did not appear to be dose or treatment related. Necropsy of both male and female rats that died as the result of a single dose (928, 1,362, or 2,000 mg/kg) of diisopropyl methylphosphonate, administered by gastric intubation, revealed some hyperemia of the lungs; however, most animals displayed no abnormalities (Hart 1976). Changes in the lungs were not observed in rats that survived treatment at 928 mg/kg or at lower doses (430, 632 mg/kg). No abnormal necropsy findings were noted in Swiss Webster mice dosed similarly (430, 632, 928, 1,362, 2,000 mg/kg) in a companion study (Hart 1976). No important abnormalities were noted in the necropsy of rats receiving diisopropyl methylphosphonate in the diet (0, 30, 100, or 300 mg/kg/day) for 90 days (Hart 1976). No noteworthy deviations were noted in the necropsy or histopathology examination of male or female beagles receiving dietary doses of 0, 4, 13, or 38 mg/kg/day for 14 days (Hart 1976). No gross lesions were noted in rats subsequent to receiving drinking water containing diisopropyl methylphosphonate at doses of  $6.6 \times 10^{-7}$ ,  $6.6 \times 10^{-5}$ , 0.011, or 1.1 mg/kg/day (Army 1978). However, as discussed in Section 2.2.2.1, there is some confusion concerning the concentration units and purity of the diisopropyl methylphosphonate used in the Army (1978) study (EPA 1989). Therefore, results from the Army (1978) study are considered inappropriate for human health risk assessment.

## 2. HEALTH EFFECTS

In a study of calves dosed with diisopropyl methylphosphonate at 62.5, 125, 250, 500, or 1,000 mg/kg via gelatin capsules placed with a balling gun, calves at the highest dose level displayed pulmonary emphysema upon autopsy (Palmer et al. 1979). No respiratory effects were observed at sublethal dose levels.

Male juvenile pastel mink receiving diisopropyl methylphosphonate in the diet at doses of 201 or 1,852 mg/kg/day for 21 days demonstrated significantly lower lung weights (toxicity is usually, but not always, associated with increased lung weight), although there were no significant changes observed during necropsy (Aulerich et al. 1979). The study authors concluded that the compound was nontoxic in the 21-day test. Respiratory effects were not observed in mink treated with diisopropyl methylphosphonate in the diet at doses of 95 mg/kg/day for 49 weeks (Aulerich et al. 1979).

**Cardiovascular Effects.** No studies were located regarding cardiovascular effects in humans after oral exposure to diisopropyl methylphosphonate.

Mean blood pressure was decreased in male and female Mallard ducks given a single dose of 1,500 mg/kg of diisopropyl methylphosphonate via proventricular intubation (Iones et al. 1992). The proventriculus is a glandular stomach that precedes the gizzard in birds. However, pulse pressure and heart rate were not affected. Therefore, it is likely that cardiac output and/or resistance were decreased. The study authors speculated that diisopropyl methylphosphonate acts in a mechanism similar to that of meprobamate and glycerol guaiacolate (two psychotropic agents), i.e., by depressing or blocking nerve impulse transmission at the internuncial neuron level of the spinal cord, brain stem, and subcortical areas of the central nervous system.

There is no evidence from animal studies that diisopropyl methylphosphonate directly affects the cardiovascular system except at very high doses (Palmer et al. 1979). No anomalous necropsy or histopathological effects were noted in rats that received diisopropyl methylphosphonate in the diet at doses of 0, 30, 100, or 300 mg/kg/day for 90 days (Hart 1976). Similarly, no abnormalities were noted in mice that received the compound in the diet at doses of 0, 27, 91, or 273 mg/kg/day for 90 days (Hart 1976). In this study, beagle dogs were also treated with the compound in the diet at doses of 0, 4, 13, or 38 mg/kg/day for 14 days (Hart 1976). No deviation in heart weight or abnormal histopathological findings were reported in beagles that received diisopropyl methylphosphonate in the diet at doses of 0, 4, 38, or 75 mg/kg/day for 13 weeks (Hart 1980).

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In a study of calves given a single dose of diisopropyl methylphosphonate at 62.5, 125, 250, 500, or 1,000 mg/kg via gelatin capsules placed with a balling gun, animals that died at the highest dose level displayed petechiae in the coronary grooves and ecchymotic hemorrhaging of the gastrointestinal tract upon autopsy (Palmer et al. 1979). No cardiovascular effects were observed at sublethal dose levels.

No significant changes in heart weight or histopathological findings were noted in mink receiving diisopropyl methylphosphonate in the diet at doses of 0.2, 2, 17,201, or 1,852 mg/kg/day for 21 days (Aulerich et al 1979). Gross and histopathological examination of mink that received 11, 37, or 95 mg/kg/day for 49 weeks revealed no consistent pathological changes (Aulerich et al. 1979).

**Gastrointestinal Effects.** No studies were located regarding gastrointestinal effects in humans after oral exposure to diisopropyl methylphosphonate.

In a study of calves dosed with diisopropyl methylphosphonate at 62.5, 125, 250, 500, or 1,000 mg/kg via gelatin capsules placed with a balling gun, calves that died at the highest dose level displayed acute gastroenteritis and ecchymotic hemorrhaging upon necropsy (Palmer et al. 1979). No gastrointestinal effects were observed at sublethal dose levels.

**Hematological Effects.** No studies were located regarding hematological effects in humans after oral exposure to diisopropyl methylphosphonate.

Hematological effects were noted in several animal studies. A few instances of significant differences were noted in the hematocytology (red blood cell [BBC] count, packed cell volume, hemoglobin, leukocyte count, and differential leukocyte count) in rats that had received diisopropyl methylphosphonate in the diet at doses of 0, 30, 100, or 300 mg/kg/day for 90 days. However, because the differences were so scattered and lacked clear dose response, they were considered of no toxicological importance (Hart 1976). Beagles that had been treated with the compound in the diet at doses of 4, 13, or 38 mg/kg/day for 14 days demonstrated values that were within normal limits for hemoglobin, hematocrit, BBC count, total leukocyte count, and differential leukocyte count (Hart 1976). In a 3-month study of beagles receiving diisopropyl methylphosphonate in the diet at concentrations that provided doses of 0, 4, 38, or 75 mg/kg/day, no clear dose-related adverse toxicological changes were noted in erythrocyte count, leukocyte count, differential leukocyte count, hemoglobin, packed cell volume, clotting time, blood glucose, or albumin (Hart 1980). Based on the NOAEL



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of 75 mg/kg/day for hematological effects in dogs, an intermediate-duration oral MRL of 0.8 mg/kg/day was derived as described in the footnote in Table 2-1 a

In a study of calves given a single dose of diisopropyl methylphosphonate at 62.5, 125, 250, 500, or 1,000 mg/kg via gelatin capsules placed with a balling gun, no hematological effects were observed at any dose level (Palmer et al. 1979).

Juvenile pastel mink that ingested 201 or 1,852 mg/kg/day diisopropyl methylphosphonate in food for 21 days showed a significantly depressed hematocrit (Aulerich et al. 1979). A significantly lower percentage of lymphocytes in peripheral blood was also noted in mink after ingestion of 2,201, or 1,852 mg/kg/day in the diet for 21 days. Mink that received food containing the compound at 17 mg/kg/day did not demonstrate a change in lymphocytes. The decreased hematocrit probably resulted from RBC damage and subsequent RBC clearance. This would explain the increase in reticulocytes and Heinz bodies noted in other studies. Increased hematocrit was noted in male dark variety mink treated with 37 or 95 mg/kg/day diisopropyl methylphosphonate in feed for 49 weeks, but not in males treated with 11 mg/kg/day or in females in any of the treatment groups. No differences in hemoglobin concentration or mean corpuscular hemoglobin were noted in either sex in any of the treatment groups (Aulerich et al. 1979).

Compared to controls, male dark brown Ranch Wild mink that were fed standard ranch diet containing diisopropyl methylphosphonate at doses of 1,009 mg/kg/day for 90 days had a significantly lower hematocrit at week 13, and lower hemoglobins at weeks 3, 7, and 13 (Bucci et al. 1994). Females that consumed 908 or 1,264 mg/kg/day had significantly lower hematocrit than the controls starting at week 3. There were no significant changes in mean erythrocyte cell volume, mean cell hemoglobin, or mean cell hemoglobin concentration. The mean number of reticulocytes in females treated at 1,264 mg/kg/day and in males at 747 mg/kg/day was significantly increased compared to the controls. The females that received 908 or 1,264 mg/kg/day had a significant increase in platelets (averaged across all time points) compared to the controls. Inconsistent changes in erythrocyte morphology were noted in the 3 highest dose groups (345, 747, 1,009 mg/kg/day for males; 455, 908, 1,264 mg/kg/day for females). All erythrocyte anomalies were normal by week 3 after treatment. Both males and females in the highest dose groups demonstrated a significant increase in Heinz bodies from week 3 through week 13, and at week 13 the females receiving 908 mg/kg/day showed a significant increase in Heinz bodies. Although not statistically significant, the number of Heinz bodies in males at 345 mg/kg/day and in females at 455 mg/kg/day was increased relative to the controls. Heinz bodies are inclusions in the RBCs resulting from the irreversible denaturation and precipitation of

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hemoglobin. An increase in Heinz bodies causes the cells to be less deformable, have a reduced lifespan, and be vulnerable to splenic phagocytes and thus induces a predisposition for the RBCs to become trapped in the splenic sinuses and destroyed by phagocytes. The Heinz body data suggest that diisopropyl methylphosphonate may cause oxidative damage to the tertiary structure of hemoglobin, thus reducing the lifespan of RBCs, which in turn ultimately induces hematopoiesis. A marginal increase in the incidence and severity of splenic extramedullary hematopoiesis was observed in male mink exposed to 1,009 mg/kg/day of diisopropyl methylphosphonate in the diet for 90 days (Bucci et al. 1994). The RBC data and the reticulocyte data are consistent with a hematopoietic response to increased RBC destruction. The study authors indicated that the observed hematological changes suggested that diisopropyl methylphosphonate or its metabolites had oxidant properties that resulted in the denaturation of hemoglobin and shortened RBC lifespan (Bucci et al. 1992).

Similar results were observed in a two-generation study of Ranch Wild brown mink. Males were fed 0, 16, 45, or 262 mg/kg/day diisopropyl methylphosphonate for 8 months, and females were fed 0, 20, 57, or 330 mg/kg/day diisopropyl methylphosphonate for 13 months (Bucci et al. 1997). In the F<sub>0</sub> generation, no changes in hematological parameters were observed in males. However, in F<sub>0</sub>-generation females fed 330 mg/kg/day, RBC counts were significantly decreased, and reticulocyte counts, Heinz body counts, and mean cell volume were significantly increased. In the F<sub>1</sub>-generation females fed 330 mg/kg/day diisopropyl methylphosphonate, Heinz body counts were increased at 6 and 13 months. Males fed this dose also had increased Heinz body counts. High-dose F<sub>1</sub> male kits had significantly decreased RBC counts at 6 weeks of age, but this effect was not observed at the age of 4.1 weeks. The increased Heinz body counts observed in this study correlated with a decrease in RBC survival, which was shown by the decreased RBC count. These effects also correlate with the histopathological findings in the spleen of these animals, in which evidence of RBC replacement was observed. Based on the NOAEL of 57 mg/kg/day for hematological effects in mink in this study, a chronic-duration oral MRL of 0.5 mg/kg/day was derived as described in the footnote in Table 2-1.

Mallard ducks fed up to 10,000 ppm diisopropyl methylphosphonate in feed for up to 24 weeks showed no significant effects on blood hemoglobin levels, leukocyte count, and hematocrit (Aulerich et al. 1979). Doses could not be calculated in mg/kg/day because food intake and body weights were not measured in this study.

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**Hepatic Effects.** No studies were located regarding hepatic effects in humans after oral exposure to diisopropyl methylphosphonate.

There is no evidence that diisopropyl methylphosphonate causes toxic effects in the liver or biliary systems. No changes of toxicologic significance were noted in serum glutamic-oxaloacetic transaminase (SGOT), serum glutamic-pyruvic transaminase (SGPT), alkaline phosphatase, liver weight, or liver histopathology in rats treated with diisopropyl methylphosphonate in the diet at doses of 0, 30, 100, or 300 mg/kg/day for 90 days (Hart 1976). No hepatic histopathological abnormalities or significant changes in liver weight were found in mice that received doses of 0, 27, 91, or 273 mg/kg/day diisopropyl methylphosphonate in food for 90 days (Hart 1976). No abnormal hepatic lesions or changes in SGPT, SGOT, or alkaline phosphatase were found in beagles treated with diisopropyl methylphosphonate in the diet at doses of 0, 4, 13, or 38 mg/kg/day for 14 days (Hart 1976). Rats pretreated with diisopropyl methylphosphonate in the diet (450 mg/kg/day for 4 days) showed a marked decrease in sleep time subsequent to the administration of hexobarbital, which is consistent with the induction of the P-450 mixed-function oxidase system. Liver weights, however, were not increased (Hart 1976). No changes in SGOT, SGPT, alkaline phosphatase, liver histopathology, or liver weight were reported in male or female beagles that received 0, 4, 38, or 75 mg/kg/day diisopropyl methylphosphonate in food for 90 days (Hart 1980).

Juvenile male pastel mink that ingested 1,852 mg/kg/day diisopropyl methylphosphonate for 21 days showed a significant decrease in liver weight; however, no associated lesions were noted (Aulerich et al. 1979). The study authors indicated that the decreases in organ weights noted in this high-dose group may be related to significantly reduced food consumption and were not necessarily a toxic response. Since a pair-fed control was not maintained for this study, the causes of the organ weight differences were not easily identifiable. Finally, the authors considered the compound nontoxic in this 21-day study (Aulerich et al. 1979). No gross or histopathological hepatic lesions or decreases in liver weight were noted in male and female dark variety mink that received diisopropyl methylphosphonate in feed at doses of 0, 11, 37, or 95 mg/kg/day for 49 weeks (Aulerich et al. 1979). No lesions were noted in the livers of Ranch Wild mink in a 90-day dietary study in which males received doses of 0, 7, 63, 345, 747, or 1,009 mg/kg/day and females received doses of 0, 9, 82, 455, 908, or 1,264 mg/kg/day (Bucci et al. 1992, 1994). The absence of hepatic lesions and the healthy appearance of the animals throughout the 90-day study are in agreement with the results reported by Aulerich et al. (1979).

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**Renal Effects.** No studies were located regarding renal effects in humans after oral exposure to diisopropyl methylphosphonate.

In a study of calves given a single dose of diisopropyl methylphosphonate at 62.5, 125, 250, 500, or 1,000 mg/kg via gelatin capsules placed with a balling gun, animals at the 500-mg/kg and higher dose levels displayed mild congestion of the renal cortex upon necropsy (Palmer et al. 1979). All calves at 1,000 mg/kg died.

Diisopropyl methylphosphonate does not appear to cause renal effects in laboratory animals. In a study of mink that were fed 1,852 mg/kg/day for 21 days, significant kidney weight loss but no renal pathology occurred (Aulerich et al. 1979). The study authors indicated that the decreases in organ weights noted in this high-dose group may be related to significantly reduced food consumption and were not necessarily a function of toxicity. In a 90-day study, small but significant increases in blood urea nitrogen were observed in Ranch Wild mink fed diisopropyl methylphosphonate in the diet at 747 and 1,009 mg/kg/day for males and 908 and 1,264 mg/kg/day for females, with no increase at 345 mg/kg/day in males and 455 mg/kg/day in females (Bucci et al. 1992). Microscopic examination of the kidneys of the mink did not reveal any treatment-related lesions (Bucci et al. 1992, 1994).

No deviations from normal were noted in the urinalyses (color, specific gravity, pH, sugar albumin, ketones, and microscopic examination of sediment), weights, or histopathology of kidneys in rats given diisopropyl methylphosphonate in the diet at doses of 0, 30, 100, or 300 mg/kg/day for 90 days (Hart 1976). No abnormal renal lesions (gross or histopathological) or changes in kidney weight were noted in mice fed chow with 0, 27, 91, or 273 mg/kg/day diisopropyl methylphosphonate for 90 days (Hart 1976). In beagle dogs given 0, 4, 13, or 38 mg/kg/day diisopropyl methylphosphonate in the diet for 14 days, urinalyses (pH, specific gravity, glucose, ketones, and total protein) resulted in values within normal limits, and no gross or histopathological renal lesions were noted (Hart 1976). No toxicologically significant anomalies in kidney weight, kidney-body weight ratios, or pathology of the kidneys were noted in dogs that received dietary diisopropyl methylphosphonate for 90 days at doses up to 75 mg/kg/day (Hart 1980). No gross diisopropyl methylphosphonate-related renal lesions were noted in rats that had received the compound in drinking water ( $6.6 \times 10^{-7}$ ,  $6.6 \times 10^{-5}$ , 0.011, 1.1 mg/kg/day) for 26 weeks (Army 1978). However, as discussed in Section 2.2.2.1, there is some confusion concerning the concentration units and purity of the diisopropyl methylphosphonate used in the Army (1978) study (EPA 1989), and therefore the results are considered inappropriate for human health risk assessment.

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**Ocular Effects.** No studies were located regarding ocular effects in humans after oral exposure to diisopropyl methylphosphonate.

An ophthalmic examination was performed during week 12 of a 13-week study of rats that received diisopropyl methylphosphonate in food (0, 30, 100, or 300 mg/kg/day); 12 of 64 animals displayed some opacity of the lens (Hart 1976). Although the finding was considered by the pathologist to be normal for the rat, the study author suggested that lens opacity may deserve attention in longer studies. No changes were noted in the eyes of beagle dogs that received diisopropyl methylphosphonate in their food (0, 4, 38, or 75 mg/kg/day) for 90 days (Hart 1980). Bucci (1997) reported no clinical observations related to treatment in mink exposed to up to 262 mg/kg/day (males) or 330 mg/kg/day (females) diisopropyl methylphosphonate in feed. The eyes were specifically examined at necropsy.

**Body Weight Effects.** No studies were located regarding body weight effects in humans after oral exposure to diisopropyl methylphosphonate.

Decreased body weight was observed in several animal studies; however, the decreases appeared related to the unpalatability of the food and to decreased food consumption rather than to a direct toxic effect. Rats that received diisopropyl methylphosphonate (0, 30, 100, or 300 mg/kg/day) in their diet for 90 days showed occasional significant changes in weight; however, the changes were inconsistent and judged not to be of toxicological significance (Hart 1976). Although individual weights of mice were not recorded, the study author indicated that it seemed “clear that growth was alike in all groups” of mice that had ingested diisopropyl methylphosphonate in the diet (0, 27, 91, or 273 mg/kg/day) for 90 days (Hart 1976). Calves given a single dose of 500 mg/kg diisopropyl methylphosphonate by gelatin capsule showed no significant body weight effects during the 14-day observation period (Palmer et al. 1979). Body weights of beagles fed diisopropyl methylphosphonate (0, 4, 13, or 38 mg/kg/day) for 14 days fluctuated within the confines of what was considered to be normal for adult dogs (Hart 1976). Rats receiving diisopropyl methylphosphonate in drinking water (0, 0.009, or 0.9 mg/kg/day) showed no apparent changes in weight after 10 weeks (Hardisty et al. 1977). However, as discussed in Section 2.2.2.1, there is some confusion regarding the actual doses used in this study. Therefore, results from Hardisty et al. (1977) are considered inappropriate for human health risk assessment. No changes in growth rate as measured by weight were noted in rats that received diisopropyl methylphosphonate in drinking water for 26 weeks (Army 1978), but as previously noted in Section 2.2.2.1, there is some confusion concerning the concentration units and purity of the diisopropyl

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methylphosphonate used in the Army (1978) study (EPA 1989). Therefore, results from this study are considered inappropriate for human health risk assessment. Pregnant female rats that were administered diisopropyl methylphosphonate in the diet (0, 10, 30, 100, or 300 mg/kg/day) during days 6-15 of gestation showed no difference in mean body weight compared to controls (Hart 1980). No differences were noted in the body weight of rats in the first generation of parents in a three-generation study, after the rats received diisopropyl methylphosphonate in the diet (0, 30, or 300 mg/kg/day) for a reproductive cycle and the 12 weeks preceding the cycle. However, second-generation parents that received 300 mg/kg/day diisopropyl methylphosphonate had a significant decrease in body weight lasting from week 4 to week 9 (Hart 1980). No significant differences were reported among beagle dogs that were fed chow containing diisopropyl methylphosphonate (0,4,38, or 75 mg/kg/day) for 90 days (Hart 1980).

Significant weight loss was noted in male and female pastel mink that received diisopropyl methylphosphonate in their feed at a dose of 1,851 mg/kg/day for 21 days (Aulerich et al. 1979). A significant reduction in food consumption was also noted in this high-dose treatment group. Subsequent to treatment (during the recovery period), the group displayed increased food consumption suggesting that the weight loss may have been due to decreased food consumption resulting from a palatability problem and was not a function of toxicity. To determine if palatability of the feed was the cause of the weight loss, an attempt was made to "pair feed" a control group to match voluntary food consumption (Bucci et al. 1994). Similar to the findings of Aulerich and coworkers (1979), male and female mink receiving feed containing diisopropyl methylphosphonate at doses of 1,009 mg/kg/day for males and 1,264 mg/kg/day for females for 21 days consumed significantly less food than did the controls and demonstrated decreased body weight throughout the study. Although only partially successful, the pair-feed data for the high-dose male group (1,009 mg/kg/day) provided substantial evidence that the weight loss was the result of decreased food consumption rather than diisopropyl methylphosphonate toxicity (Bucci et al. 1994). No differences in body weight or in the percentage change in body weight were found in mink receiving diisopropyl methylphosphonate in feed at doses of 0, 11, 37, or 95 mg/kg/day for 49 weeks (Aulerich et al. 1979). Similarly, no significant changes in food consumption or mean body weight were observed in any generation of mink fed 0, 16, 45, or 262 mg/kg/day (males) or 0, 20, 57, or 330 mg/kg/day (females) diisopropyl methylphosphonate in the diet compared to controls for up to 8 or 13 months, respectively, in a two-generation reproductive study (Bucci et al. 1997). However, it should be noted that F<sub>0</sub> females consumed almost 50% more feed than F<sub>1</sub> females in this study. The study authors suggested that there may have been food wastage in the F<sub>0</sub> generation or that because the F<sub>0</sub> generation was 8 months old when they arrived in the laboratory from the ranch they may

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have retained more of their ranch leanness, whereas because the F<sub>1</sub> animals were housed in the laboratory throughout their lives they may have become heavier on less food (Bucci et al. 1997).

Aulerich et al. (1979) reported a 14.8% weight loss in Mallard ducks given a single gavage dose of 1,800 mg/kg of diisopropyl methylphosphonate. Twelve-day-old Mallard ducklings dosed with 1,796 or 2,062 mg/kg/day of diisopropyl methylphosphonate in their feed for 5 days, followed by a 3-day normal feed period, showed significant decrease in body weight gain and food consumption (Aulerich et al. 1979). However, the study authors speculated that this decrease was due to the animals refusing to eat the feed that contained very high percentages of the chemical instead of from a loss of appetite from toxic effects. Following the dosing period, animals consumed an amount inversely proportional to the decreases in food consumption observed during the dosing period, indicating a lack of residual effects on appetite. Female Mallard ducks that received diisopropyl methylphosphonate (beginning 10 weeks prior to egg production) in their feed at a concentration of 1,000 ppm or more lost significantly less weight than the controls. Food intake and body weights were not measured in this study, and therefore, mg/kg/day doses could not be calculated.

### 2.2.2.3 Immunological and Lymphoreticular Effects

No studies were located regarding immunological or lymphoreticular effects in humans after oral exposure to diisopropyl methylphosphonate.

Histological changes in the spleen related to diisopropyl methylphosphonate intake were not observed in male or female rats exposed to 1 mg/kg/day of diisopropyl methylphosphonate in their drinking water for 26 weeks (Army 1978). As discussed in Section 2.2.2.1, there is some confusion concerning the concentration units and purity of the diisopropyl methylphosphonate used in the Army (1978) study (EPA 1989), and therefore results from the Army (1978) study are considered inappropriate for human health risk assessment. No changes in spleen weight were noted in male or female mink exposed to 1,851 mg/kg/day of diisopropyl methylphosphonate in the diet for 21 days (Aulerich et al. 1979). Similarly, spleen weight measurements and gross and histopathological examinations did not reveal any significant immunological or lymphoreticular effects in male or female mink exposed to 95 mg/kg/day of diisopropyl methylphosphonate in the diet for 12 months (Aulerich et al. 1979). A marginal increase in the incidence and severity of splenic extramedullary hematopoiesis was observed in male mink exposed to 1,009 mg/kg/day of diisopropyl methylphosphonate in the diet for 90 days (Bucci et al. 1994). No adverse effects were observed in mink exposed to 747

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mg/kg/day. Similarly, female mink fed 330 mg/kg/day, but not 57 mg/kg/day, diisopropyl methylphosphonate in the diet for 4 months exhibited significant increases in absolute and relative spleen weight and the ratio of spleen-to-brain weight (Bucci et al. 1997). This was accompanied by splenic hematopoietic cell proliferation. The splenic extramedullary hematopoiesis in both studies is probably due to erythrocyte damage and shortened survival of erythrocytes rather than a direct lymphoreticular effect.

#### 2.2.2.4 Neurological Effects

No studies were located regarding neurological effects in humans after oral exposure to diisopropyl methylphosphonate.

Single-dose oral toxicity studies in both rats and mice indicated that signs of diisopropyl methylphosphonate intoxication included decreased activity, occasional ataxia, and prostration within 1-4 hours after dosing (Hart 1976). Acute toxicity studies on Mallard ducks (Aulerich et al. 1979) treated orally with doses of up to 1,800 mg/kg diisopropyl methylphosphonate demonstrated that the most notable clinical sign was salivation. Acute studies in female minks at doses up to 600 mg/kg (Aulerich et al. 1979) indicated clinical signs included salivation, lethargy, myasthenia, immobilization, vomiting, and death. Male B6C3F<sub>1</sub> mice treated with 1,000 mg/kg diisopropyl methylphosphonate for 3 days were lethargic or comatose after each treatment, but recovered within an hour (DOD 1991a). Calves that received a single dose of 1,000 mg/kg diisopropyl methylphosphonate in a gelatin capsule suffered neurological effects including depression, ataxia, tympanitis, engorgement of the meningeal vessels, excess fluid in cerebral ventricles, and prostration followed by death within 2 hours after treatment (Palmer et al. 1979). Mink that received 1,852 mg/kg/day diisopropyl methylphosphonate in the feed for 21 days displayed aggressive behavior (Aulerich et al. 1979). However, the study authors concluded that this behavior was probably due to hunger resulting from the unpalatability of the feed.

Inconsistent fluctuations were observed in RBC or plasma cholinesterase activity in rats that received diisopropyl methylphosphonate in the diet (0, 30, 100, or 300 mg/kg/day) for 90 days (Hart 1976). The most significant difference was a decrease in plasma cholinesterase activity in the high-dose female group at 13 weeks. The study author pointed out, however, that the control group value was exceptionally high compared to that of other time intervals (Hart 1976). In a 90-day study of dogs using diisopropyl methylphosphonate in the diet (0, 4, 38, or 75 mg/kg/day), a slight increase in plasma cholinesterase was observed; however, problems with the experimental protocol (failure to measure cholinesterase at 4 and 8



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weeks) make any interpretations difficult (Hart 1980). Dark brown Ranch Wild mink that ingested 375, 742, or 1,009 mg/kg/day (males) or 455, 908, or 1,264 mg/kg/day (females) demonstrated decreased plasma cholinesterase compared to the controls, although the change was rapidly reversible. No effect was noted in erythrocyte cholinesterase activity. The study authors indicated that plasma cholinesterase is labile and sufficiently unrelated to the nervous system and that, therefore, the observed changes in the low-dose groups were toxicologically unimportant. Further, they noted that, even in the high-dose groups with large decreases (61%) in plasma cholinesterase, there was no reduction in erythrocyte cholinesterase or signs of acetylcholinesterase inhibition (Bucci et al. 1992, 1994). Similar results were also observed in a two-generation reproductive study using brown Ranch Wild mink that ingested 0, 16, 45, or 262 mg/kg/day (males) or 0, 20, 57, or 330 mg/kg/day (females) (Bucci et al. 1997). No changes in brain acetylcholinesterase were observed in either the F<sub>1</sub> or F<sub>2</sub> offspring. Decreases (42%) in plasma cholinesterase were observed in F<sub>0</sub> females at 4 months, and F<sub>1</sub> females at 4 (22%), 8 (21%), and 13 (31%) months of age at high doses. These effects were not observed in males or F<sub>2</sub> kits. No significant changes or biological significance (>20%) were observed in RBC or whole blood cholinesterase in any group or generation. There were no clinical signs of acetylcholinesterase inhibition observed in any generation or treatment group in this study. Based on the studies discussed, it appears that diisopropyl methylphosphonate may inhibit plasma cholinesterase but not RBC cholinesterase.

Consistent decreases in plasma cholinesterase may not have been observed in rats and dogs because they were treated with lower doses of diisopropyl methylphosphonate. In general, depression of plasma cholinesterase, also known as pseudocholinesterase or butyrylcholinesterase, is considered a marker of exposure rather than an adverse effect. Depression of cholinesterase activity in red blood cells (acetylcholinesterase) is a neurological effect thought to parallel the inhibition of brain acetylcholinesterase activity. It is considered an adverse effect. Acetylcholinesterase is found mainly in nervous tissue and erythrocytes. Diisopropyl methylphosphonate was not found to inhibit RBC cholinesterase at doses at which plasma cholinesterase was significantly inhibited. Thus, although the 330-mg/kg/day dose in females appears to be a lowest-observable-effect level (LOAEL) for plasma cholinesterase inhibition and 57 mg/kg/day females or 262 mg/kg/day in males appears to be a no-observable-effect level (NOAEL) for this end point based on the study by Bucci et al. (1997), it is not a biologically significant change.

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**2.2.2.5 Reproductive Effects**

No studies were located regarding reproductive effects in humans after oral exposure to diisopropyl methylphosphonate.

In a single-generation reproductive study, male and female rats received diisopropyl methylphosphonate in the drinking water for 10 weeks at doses of 0, 0.009, or 0.9 mg/kg/day (Hardisty et al. 1977). Dosing continued during gestation and lactation in females. There were no differences in male fertility (calculated by the number of females that became pregnant) among the treatment groups and controls. No differences were noted in the litter sizes among those treated and the controls. No differences were noted in the number of stillborn pups or in pup weights. The study authors concluded that there was no evidence of adverse diisopropyl methylphosphonate-induced reproductive effects. However, as discussed in Section 2.2.2.1, there is some confusion regarding the actual doses to which the animals were exposed in the Hardisty et al. (1977) study. Therefore, results from this study are considered inappropriate for human health risk assessment.

In a three-generation reproductive study, male and female rats received diisopropyl methylphosphonate in their feed (0, 30, or 300 mg/kg/day) for 11 weeks before being mated with animals of the same dose group in the 12th week (Hart 1980). Dosing continued during gestation and lactation. A week after lactation of the F<sub>1A</sub> pups, the F<sub>0</sub> females were remated with a different male. A week after lactation of the F<sub>1B</sub> pups, the F<sub>0</sub> parents were sacrificed and necropsied. Male and female F<sub>1B</sub> animals were selected and mated as above. Similarly F<sub>2B</sub> offspring were mated, yielding third-generation (F<sub>3A</sub> and F<sub>3B</sub>) offspring. No differences in male virility or female fertility were noted in the F<sub>0</sub> and F<sub>1</sub> parents, and no differences in newborn viability or pup weights were noted in the F<sub>1</sub> and F<sub>2</sub> offspring. A significant number of pup losses were noted in the F<sub>3A</sub> offspring from the 300-mg/kg/day group; however, since the losses were not observed in the second mating (F<sub>3B</sub> offspring), the losses were probably not related to treatment. Further, pup appearance and gross examination at necropsy did not reveal any evidence of diisopropyl methylphosphonate-related effects in the F<sub>3A</sub> or F<sub>3B</sub> pups, although the histopathological changes were apparently not evaluated. No significant differences in body weight and food consumption among the F<sub>0</sub> (parent), F<sub>1B</sub> and F<sub>2B</sub> generations were observed. Necropsy observations did not indicate any dose-dependent relationships of diisopropyl methylphosphonate in the feed at doses of 30 or 300 mg/kg/day in the rat in 3 successive generations with 2 matings per generation (Hart 1980).

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Reproductive toxicity in mink was assayed in a 49-week feed study (Aulerich et al. 1979). Male and female dark variety mink received feed containing diisopropyl methylphosphonate at doses of 0, 11, 37, or 95 mg/kg/day. Male fertility, estimated by the presence of sperm in post-coital vaginal aspirations, was not adversely affected. Further, no significant differences were noted in whelping dam and kit performance, kit mortality, kit weight, or the body weight of lactating females at 4 weeks post-par-turn (Aulerich et al. 1979). h-r the study, an increase in deaths occurred in females that was statistically significant at the high dose. No control females died, while 2 of 23, 3 of 24, and 5 of 24 died at the low, middle, and high doses, respectively. However, the deaths may not be treatment related. In a concurrent study conducted to assess the toxicity of dicyclopentadiene which used mink from the same lot, the mortality in the untreated female mink was 4 of 24, with 2 mink dying between the time of mating and lactation. The conclusion that female deaths in the Aulerich et al. (1979) study were probably not DIMP treatment-related was supported by a two-generation reproductive study performed using Ranch Wild mink fed 0, 16, 45, or 262 mg/kg/day (males) or 0, 20, 57, or 330 mg/kg/day (females) diisopropyl methylphosphonate in the diet (Bucci et al. 1997). The F<sub>0</sub> males and females were exposed for 1 and 4 months, respectively, and the F<sub>1</sub> males and females for 8 and 13 months, respectively. No treatment-related effects were observed in kits/litter, live kits/litter, litter weights at birth or at 28 days, or kit development. No dose-related deaths was observed in females in either the F<sub>0</sub> and F<sub>1</sub> generations. Ovarian follicles were counted in control and high-dose F<sub>1</sub> females to examine possible ovarian toxicity. There was a significant ( $p < 0.01$ ) increase in the mean follicle count of high-dose females ( $645 \pm 157$ ) compared to controls ( $329 \pm 153$  or  $460 \pm 148$ ). Only the control and high-dose animals' ovaries were examined. However, it is not clear whether this end point represents an adverse effect because the treated dams of both the F<sub>0</sub> and F<sub>1</sub> generations produced as many offspring as controls. The study authors noted that the effect could be representative of disrupted follicle maturation with retention of ova. Semen quality in F<sub>0</sub> and F<sub>1</sub> males, as measured by sperm motility, epididymal sperm count, and incidence of head/tail abnormalities, was unaffected by treatment.

Another study was performed in which both male and female Mallard ducks received feed containing 1,000, 3,200, or 10,000 ppm diisopropyl methylphosphonate from 40 weeks prior to the beginning of egg production (Aulerich et al. 1979). The dosing continued through the egg production period and for an additional 10 weeks after egg production levels reached 50%. No significant changes in maternal death or in gonad weight in either males or females were reported at any dose level. Food intake and body weights were not measured in this study, and therefore mg/kg/day doses could not be calculated.

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**2.2.2.6 Developmental Effects**

No studies were located regarding developmental effects in humans after oral exposure to diisopropyl methylphosphonate.

Diisopropyl methylphosphonate was administered to pregnant female rats in the diet at doses of 0, 10, 30, 100, or 300 mg/kg/day on day 6 through day 15 of gestation (Hart 1980). No deaths occurred among the rats. No differences were noted in mean body weight or food consumption. Observations of the uterine contents indicated no compound-related effects.” Gross examination revealed no abnormalities of the internal organs. There was a slight increase in the abnormal-normal fetus ratio in the 300-mg/kg/day group; however, the effect was not statistically significant. Further, the changes noted did not suggest a specific area of involvement. The study author concluded that the administration of diisopropyl methylphosphonate in the diet at these concentrations produced no effect on dams and no compound-induced terata, embryo toxicity, or inhibition of fetal growth and development. In a single-generation study, male and female rats received diisopropyl methylphosphonate in the drinking water (0, 0.009, 0.9 mg/kg/day) for 10 weeks (Hardisty et al. 1977). Dosing continued through gestation and lactation in females. No skeletal or visceral anomalies were found among the rats that were examined. However, as discussed in Section 2.2.2.1, there is some confusion regarding the actual dosages that the animals were exposed to in this study. Therefore, results from the Hardisty et al. (1977) study are considered inappropriate for human health risk assessment.

Decreased red blood cell counts were observed in F<sub>2</sub> male, but not female, kits at 6 weeks of age in a two-generation reproductive study in which Ranch Wild mink were fed 0, 16,45, or 262 mg/kg/day (males) or 0, 20, 57, or 330 mg/kg/day (females) diisopropyl methylphosphonate (Bucci et al. 1997). This effect was not observed in F<sub>1</sub> kits at 11 weeks.

Both male and female Mallard ducks received feed containing 1,000, 3,200, or 10,000 ppm diisopropyl methylphosphonate beginning 10 weeks prior to the beginning of egg production (Aulerich et al. 1979). The dosing continued through the egg production period and for an additional 10 weeks after egg production levels reached 50%. No significant difference was reported between dosed animals and controls in number of cracked shells, shell thickness, egg incubation parameters (viability, fertile versus nonfertile eggs, survival in shell), incidence of developmental abnormalities, or 14-day survival in chicks. Food intake and body weights were not measured in this study, and therefore mg/kg/day doses could not be calculated.

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**2.2.2.7 Genotoxic Effects**

No studies were located regarding genotoxic effects in humans after oral exposure to diisopropyl methylphosphonate.

Genotoxicity studies in animals are discussed in Section 2.5.

**2.2.2.8 Cancer**

No studies were located regarding carcinogenic effects in humans or animals after oral exposure to diisopropyl methylphosphonate.

**2.2.3 Dermal Exposure**

Limited data are available concerning health effects in humans and animals following dermal exposure to diisopropyl methylphosphonate. Results of these studies are discussed below and presented in Table 2-2.

**2.2.3.1 Death**

No studies were located regarding death in humans after dermal exposure to diisopropyl methylphosphonate.

Twelve New Zealand White rabbits were divided into three dose groups, each consisting of four animals (Hart 1976). Diisopropyl methylphosphonate was applied neat, in single doses (200, 630, or 2,000 mg/kg) to areas of the back that had been closely clipped. Three rabbits that received 2,000 mg/kg were found dead the morning after application, and a fourth animal that had received 630 mg/kg was found dead on the third morning. Based on these results, a dermal LD<sub>50</sub> of 1,100 mg/kg was calculated. The study author noted that there was “no antemortem indication of systemic intoxication based on the general appearance and behavior” of the animals. In fact, the body weights of the surviving animals increased during the 2-week observation period. Further, no edema or eschar formation of the skin was observed, and the hair growth appeared normal in the shaved areas. No treatment-related abnormalities were noted during necropsy (Hart 1976).

TABLE 2-2. Levels of Significant Exposure to Diisopropyl Methylphosphonate - Dermal

Species (strain)	Exposure duration/ frequency/ (specific route)	System	NOAEL (mg/kg/day)	LOAEL (effect)		Reference
				Less serious (mg/kg/day)	Serious (mg/kg/day)	
<b>ACUTE EXPOSURE</b>						
<b>Death</b>						
Rabbit (New Zealand)	once				1100 M (LD <sub>50</sub> )	Hart 1976
<b>Systemic</b>						
Rabbit (New Zealand)	once	Ocular		0.1	(diffuse opacity of the corneal surface)	Hart 1976
Rabbit (New Zealand)	once	Derm	2000			Hart 1976

Derm = dermal; LD<sub>50</sub> = lethal dose, 50% kill; LOAEL = lowest-observed-adverse-effect level; M = male; NOAEL = no-observed-adverse-effect level

## 2. HEALTH EFFECTS

**2.2.3.2 Systemic Effects**

No studies were located regarding respiratory, cardiovascular, gastrointestinal, hematological, musculoskeletal, hepatic, or renal effects in humans or animals after dermal exposure to diisopropyl methylphosphonate.

**Dermal Effects.** Skin irritation was noted in wildlife officers at the RMA after they handled sick or dead ducks without gloves (NIOSH 1981). Although the investigators concluded that diisopropyl methylphosphonate contributed to the local effects, a number of other compounds were present. Analysis of the pond water indicated the presence of a number of organic and inorganic contaminants, including diisopropyl methylphosphonate (11.3 ppm); aldrin (0.368 ppm); dieldrin (0.0744 ppm); dicyclopentadiene, bicycloheptadiene, diethyl benzene, dimethyl disulfide, methyl acetate, methyl isobutyl ketone, toluene, and sodium (49,500 ppm); chloride (52,000 ppm); arsenic (1,470 ppm); potassium (180 ppm); fluoride (63 ppm); copper (2.4 ppm); and chromium (0.27 ppm). Because of the presence of numerous compounds, it is unclear whether diisopropyl methylphosphonate was related to the irritation.

Only minimal indications of skin irritation were noted in New Zealand White rabbits that received a single dose of diisopropyl methylphosphonate (200, 630, or 2,000 mg/kg) on intact or abraded skin (Hart 1976). In the same series of studies, the sensitization potential for diisopropyl methylphosphonate was evaluated in guinea pigs. Eight albino guinea pigs received 10 intracutaneous injections (3 times per week to a total of 10) of diisopropyl methylphosphonate in corn oil on one side and of the corn oil vehicle on the other side. Four positive control guinea pigs received intracutaneous injections of a known sensitizer (2,4-dinitro-1-chlorobenzene). Following a 2-week period, the animals received one or more challenges, applied in the same manner as the initial material, and were graded for sensitization. Diisopropyl methylphosphonate was not judged by the study author to be a strong sensitizer in guinea pigs; however, this was considered a preliminary study (Hart 1976).

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**Ocular Effects.** No studies were located regarding ocular effects in humans after dermal exposure to diisopropyl methylphosphonate.

The potential of diisopropyl methylphosphonate to cause eye irritation was evaluated with the Draize Test (Hart 1976). The compound was directly applied to the conjunctival sac of one eye in each of nine New Zealand White rabbits. Significant irritation of the conjunctivae was observed in all rabbits, and the corneal surface was characterized by a diffuse opacity. The opacity was temporary and cleared within 8 days. Irrigation with lukewarm water following application of diisopropyl methylphosphonate reduced but did not prevent irritation (Hart 1976).

No studies were located regarding the following health effects in humans or animals after dermal exposure to diisopropyl methylphosphonate.

**2.2.3.3 Immunological and Lymphoreticular Effects****2.2.3.4 Neurological Effects****2.2.3.5 Reproductive Effects****2.2.3.6 Developmental Effects****2.2.3.7 Genotoxic Effects**

Genotoxicity studies are discussed in Section 2.5.

**2.2.3.8 Cancer**

No studies were located regarding cancer in humans or animals after dermal exposure to diisopropyl methylphosphonate.

**2.3 TOXICOKINETICS**

Diisopropyl methylphosphonate is rapidly absorbed following oral administration in animals and is widely distributed throughout the body. Two metabolites of diisopropyl methylphosphonate are isopropyl methylphosphonic acid (IMPA) and methylphosphonic acid (MPA). Excretion of these metabolites in animals occurs primarily through the urine.



### 2.3.1 Absorption

#### 2.3.1.1 Inhalation Exposure

No studies were located regarding absorption in humans or animals after inhalation exposure to diisopropyl methylphosphonate.

#### 2.3.1.2 Oral Exposure

No studies were located regarding absorption in humans after oral exposure to diisopropyl methylphosphonate.

Diisopropyl methylphosphonate was absorbed from the gastrointestinal tract of mink, rats, mice, dogs, and cattle after oral administration of the [<sup>14</sup>C]-radiolabeled compound as indicated by the appearance of the radiolabel in the blood after exposure (Bucci et al. 1992; Hart 1976; Ivie 1980). Quantitative measurements of absorption can only be approximated (Hart 1976; Ivie 1980).

Single doses (225 mg/kg) of [<sup>14</sup>C]-radiolabeled diisopropyl methylphosphonate dissolved in polyethylene glycol were administered by gavage to fasted male mice, rats, and dogs (Hart 1976). Plasma concentrations of the labeled diisopropyl methylphosphonate were monitored after 5, 15, and 30 minutes and after 1, 2, 4, 6, and 24 hours in the mice, rats, and dogs as well as at 48 and 72 hours in the mice and rats. The plasma data indicated that there were species differences in absorption. In mice, the highest level of radioactivity (172 µg/mL) was noted in the blood at 15 minutes. In rats, plasma concentrations peaked at 151 µg/mL about 2 hours after compound administration, suggesting slower movement through the gastrointestinal tract and uptake by the mucosa. In dogs, peak plasma concentrations also occurred at 2 hours post-administration at a concentration of 276 µg/mL. This value suggests that movement through the gastrointestinal tract in dogs was comparable to that in rats. However, for each measurement taken in the first 2 hours, the plasma levels of diisopropyl methylphosphonate in dogs were higher than those in rats. This suggests that the total amount of intestinal uptake in the dogs exceeded that for the rats.

Studies in mink (Bucci et al. 1992) and rats (Weiss et al. 1994) indicate that absorption following oral exposure to a single dose of [<sup>14</sup>C]-radiolabeled diisopropyl methylphosphonate is rapid. Male and female rats

## 2. HEALTH EFFECTS

were treated with a gavage dose of 66 or 660 mg/kg, and mink were treated with a gavage dose of 27 or 270 mg/kg. Blood levels of radioactivity peaked in 2-3 hours in rats and at 2 hours in mink, except in high-dose male mink, in which the peak was observed at 4 hours. In male rats, 86-97% of the radioactivity was recovered in the urine. Recovery in the urine of female rats (67-72%) and mink (70-91%) was lower, but this was thought to be a result of difficulties in urine collection during blood sampling rather than a difference in the absorption or disposition of diisopropyl methylphosphonate.

Plasma levels of diisopropyl methylphosphonate were measured in a single lactating Jersey cow after the sixth day of diisopropyl methylphosphonate oral administration (10 mg/kg/day) by gelatin capsule (Ivie 1980). For the first 5 days the cow was given unlabeled compound and fed hay *ad libitum*. The diisopropyl methylphosphonate administered on the 6th day was labeled with carbon 14. Based on measurements of label in the plasma, absorption in the cow paralleled that in rats and dogs, with the highest concentration detected in the plasma at 2 hours after administration.

After doses of 10 mg/kg/day in a cow and 225 mg/kg in male mice, rats, and dogs (fasted for 18 hours prior to administration of diisopropyl methylphosphonate), approximately 90% of the diisopropyl methylphosphonate was absorbed from the gastrointestinal tract. This estimate is based on the small percentage of the label found in the feces in the 2-3-day period after dosing and the 84-97% excreted in the urine (Hart 1976; Ivie 1980).

### 2.3.1.3 Dermal Exposure

No studies were located regarding absorption in humans after dermal exposure to diisopropyl methylphosphonate.

Some dermal uptake of diisopropyl methylphosphonate through the skin of rabbits can be inferred based on the death of 3 of 4 animals treated with 2,000 mg/kg neat applied to shaved abraded or unabraded skin for a 24-hour period (Hart 1976).

Snodgrass and Metker (1992) reported that male pigs dermally treated with [<sup>14</sup>C]-radiolabeled diisopropyl methylphosphonate dissolved in 95% ethanol (1-mL dose volume over a 100-cm<sup>2</sup> area) absorbed less than 7% of the percutaneously-applied dose regardless of dose level. The remaining dermally-applied diisopropyl methylphosphonate is assumed to have volatilized or been lost along with exfoliated skin.

### 2.3.2 Distribution

#### 2.3.2.1 Inhalation Exposure

No studies were located regarding distribution in humans or animals after inhalation exposure to diisopropyl methylphosphonate.

#### 2.3.2.2 Oral Exposure

No studies were located regarding distribution in humans after oral exposure to diisopropyl methylphosphonate.

In animals, diisopropyl methylphosphonate absorbed from the gastrointestinal tract is rapidly distributed to the tissues as indicated by the decay in peak plasma levels after absorption (Hart 1976; Ivie 1980). In mice, plasma radioactivity declined slowly from 15 minutes to 1 hour after exposure and then dropped rapidly during the next 2 hours. At the end of 24 hours, the label in the blood was 0.63 µg/mL or 0.3% of the 173-µg/mL peak concentration (Hart 1976). The radiolabel in the plasma of rats after 24 hours was nearly identical to that in mice (0.61 µg/L) and was 0.4% of the peak concentration. Clearance of label was slower in dogs with 1.3% of the 276-µg/mL peak concentration present after 24 hours.

Diisopropyl methylphosphonate is initially distributed to the liver by way of the portal circulation after absorption from the intestines, and then to the kidneys for excretion (Hart 1976). High concentrations of radiolabel were detected in the urinary bladder itself, exclusive of any urine of mice at 15 minutes and persisted for up to 6 hours. A similar pattern of distribution to the liver, kidney, and urinary bladder was seen in rats over the first 6 hours after oral administration of diisopropyl methylphosphonate.

Tissue levels of diisopropyl methylphosphonate radiolabel in dogs were determined at 4 and 24 hours on the day of compound administration (Hart 1976). When the liver, kidney, and bladder values for dogs at 4-hours are compared to those for mice and rats, it appears that clearance from the body is slower in canine species. At 24 hours, the organ concentrations of radiolabel in the dogs' tissues were generally 4-8 times higher than those in mice and rats, but tissue:blood concentration ratios were lower (Hart 1976).

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Based on a single-dose study and tissue:blood concentration ratios, diisopropyl methylphosphonate is taken up by the lungs of mice, rats, and dogs (Hart 1976). The lung:blood concentration ratios are 4.5 for mice, 3.6 for rats, and 2.0 for dogs. There is also some uptake of the [<sup>14</sup>C]-label by the testes where the testes:blood concentration ratios are 2.7 for mice, 2.3 for rats, but only 1.1 for dogs.

Fat tissues do not appear to highly concentrate diisopropyl methylphosphonate or its metabolites. Tissue:blood ratios for adipose deposits range from 1.3 to 3.6 in the species studied (Hart 1976). There was a surprisingly high concentration of radiolabel in the skin for mice with a tissue:blood ratio of 14.6 (Hart 1976). It has been suggested, however, that the skin samples were contaminated with urine. Values for rats and dogs were much lower (Hart 1976).

In a single-dose oral study in male and female rats (Bucci et al. 1992), only 0.5% of the radioactivity from a dose of 660 mg/kg [<sup>14</sup>C]-radiolabeled diisopropyl methylphosphonate was found in the tissues 120 hours after dosing. The investigators indicated that no important tissue depot for diisopropyl methylphosphonate or its metabolites could be identified from the data obtained.

The study of diisopropyl methylphosphonate distribution in a lactating Jersey cow was the only study that used multiple doses of diisopropyl methylphosphonate (Ivie 1980). In this single cow, radioactivity was detected in the blood 2 hours after dosing with [<sup>14</sup>C]-radiolabeled compound but not in the tissues. The animal had received diisopropyl methylphosphonate in one gelatin capsule for 5 days before the radiolabeled dose was administered. If tissue uptake in the cow was similar to that in dogs, measurements made 2 hours after dosing may not have provided an opportunity to measure tissue uptake of label. After 24 hours, 0.1% of the administered label was found in the cow's milk (Ivie 1980).

### 2.3.2.3 Dermal Exposure

No studies were located regarding distribution in humans or animals after dermal exposure to diisopropyl methylphosphonate.

### 2.3.3 Metabolism

Metabolism of diisopropyl methylphosphonate can be inferred based on the identification (Hart 1976; Ivie 1980) and quantification of its urinary metabolites in various animal species (Bucci et al. 1992; Hart 1976; Ivie 1980; Snodgrass and Metker 1992; Weiss et al. 1994). Hydrolysis of one of the two phosphate ester bonds liberates isopropanol and converts diisopropyl methylphosphonate to IMPA. The locations of the enzymes capable of catalyzing diisopropyl methylphosphonate phosphate ester hydrolysis have not been identified.

At low doses, the metabolism of diisopropyl methylphosphonate to IMPA in the body is rapid and nearly complete. After oral exposure to diisopropyl methylphosphonate, the principal metabolite isolated from both urine (93-99%) and feces ( $\geq 97\%$ ) in mink, mice, rats, dogs, and cattle is IMPA (Bucci et al. 1992; Hart 1976; Ivie 1980). Less than 0.5% of the radiolabel was detected in the exhaled air of rats and mice as carbon dioxide after diisopropyl methylphosphonate ingestion (Hart 1976). Thus, complete metabolism of diisopropyl methylphosphonate occurs only to a minor extent.

A sex difference in the rate of conversion of DIMP to its primary metabolite was observed after intravenous administration of  $^{14}\text{C}$ -DIMP in rats (Bucci et al. 1992). The males appeared to convert DIMP to IMPA more actively than the females. The apparent plasma elimination half-life of DIMP was about 45 minutes in males and up to 250 minutes in females. Both the rate and total excretion of the administered dose in urine were also higher in male rats. However, this sex difference was not observed for orally-administered DIMP in minks (Bucci et al. 1992; Weiss et al. 1994).

In studies in rats and mink that used more than one dose, the area under the plasma-IMPA concentration time curves indicated that at high doses the principal pathway for the conversion of diisopropyl methylphosphonate to IMPA was saturated (Bucci et al. 1992). In rats, metabolism was saturated at an oral dose of 660 mg/kg, but not at 66 mg/kg; in mink, an oral dose of 270 mg/kg caused metabolic saturation which did not occur at 27 mg/kg.

The second ester bond in diisopropyl methylphosphonate is more stable to hydrolysis than the first, and IMPA undergoes relatively little decomposition to MPA. After intraperitoneal administration of 160 mg/kg [ $^{32}\text{P}$ ]-labeled IMPA to a single male rat, the IMPA was excreted unmodified in the urine with no evidence of further metabolism (Hoskin 1956).

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Support for the limited metabolism of IMPA to MPA with resultant retention of the MPA is provided by distribution studies of Sarin (GB). Twenty-four hours after intravenous injection of [ $^3\text{H}$ ]-labeled Sarin, unextractable label was present in all tissues except the plasma and kidney. At least some unextractable label was presumed by the study authors to be protein-bound MPA (Little et al. 1986, 1988). The concentration of bound MPA was found to be 20-65% of bound IMPA in different mouse brain areas, with the highest concentration found in the hypothalamus.

### 2.3.4 Elimination and Excretion

#### 2.3.4.1 Inhalation Exposure

No studies were located regarding excretion in humans or animals after inhalation exposure to diisopropyl methylphosphonate.

#### 2.3.4.2 Oral Exposure

No studies were located regarding excretion in humans after oral exposure to diisopropyl methylphosphonate.

Urine is the principal excretory route for elimination of diisopropyl methylphosphonate after oral administration to mice, rats, pigs, mink, or dogs (Hart 1976; Snodgrass and Metker 1992; Weiss et al. 1994). However, the rate of excretion differs among species. Peak urinary excretion of a single oral dose of 225 mg/kg [ $^{14}\text{C}$ ]-radiolabeled diisopropyl methylphosphonate occurred at 6 hours in mice, 24 hours in rats, and 72 hours in dogs (Hart 1976). Over the 72-hour period after dosing, a total of 96% of the recovered label was found in the urine of mice, 86% of the recovered label was found in the urine of rats, and 97% was found in the urine of dogs. Fecal excretion was low in some species (3-30%). Only 0.06% of the label was found in the bile of dogs; biliary excretion in mice and rats was not determined. Minimal label (<0.5%) was removed from the body with exhaled air. Dose administration and recovery in the Hart (1976) study may not have been very accurate as total recovery of radioactivity was 126% in mice, 108% in rats, and 90% in dogs.

In rats given 66 or 660 mg/kg diisopropyl methylphosphonate, peak radioactivity in the blood was at 2-3 hours in both sexes at both doses; however, radioactivity was still detectable in the blood 24 hours post-administration in the 66-mg/kg group (Weiss et al. 1994). After intravenous administration of 66 mg/kg, the elimination half-life of diisopropyl methylphosphonate was estimated at 45 minutes in males and 250 minutes

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in females; the rate of excretion was greater in males than in females. Urine and feces together accounted for 86-97% of administered radioactivity in males and 57-62% in females.

Mink fasted overnight and given 27 or 270 mg/kg diisopropyl methylphosphonate by gavage exhibited peak radioactivity in the blood at 2-4 hours in both sexes (Weiss et al. 1994). In male and female mink given 270 mg/kg, urinary excretion accounted for 83.3% and 83.6%, respectively, of the total radioactivity administered. In male and female mink given 27 mg/kg, urine accounted for 86.9% and 91.5%, respectively, of the administered radioactivity. In male and female mink given 270 mg/kg, feces accounted for only 1.7% and 2.5%, respectively, of the total radioactivity administered. In male and female mink given 27 mg/kg, feces accounted for only 3.1% and 3.7%, respectively, of the total radioactivity administered. The study authors indicated that total recoveries may have been low because of difficulties in urine collection, especially while blood samples were being taken.

In a single lactating Jersey cow, 30% of a [<sup>14</sup>C]-radiolabeled dose of 10 mg/kg/day was excreted in the urine 4 hours after dosing, and 84% was excreted in a 96-hour period (Ivie 1980). The amount of label after 96 hours was 7% in the feces and less than 1% in the cow's milk. Before being given the radiolabeled material, the cow had been administered unlabelled diisopropyl methylphosphonate in a gelatin capsule for 5 consecutive days.

#### 2.3.4.3 Dermal Exposure

No studies were located regarding excretion in humans after dermal exposure to diisopropyl methylphosphonate.

Yorkshire cross pigs were treated with diisopropyl methylphosphonate percutaneously and subcutaneously (Snodgrass and Metker 1992). In animals treated subcutaneously with 40 mg labelled diisopropyl methylphosphonate, 91% of the injected dose was recovered in the urine within 24 hours, with a total of 94% recovered in urine and 6% recovered in feces by 7 days. In animals exposed percutaneously to 0.4, 4, or 40 mg diisopropyl methylphosphonate (dissolved in ethanol and applied to a 100-cm<sup>2</sup> clipped area on the back), percentages of the total radioactivity administered (including nonabsorbed diisopropyl methylphosphonate) in urine were 7%, 3%, and 4%, respectively. Radioactivity in feces was very low, ranging from 0.31% to 0.45%. As the total percentage of administered radioactivity absorbed was only 7%, 3%, and 4% for the three dose levels, almost all radioactivity absorbed was accounted for in urine or feces.

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**2.3.5 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). 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 parametrization, (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) is adequately described, however, this simplification is desirable because data are often unavailable for many biological processes. A



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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 2-2 shows a conceptualized representation of a PBPK model.

If PBPK models for diisopropyl methylphosphonate 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.

No data were located regarding PBPK/PD modeling to explain the biological basis for the dose-response relationship in humans or animals after exposure to diisopropyl methylphosphonate.

## 2.4 MECHANISMS OF ACTION

### 2.4.1 Pharmacokinetic Mechanisms

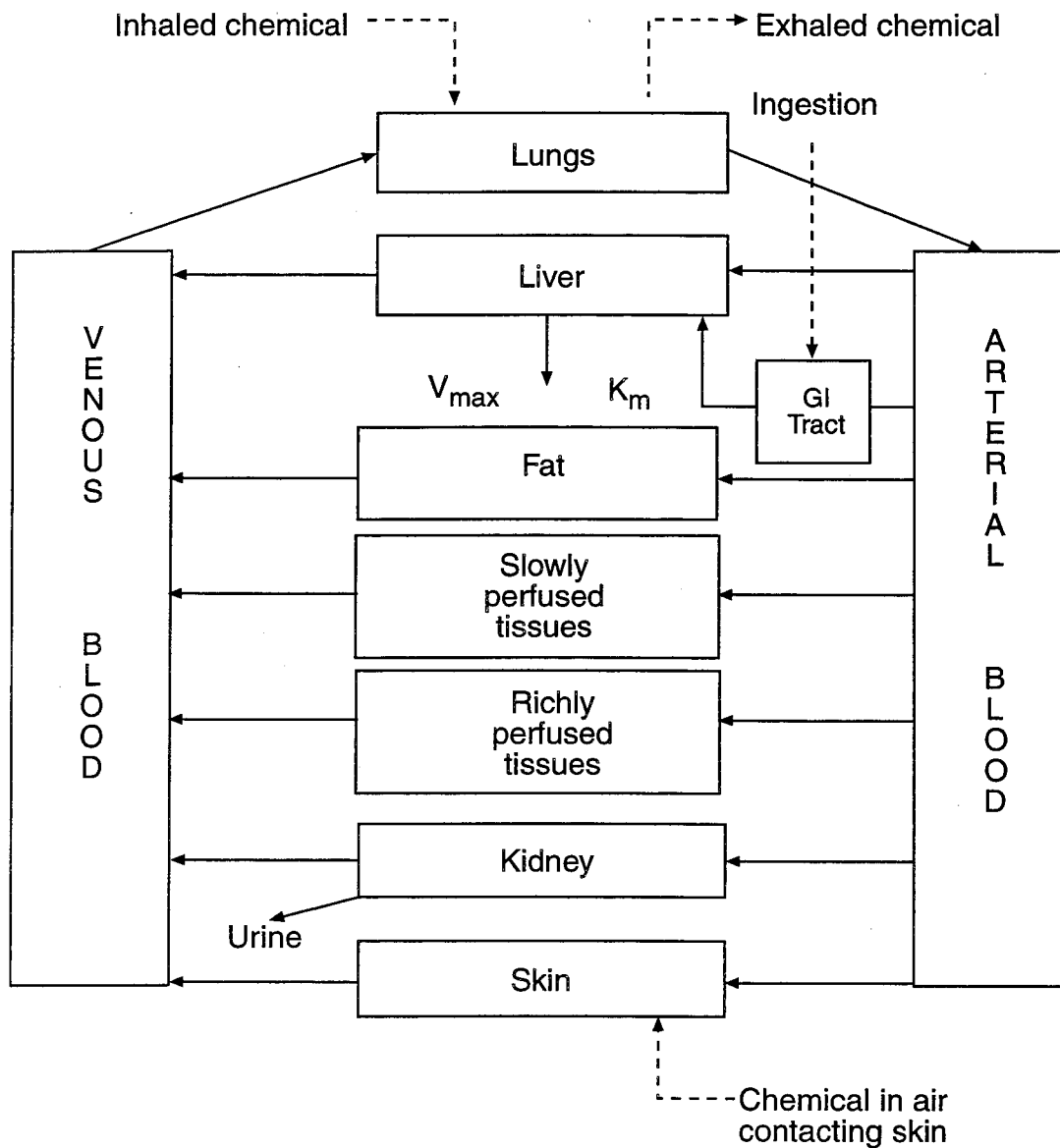
**Absorption.** No studies were located regarding the mechanism of absorption in humans or animals after inhalation, oral, or dermal exposure to diisopropyl methylphosphonate. Both facilitated transport and diffusion through the lipophilic portions of the membrane could be involved in absorption processes. No data were found regarding lipid solubility or partition coefficients.

**Distribution.** No studies were located regarding the mechanism of distribution in humans after inhalation, oral, or dermal exposure to diisopropyl methylphosphonate.

In animals, radiolabel distribution studies demonstrate rapid movement of the label to the liver. Average plasma concentrations of label were greater than average blood concentrations in mice, rats, and dogs after oral administration of a single dose of 225 mg/kg diisopropyl methylphosphonate (Hart 1976). This suggests

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**Figure 2-2. Conceptual Representation of a Physiologically Based Pharmacokinetic (PBPK) Model for a Hypothetical Chemical Substance**



Source: adapted from Krishnan et al. 1994

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.

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greater solubility of the radiolabeled compound in the plasma or protein adsorption rather than adsorption to the cellular components of the blood.

There may be some hydrolysis of IMPA to MPA in the liver or in other tissues, but the MPA that is liberated is apparently bound to other molecules and is not immediately excreted. Most of the IMPA is rapidly removed from the blood by the kidneys and excreted by way of the urinary bladder. In mice, the concentrations of radiolabel in the empty urinary bladder from a single 225-mg/kg dose were greater than 1,000 µg/mL in all samples taken between 30 minutes and 4 hours after compound administration (Hart 1976). In rats, concentrations of the label were 1,000 µg/mL or greater for all measurements taken between 2 and 6 hours after dosing (Hart 1976).

**Storage.** No studies were located regarding storage of the parent compound or its metabolites in humans after inhalation, oral, or dermal exposure to diisopropyl methylphosphonate.

Animal data suggest that there is no storage of diisopropyl methylphosphonate, IMPA, or MPA in the body, although portions of the [<sup>3</sup>H]-label may become incorporated in biomolecules leading to some retention of label in the form of an unextractable labeled compound (Little et al. 1986, 1988).

The concentrations of [<sup>14</sup>C]-radiolabel were very low in the adipose deposits of mice, rats, and dogs at 1, 2, and 3 days after administration of a single dose of diisopropyl methylphosphonate (Hart 1976). This demonstrates lack of storage in body lipids.

**Excretion.** No studies were located regarding the mechanism of excretion in humans after inhalation, oral, or dermal exposure to diisopropyl methylphosphonate.

Based on the data from animal studies, diisopropyl methylphosphonate is principally excreted in the urine as the metabolite IMPA (Hart 1976; Ivie 1980). Chromatographic behavior of urinary metabolites does not change after the urine is treated with glucuronidase and sulfatase, so there is no conjugation of diisopropyl methylphosphonate or IMPA by microsomal enzymes (Hart 1976). There was minimal excretion of diisopropyl methylphosphonate metabolites in bile (Hart 1976) or in the milk of a lactating cow (<1 %) (Palmer et al. 1979).

### 2.4.2 Mechanisms of Toxicity

**Effects of Metabolism on Toxicity.** Whether the toxic effects seen after exposure to diisopropyl methylphosphonate are caused by the parent compound or its metabolites is unknown. Studies of IMPA show that acute-duration exposure to IMPA results in reduced motor activity, prostration, and ataxia-effects also seen after exposure to diisopropyl methylphosphonate (EPA 1992). Other studies (Little et al. 1986, 1988) show that IMPA, the major metabolite of diisopropyl methylphosphonate, has an affinity for both lung and brain tissues and will bind to proteins in these tissues-effects that were not seen after exposure to diisopropyl methylphosphonate (EPA 1992; Little et al. 1988). These data and other data on the toxicity of IMPA neither support nor contradict the data found in the diisopropyl methylphosphonate studies, so it is not possible to attribute the effects after exposure to diisopropyl methylphosphonate to IMPA. Metabolites of IMPA other than MPA have not been identified.

**Target Organ Toxicity.** The mechanism of toxicity for diisopropyl methylphosphonate on the red blood cells is not understood. However, it has been suggested that its toxicity in general may be due to the ability of the chemical to inhibit the enzyme cholinesterase (NIOSH 1981). However, no quantitative data exist to support this hypothesis, and Bucci et al. (1992, 1994) indicated that there were no signs of acetylcholinesterase inhibition in mink that received diisopropyl methylphosphonate in the diet at concentrations of 2,700, 5,400, or 8,000 ppm (400, 827, or 1,136 mg/kg/day) for 90 days. The effects of diisopropyl methylphosphonate exposure in acute animal toxicity studies range from ataxia and decreased activity to coma and death at higher doses (Aulerich et al. 1979; Hart 1976). These effects suggest that diisopropyl methylphosphonate, like a number of other organophosphorous compounds, affects the nervous system. This mechanism, however, is speculative, and biologically significant changes in brain cholinesterase activity have not been measured. Neurotoxicity data for diisopropyl methylphosphonate are limited; therefore, the mechanism of nervous system toxicity is not known.

**Carcinogenesis.** There are no human or animal data concerning the carcinogenic effects of diisopropyl methylphosphonate. Diisopropyl methylphosphonate has been classified by the EPA as a Group D substance, not classifiable as to human carcinogenicity (EPA 1994).

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**2.4.3 Animal-to-Human Extrapolations**

No studies were located regarding toxicokinetic data in humans. Limited information is available regarding the toxicokinetic differences among animal species. Rats, mice, mink, and dogs showed rapid absorption, wide distribution, and over 90% urinary excretion of diisopropyl methylphosphonate or its metabolites. However, the rates of absorption and patterns of distribution varied (Hart 1976; Weiss et al. 1994). The mechanism of toxicity is also undetermined. From the limited data available, it is not possible to determine the degree of correlation between humans and animals

The use of data derived from mink for human health assessment, and in particular the mink diisopropyl methylphosphonate data, has been recommended by Calabrese (1990). His recommendation is primarily based on the documentation of “the biology of the mink” available in the scientific literature (including information concerning “nutrition, reproduction and breeding, physiology and biochemistry, disease and parasitism, anatomy and embryology, genetics, behavior, ecology, paleontology and taxonomy”) and on the development of a historical database and quantitative assessment of natural background mortality. Calabrese indicated that mink have been used for toxicological research into a number of compounds (e.g., lead, mercury, polychlorinated biphenyls, selenium, and dioxin). It should be noted, however, while mink can be used as a predictive animal model for animal-to-human extrapolations, the database of mink studies being developed has far fewer data than are available for rodents, rabbits, and dogs, species that have traditionally been used for toxicity studies.

**2.5 RELEVANCE TO PUBLIC HEALTH**

No animal or human data were available for inhalation exposure. There are no data regarding effects in humans after oral exposure. Information is available in animals regarding health effects following acute, intermediate, and chronic oral ingestion of diisopropyl methylphosphonate. The animal data obtained after oral exposure indicate that diisopropyl methylphosphonate is moderately toxic after acute bolus exposure but has a lower order of toxicity after intermediate and chronic exposures in food. No data were found on the toxicity of diisopropyl methylphosphonate after exposure in drinking water. Further, diisopropyl methylphosphonate is rapidly metabolized and excreted and does not accumulate. It does not appear to have reproductive or developmental effects. At the doses tested, it does not appear to be an acetylcholinesterase inhibitor, although this issue has not been resolved yet. Limited data are available for dermal exposure in humans and animals. Diisopropyl methylphosphonate does not appear to be a skin irritant; dermal

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absorption data on pigs (Snodgrass and Metker 1992) indicated skin absorption in humans is expected to be less than 10%. It does appear to be an eye irritant. Diisopropyl methylphosphonate is not a strong sensitizer in animals, although this conclusion is based on a preliminary study. Animals have shown hematological effects to diisopropyl methylphosphonate after oral exposure.

### **Minimal Risk Levels for Diisopropyl Methylphosphonate**

#### *Inhalation MRLS*

Minimal risk level (MRL) values were not derived for acute, intermediate, or chronic inhalation exposures to diisopropyl methylphosphonate because no toxicity data were identified concerning the inhalation of the compound.

#### *Oral MRLs*

- An intermediate-duration MRL of 0.8 mg/kg/day was derived from a NOAEL of 75 mg/kg/day (3,000-ppm concentration in the diet) at which no hematological or other effects were noted in beagle dogs (Hart 1980). The NOAEL was divided by an uncertainty factor of 100 (10 each for interspecies and intraspecies variability).

Purebred beagles, 4 males and 4 females, received diisopropyl methylphosphonate in the diet at doses of 4, 38, or 75 mg/kg/day for 90 days (Hart 1980). A control group of four males and four females was also maintained. At the outset of the study and at 4, 8, and 13 weeks, hemograms and clinical chemistry parameters were determined for all of the dogs. The dogs appeared in good condition throughout the study. No hematological effects were ascribed to diisopropyl methylphosphonate. At the termination of the study, a gross necropsy was performed on all of the dogs and no meaningful changes were observed. In addition, the liver, brain, thyroid, kidneys, adrenal glands, testes, ovaries, heart, and spleen were removed and weighed. No significant weight changes were noted. Microscopic examination of major organs did not reveal any adverse effects.

Although this study (Hart 1980) did not identify an effect level, the NOAEL is below the LOAEL found in all studies examining the toxicity of diisopropyl methylphosphonate. The LOAEL for diisopropyl methylphosphonate is 262 mg/kg/day for male mink and 330 mg/kg/day for female mink

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(Bucci et al. 1997), doses at which statistically significant decreases in plasma cholinesterase (butyrylcholinesterase) but not RBC cholinesterase (acetylcholinesterase) activity were observed (Bucci et al. 1997). In general, a decrease in plasma cholinesterase activity is considered to be a marker of exposure rather than a marker of adverse effect, while a decrease in RBC acetyl-cholinesterase activity is a neurological effect thought to parallel the inhibition of brain acetyl-cholinesterase activity and is thus considered an adverse effect. Diisopropyl methylphosphonate was not found to inhibit red blood cell cholinesterase at doses at which plasma cholinesterase was significantly inhibited. No effects were observed in males at 45 mg/kg/day (Bucci et al. 1997) or at 63 mg/kg/day (Bucci et al. 1994), and no effects were observed in females at 82 mg/kg/day (Bucci et al. 1994), or at 57 mg/kg/day (Bucci et al. 1997).

Adverse effects (shortened RBC survival, increased Heinz body formation, increased number of reticulocytes, and reduced blood cell counts) were observed at 747 mg/kg/day in males and 907 mg/kg/day in females (average 827 mg/kg/day) (Bucci et al. 1994). Although not statistically significant, the number of Heinz bodies was increased relative to the controls and to the rats treated at 400 mg/kg/day. The observed effects are consistent with a direct effect on RBC and a decrease in their survival.

EPA derived a reference dose (RfD) of 0.08 mg/kg/day based on the same NOAEL (75 mg/kg/day) from the Hart (1980) study. The RfD, however, utilized an additional uncertainty factor of 10 to extrapolate to chronic exposure.

- A chronic-duration MRL of 0.6 mg/kg/day was derived from a NOAEL of 57 mg/kg/day (490-ppm concentration in the diet) at which no hematological or other effects were noted in female mink (Bucci et al. 1997). The NOAEL was divided by an uncertainty factor of 100 (10 for extrapolations from animals to humans and 10 for human variability).

In a two-generation reproductive study, Ranch Wild mink received diisopropyl methylphosphonate in the diet at doses of 0, 16, 45, or 262 mg/kg/day (males) or 0, 20, 57, or 330 mg/kg/day (females) (Bucci et al. 1997). F<sub>1</sub> -generation females were treated for up to 13 months, while other generations and F<sub>1</sub> males were treated for 8 months or less. Two groups of control animals were used. In addition to standard examinations (body and organ weights, hematology, clinical chemistry, gross and

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histopathological parameters), plasma and whole blood cholinesterase, and brain acetylcholinesterase were measured. Ovarian follicles were also counted in high-dose females.

No effects were observed in F<sub>1</sub> females at 57 mg/kg/day that were attributable to diisopropyl methylphosphonate after 13 months of exposure. However, at 330 mg/kg/day, Heinz body counts were increased in F<sub>1</sub> females. There was a 31% decrease in plasma cholinesterase in animals fed 330 mg/kg/day for 13 months, but this is not considered to be biologically significant. There was also a significant increase in ovarian follicles among animals at this concentration (the only level examined). However, because treated dams of both generations produced as many offspring as the control animals, the biological significance of these findings is unclear.

This study is supported by intermediate-duration NOAELs for hematological effects of 75 mg/kg/day in a dog study (Hart 1980) and 45 mg/kg/day (males) and 57 mg/kg/day (females) in a mink study (Bucci et al. 1997). In the Bucci et al. (1997) mink study, the next highest level, 262 or 330 mg/kg/day in males and females, respectively, produced hematological changes that included increased Heinz body counts, reticulocytes, mean cell volume, and decreased RBC counts.

No MRL was derived for acute oral exposure to diisopropyl methyl phosphonate because the data were considered too limited to determine an appropriate threshold and derive an MRL.

**Death**

No studies were located regarding death in humans after inhalation, oral, or dermal exposure to diisopropyl methylphosphonate.

Oral LD<sub>50</sub>s of 1,125 and 826 mg/kg have been reported for male and female rats, respectively (Hart 1976). Swiss Webster mice yielded oral LD<sub>50</sub>s of 1,041 and 1,363 mg/kg for males and females, respectively (Hart 1976). No deaths occurred in female rats receiving diisopropyl methylphosphonate for 10 days in the diet at doses of 10, 30, or 300 mg/kg/day on days 6-15 of gestation (Hart 1980). An oral LD<sub>50</sub> of approximately 750 mg/kg for calves was estimated in a study of pairs of calves given 62.5, 125, 250, 500, or 1,000 mg/kg diisopropyl methylphosphonate in a gelatin capsule (Palmer et al. 1979). All animals at the highest dose died. No deaths of adult rats were recorded in a three-generation study of reproductive effects in rats receiving diisopropyl methylphosphonate in the diet at 0, 30, or 300 mg/kg/day (Hart 1980). Four male rats



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died (1 control, 1 low dose, and 2 high dose) out of a total of 256 animals in a 90-day study in which rats received diisopropyl methylphosphonate in the diet at doses of 30, 100, or 3,000 mg/kg/day. The study author concluded that the deaths were not treatment related and that there was no evidence of toxicity (Hart 1976). Similarly, the 3 deaths in the 91-mg/kg/day group were not considered evidence of toxicity since no deaths occurred in the low- or high-dose groups among 180 mice receiving diisopropyl methylphosphonate in the diet at doses of 27, 91, or 273 mg/kg/day (Hart 1976). No mortality occurred, nor were there judged to be any signs of toxicity in beagle dogs receiving dietary doses of 4, 13, or 38 mg/kg/day for 2 weeks (Hart 1976). No deaths or evidence of toxicity were attributable to diisopropyl methylphosphonate administered for 26 weeks in the drinking water of rats at doses of  $6.6 \times 10^{-7}$ ,  $6.6 \times 10^{-5}$ , 0.011, or 1.1 mg/kg/day (Army 1978). However, as discussed in Section 2.2.2.1, there is some confusion concerning the concentration units and purity of the diisopropyl methylphosphonate used in the Army (1978) study (EPA 1989), and therefore results from the Army (1978) study are considered inappropriate for human health risk assessment.

Adult female mink dosed by gavage with single doses of diisopropyl methylphosphonate (75, 150, 300, 450, 500, 550, and 600 mg/kg) yielded an LD<sub>50</sub> of 503 mg/kg (Aulerich et al. 1979). No mortality related to diisopropyl methylphosphonate was noted in juvenile pastel mink receiving dietary doses of 0.2, 2, 17, 201, or 1,852 mg/kg/day in a 21-day feed study. The calculated doses of diisopropyl methylphosphonate suggest that it is not a cumulative toxin since the amounts ingested with food exceed the LD<sub>50</sub> found in a companion study. The effects may also have been lessened because it was mixed with food (Aulerich et al. 1979).

During a 90-day study, no mortality occurred in male or female mink receiving diisopropyl methylphosphonate in the diet at doses of 7, 63, 345, 747, or 1,009 mg/kg/day for males and 9, 82, 455, 908, or 1,264 mg/kg/day for females (Bucci et al. 1992, 1994). No treatment-related mortality was observed in parental or F<sub>1</sub> animals in a two-generation reproductive study in mink in which animals consumed 16, 45, or 262 mg/kg/day (males) or 20, 57, or 330 mg/kg/day (females) of diisopropyl methylphosphonate in the diet for up to 13 months (Bucci et al. 1997). A slight though significant increase in mortality (5/24) was noted among female dark variety mink in the high-dose group in a reproductive study approximately 49 weeks in duration in which the animals received doses of 11, 37, or 95 mg/kg/day diisopropyl methylphosphonate in the diet (Aulerich et al. (1979). It is not clear if the deaths in the Aulerich et al. (1979) study were actually a result of diisopropyl methylphosphonate treatment. No symptoms of toxicity were noted before the mink died. In a concurrent study conducted to assess the toxicity of dicyclopentadiene, which used mink from the same lot, the mortality in the untreated female mink was 4 of 24; 2 died between the time of mating and lactation. It should also be noted that mink have a relatively high natural mortality (EPA 1989). The natural mortality for 1st year mink in a commercial fur ranch operation approaches 6% annually, and up to 15% of

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lactating females may die in the late gestation period from “nursing sickness” (Schneider and Hunter 1993). Although it is not clear when in relation to lactation the mink in the Aulerich et al. (1979) study died, some female mink did die during the period that included lactation. The Bucci et al. (1994, 1997) studies, which did not observe deaths in mink that were fed higher doses, provide further evidence that the deaths of mink in the Aulerich et al. (1979) study probably were not related to diisopropyl methylphosphonate treatment.

Four out of 12 rabbits died after a single dermal administration of diisopropyl methylphosphonate (dose range, 200 to 2,000 mg/kg), yielding a dermal LD<sub>50</sub> of 1,100 mg/kg. The author notes that there was “no antemortem indication of systemic toxicity based on general appearance and behavior.” In fact, the body weights of the surviving animals increased during the 2-week observation period. Further, no edema or eschar formation of the skin was observed, and the hair growth appeared normal in the shaved areas. No treatment-related abnormalities were noted during necropsy (Hart 1976). Based on the available animal data, it is highly improbable that people living near the RMA will be exposed to lethal exposures of diisopropyl methylphosphonate in their drinking water.

**Systemic Effects**

***Respiratory Effects.*** No studies were located regarding respiratory effects in humans after inhalation, oral, or dermal exposure or in animals after dermal or inhalation exposure to diisopropyl methylphosphonate.

Oral ingestion of diisopropyl methylphosphonate does not appear to induce respiratory effects. Necropsy of both male and female rats that died as the result of a single dose (928, 1362, or 2,000 mg/kg) of diisopropyl methylphosphonate administered by gastric intubation revealed some hyperemia of the lungs; however, most animals displayed no abnormalities (Hart 1976). No abnormal necropsy findings were noted in Swiss Webster mice dosed similarly in a companion study (Hart 1976). No important abnormalities were noted in the necropsy of rats receiving diisopropyl methylphosphonate in the diet (0, 100, or 300 mg/kg/day) for 90 days (Hart 1976). A study in pairs of calves given 62.5, 125, 250, 500, or 1,000 mg/kg diisopropyl methylphosphonate in a gelatin capsule (Palmer et al. 1979) showed that animals at the highest dose displayed pulmonary emphysema; however, no effects were seen at non-lethal doses. No noteworthy deviations were noted in the necropsy or histopathology examination of male or female beagles receiving dietary doses of 38 mg/kg/day (Hart 1976). No gross lesions were noted in rats subsequent to receiving drinking water containing diisopropyl methylphosphonate at doses of  $6.6 \times 10^{-7}$ ,  $6.6 \times 10^{-5}$ , 0.011, or 1.1 mg/kg/day (Army 1978). However, as described in Section 2.2.2.1, there is some confusion concerning

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the concentration units and purity of the diisopropyl methylphosphonate used in the Army (1978) study (EPA 1989), and therefore results from Army (1978) are considered inappropriate for human health risk assessment. Available animal data indicates that no respiratory effects will be expected in the population living near the RMA.

***Cardiovascular Effects.*** No studies were located regarding cardiovascular effects in humans after inhalation, oral, or dermal exposure or in animals after dermal or inhalation exposure to diisopropyl methylphosphonate.

Ingestion of diisopropyl methylphosphonate does not appear to induce cardiovascular effects. No anomalous necropsy or histopathological effects were noted in rats receiving diisopropyl methylphosphonate in the diet at doses of 30, 100, or 300 mg/kg/day for 90 days (Hart 1976). Similarly, no abnormalities were noted in mice receiving the compound in the diet at doses of 27, 91, or 273 mg/kg/day for 90 days (Hart 1976). A study in pairs of calves given 62.5, 125, 250, 500, or 1,000 mg/kg diisopropyl methylphosphonate in a gelatin capsule (Palmer et al. 1979) showed that animals at the highest dose displayed petechiae in the coronary grooves and ecchymosis in the gastrointestinal tract. However, no effects were seen at non-lethal doses. Gross necropsy and histopathological examination revealed hemorrhage and erythrophagocytosis in the mesenteric lymph node, although the incidences in the treated and control groups did not indicate a relation with the treatment. In no instance was the tissue or organ examined different from corresponding tissue or organ in the same or other treatment groups or in the controls in beagle dogs receiving the compound in the diet at doses of 4, 13, or 38 mg/kg/day for 14 days (Hart 1976). No deviation in heart weight or abnormal histopathological findings were reported in beagles that received diisopropyl methylphosphonate in the diet at doses of 4, 38, or 75 mg/kg/day for 90 days (Hart 1980).

No significant changes in heart weight or histopathological findings were noted in mink receiving diisopropyl methylphosphonate in the diet at doses of 0.2, 2, 17, 201, or 1,852 mg/kg/day (Aulerich et al 1979). Gross and histopathological examination of mink that received 11, 37, or 95 mg/kg/day revealed no consistent pathological changes (Aulerich et al. 1979). Persons living near the RMA who are exposed to low levels of diisopropyl methylphosphonate are not expected to experience cardiovascular effects.

***Gastrointestinal Effects.*** No studies were located regarding gastrointestinal effects in humans after inhalation, oral, or dermal exposure or in animals after dermal or inhalation exposure to diisopropyl methylphosphonate.

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A study in pairs of calves given 62.5, 125, 250, 500, or 1,000 mg/kg diisopropyl methylphosphonate in a gelatin capsule (Palmer et al. 1979) showed that animals at the highest dose displayed acute gastroenteritis and ecchymotic hemorrhaging. However, no effects were seen at non-lethal doses. It is not expected that persons living near the RMA and exposed to low levels of diisopropyl methylphosphonate would experience gastrointestinal effects.

***Hematological Effects.*** No studies were located regarding hematological effects in humans after inhalation, oral, or dermal exposure or in animals after dermal or inhalation exposure to diisopropyl methylphosphonate.

No significant differences were noted in the hemocytology of ducks, rats, calves, or dogs subsequent to ingestion of diisopropyl methylphosphonate in acute or intermediate protocols (Aulerich et al. 1979; Hart 1976, 1980; Palmer et al. 1979). However, increase in activated partial thromboplastin time (APTT) was observed in calves dosed orally with DIMP in an acute study (Palmer et al. 1979). Alternatively, mink ingesting diisopropyl methylphosphonate for 90 days demonstrated significant changes in hemocytology. Such changes included depressed hematocrits, increased numbers of reticulocytes, and an increase in Heinz bodies (Bucci et al. 1994, 1997). The increase in Heinz bodies suggests that diisopropyl methylphosphonate may cause oxidative damage to the tertiary structure of hemoglobin, thus reducing the lifespan of RBCs, which in turn ultimately induces hematopoiesis. The RBC data and the reticulocyte data are consistent with a mechanism for this hematopoietic response. A marginal increase in the incidence and severity of splenic extramedullary hematopoiesis was observed in male mink exposed to diisopropyl methylphosphonate in the diet at 1,009 mg/kg/day for 90 days (Bucci et al. 1994) and in female mink exposed to 330 mg/kg/day in the diet for 4 months (Bucci et al. 1997). This effect was not observed in the male mink exposed to 747 mg/kg/day in the Bucci et al. (1994) study, or in the female mink exposed to 57 mg/kg/day in the Bucci et al. (1997) study. Results of groundwater monitoring near the RMA in 1997 indicated the maximum detected concentration of DIMP is 1,500 µg/L; and DIMP is not detected in surface soil. The exposure dose associated with the maximum concentration of DIMP in the groundwater is below the chronic MRL of 0.6 mg/kg/day for hematological effect. Therefore, no hematological effects are expected to occur in persons exposed to diisopropyl methylphosphonate while living near the RMA.

***Hepatic Effects.*** No studies were located regarding hepatic effects in humans after inhalation, oral, or dermal exposure or in animals after dermal or inhalation exposure to diisopropyl methylphosphonate.

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No changes of toxicological significance were noted in SGOT, SGPT, alkaline phosphatase, liver weight, or liver histopathology in rats, mice, or dogs receiving diisopropyl methylphosphonate in their feed in acute or intermediate protocols (Hart 1976). However, rats pretreated with diisopropyl methylphosphonate in the diet (450 mg/kg/day for 4 days) showed a marked decrease in sleep time subsequent to the administration of hexobarbital, which is consistent with the induction of the P-450 mixed-function oxidase system. Liver weights, however, were not increased (Hart 1976).

Juvenile male pastel mink that ingested 1,852 mg/kg/day diisopropyl methylphosphonate for 21 days showed a significant decrease in liver weight. However, no associated hepatic lesions were noted. The study authors indicated that the decreases in organ weights noted in this high-dose group may be related to significantly reduced food consumption and were not necessarily a function of toxicity. It was noted that, since a pair-fed control was not maintained for this study, the causes of the organ weight differences were not easily identifiable. Finally, the study authors considered the compound nontoxic in this 21-day study (Aulerich et al. 1979).

The available animal data suggest that diisopropyl methylphosphonate is not a hepatotoxin, at least in the doses and time courses studied. Therefore, exposure of persons living near the RMA is not expected to cause hepatic effects.

**Renal Effects.** No studies were located regarding renal effects in humans after inhalation, oral, or dermal exposure or in animals after dermal or inhalation exposure to diisopropyl methylphosphonate.

Renal toxicity data are limited. In rats, dogs, and mice, no deviations from normal were noted in urinalyses (color, specific gravity, pH, sugar albumin, ketones, and microscopic examination of sediment), weights, or histopathology of kidneys after administration of diisopropyl methylphosphonate in the diet (Hart 1976) or drinking water (Army 1978). As discussed in Section 2.2.2.1, however, there is some confusion concerning the concentration units and purity of the diisopropyl methylphosphonate used in the Army (1978) study (EPA 1989). Therefore, results from the Army (1978) study are considered inappropriate for human health risk assessment. A study in pairs of calves given 62.5, 125, 250, 500, or 1,000 mg/kg diisopropyl methylphosphonate in a gelatin capsule (Palmer et al. 1979) showed that animals at the 500-mg/kg dose and above displayed mild congestion of the renal cortex; however, no effects were seen at the lower doses.

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Significant kidney weight loss, but no renal pathology, was found in mink that ingested 1,852 mg/kg/day for 21 days. The study authors indicated that the decreases in organ weights noted in this high-dose group may be related to significantly reduced food consumption and were not necessarily a function of toxicity (Aulerich et al. 1979). No renal pathology was noted in a similar study (Bucci et al. 1992, 1994). The absence of renal lesions and the healthy appearance of the animals throughout the 90-day study are in agreement with Aulerich et al. (1979). Further, the decrease in kidney weight that was attributed to decreased food intake suggests that the renal toxicity of diisopropyl methylphosphonate may be limited. However, the available data are inadequate to draw firm conclusions.

***Dermal Effects.*** No studies were located regarding dermal effects in humans after inhalation or oral exposure or in animals after oral or inhalation exposure to diisopropyl methylphosphonate.

Skin irritation was noted in wildlife officers at the IWA who handled dead or sick ducks without gloves. Analysis of the pond water indicated the presence of a number of organic and inorganic contaminants, including diisopropyl methylphosphonate. Although the investigators concluded that diisopropyl methylphosphonate contributed to the effects, a number of other compounds were identified in the pond, the presence of which makes it unclear whether diisopropyl methylphosphonate was related to the irritation (NIOSH 1981).

Only minimal indications of skin irritation were noted in New Zealand White rabbits that received a single dose of diisopropyl methylphosphonate (200, 630, or 2,000 mg/kg) applied to intact or abraded skin (Hart 1976). Diisopropyl methylphosphonate was not considered a strong sensitizer in guinea pigs (Hart 1976). These studies, however, do not conclusively predict the human risk of skin irritation or sensitivity, but it is not believed that levels found in water near the IWA would cause dermal effects.

***Ocular Effects.*** No studies were located regarding ocular effects in humans after inhalation, oral, or dermal exposure or in animals after inhalation exposure to diisopropyl methylphosphonate.

An ophthalmic examination was performed during week 12 of a 13-week study of rats that received diisopropyl methylphosphonate in their feed (30, 100, or 300 mg/kg/day); 12 of 64 animals displayed some opacity of the lens (Hart 1976). Although the finding was considered normal, the study author suggested that lens opacity may deserve attention in longer studies. No changes were noted in the eyes of beagles that received diisopropyl methylphosphonate in their food (at doses up to 75 mg/kg/day) for 90 days (Hart 1980)

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or in mink that received dietary concentrations up to 330 mg/kg/day in a two-generation reproductive study (Bucci et al. 1997).

The potential of diisopropyl methylphosphonate to cause eye irritation was evaluated by direct application of the compound to the conjunctival sac of one eye in each of nine New Zealand White rabbits. Significant irritation of the conjunctivae was observed in all rabbits, and the corneal surface was characterized by diffuse opacity. The opacity was temporary and cleared within 8 days. Irrigation with lukewarm water following application of diisopropyl methylphosphonate reduced but did not prevent irritation (Hart 1976). These data indicate that diisopropyl methylphosphonate is probably an eye irritant in humans at sufficiently high concentrations. However, eye irritation is not expected to occur as a result of groundwater contamination in persons living near the RMA.

***Body Weight Effects.*** No studies were located regarding body weight effects in humans after inhalation, oral, or dermal exposure or in animals after inhalation or dermal exposure to diisopropyl methylphosphonate.

No toxicologically important compound-related weight loss in rats (Army 1978; Hardisty et al. 1977; Hart 1976), pregnant rats (Hart 1980), calves (Palmer et al. 1979), or dogs (Hart 1980) was noted subsequent to acute (Hart 1980) or intermediate (Army 1978; Hardisty et al. 1977; Hart 1976, 1980) oral administration of diisopropyl methylphosphonate. However, as discussed in Section 2.2.2.1, there is some confusion concerning the concentration units of the diisopropyl methylphosphonate used in the Army (1978) and Hardisty et al. (1977) studies (EPA 1989), and the actual doses received may be lower. Therefore, results from the Army (1978) and Hardisty et al. (1977) studies are considered inappropriate for human health risk assessment. In addition, the purity of the compound was only 65% in the Army (1978) study. Significant weight loss and decreased food consumption were noted in male and female pastel mink that received diisopropyl methylphosphonate in their feed at a dose of 1,852 mg/kg/day. However, subsequent to treatment (during the recovery period), the group displayed increased food consumption, suggesting that the weight loss may have been due to decreased food consumption resulting from a palatability problem and not a function of toxicity (Aulerich et al. 1979). To determine if unpalatability of the feed was the cause of the weight loss, an attempt was made to “pair feed” a control group to match voluntary food consumption. Similar to the findings of Aulerich and coworkers (1979), mink that received feed containing diisopropyl methylphosphonate at doses of 1,009 mg/kg/day for males and 1,264 mg/kg/day for females consumed significantly less feed than did the controls and demonstrated decreased body weight throughout the study. Although only partially successful, the pair-feed data for the male high-dose group provided substantial

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evidence that the weight loss was the result of decreased food consumption rather than diisopropyl methylphosphonate toxicity (Bucci et al. 1994). No weight loss was noted in mink subsequent to oral administration of diisopropyl methylphosphonate for 49 weeks (Aulerich et al. 1979) or up to 13 months in a two-generation reproductive study (Bucci et al. 1997). Mallard ducks exposed to a single dose of 1,800 mg/kg by gavage or fed 1,796 mg/kg/day for 5 days exhibited decreased weight gain, although this may have been because of refusal to eat feed containing a very high concentration of the chemical (Aulerich et al. 1979). As a whole, these data suggest that intermediate-duration oral administration of diisopropyl methylphosphonate does not cause serious toxicity.

**Immunological and Lymphoreticular Effects.** No studies were located regarding immunological and lymphoreticular effects in humans after inhalation, oral, or dermal exposure or in animals after inhalation or dermal exposure to diisopropyl methylphosphonate.

Histological changes in the spleen related to diisopropyl methylphosphonate intake were not observed in male or female rats exposed to 1.1 mg/kg/day of diisopropyl methylphosphonate in their drinking water for 26 weeks (Army 1978). However, as stated in Section 2.2.2.1, there is some confusion concerning the concentration units and purity of the diisopropyl methylphosphonate used in the Army (1978) study (EPA 1989). Therefore, results from the Army (1978) study are considered inappropriate for human health risk assessment. No changes in spleen weight were noted in male or female mink exposed to 1,852 mg/kg/day of diisopropyl methylphosphonate in the diet for 21 days (Aulerich et al. 1979). Similarly, spleen weight measurements and gross and histopathological examinations did not reveal any significant immunological or lymphoreticular effects in male or female mink exposed to 95 mg/kg/day of diisopropyl methylphosphonate in the diet for 49 weeks (Aulerich et al. 1979). A marginal increase in the incidence and severity of hematopoiesis in the spleen was observed in male mink exposed to 1,009 mg/kg/day of diisopropyl methylphosphonate in the diet for 90 days (Bucci et al. 1994). An increase in hematopoiesis was not observed in male mink exposed to 747 mg/kg/day. Splenic hematopoiesis accompanied by increased spleen weights also were observed in female mink fed 330 mg/kg/day diisopropyl methylphosphonate in the diet for 4 months (Bucci et al. 1997). However, in both of these studies the splenic extramedullary hematopoiesis was probably a compensatory effect due to damage to erythrocytes and not a direct lymphoreticular effect. Data are insufficient to determine if diisopropyl methylphosphonate affects the function of the immune system.



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**Neurological Effects.** No studies were located regarding neurological effects in humans after inhalation, oral, or dermal exposure or in animals after inhalation or dermal exposure to diisopropyl methylphosphonate.

Single-dose oral toxicity studies in rats, mice, and cattle indicate that signs of diisopropyl methylphosphonate intoxication include decreased activity, ataxia, tympanitis, and prostration within 1-4 hours after dosing (Hart 1976; Palmer et al. 1979). Mink that received 1,852 mg/kg/day diisopropyl methylphosphonate in their feed displayed aggressive behavior. However, it was concluded that this behavior was probably due to hunger resulting from the unpalatability of the feed (Aulerich et al. 1979).

Inconsistent fluctuations were observed in RBC and plasma cholinesterase activity in rats that received diisopropyl methylphosphonate in the diet (30, 100, or 300 mg/kg/day) for 90 days. The most significant difference was a decrease in plasma cholinesterase activity in the 300-mg/kg/day female group at 13 weeks (Hart 1976). The study author pointed out, however, that the control group value was exceptionally high compared to those at other time intervals (Hart 1976). In a go-day study of diisopropyl methylphosphonate in the diet (4, 38,75 mg/kg/day) of dogs, a slight increase in plasma cholinesterase was observed; however, problems with the experimental protocol (failure to measure cholinesterase at 4 and 8 weeks) make any interpretations difficult (Hart 1980). Dark brown Ranch Wild mink that ingested 345, 747, or 1,009 mg/kg/day for males and 455, 908, or 1,284 mg/kg/day for females demonstrated decreased plasma cholinesterase compared to the controls, although the change was rapidly reversible. No effect was noted in erythrocyte acetylcholinesterase activity. Similarly, in a two-generation reproductive study using the same species fed 16, 45, or 262 mg/kg/day (males) or 20, 57, or 330 mg/kg/day (females) of diisopropyl methylphosphonate in the diet for up to 13 months, high-dose females of the parental and F<sub>1</sub> generations showed decreased plasma cholinesterase compared to controls (Bucci et al. 1997). The study authors indicated that plasma cholinesterase is labile and sufficiently unrelated to the nervous system, and that therefore any changes in the low-dose groups were toxicologically unimportant. Further, they noted that even in the high-dose groups with large decreases (61%) in plasma cholinesterase, there was no reduction in erythrocyte or brain cholinesterase and no signs of acetylcholinesterase inhibition (Bucci et al. 1992, 1994, 1997). Based on the studies discussed, it appears that diisopropyl methylphosphonate may inhibit plasma cholinesterase but not RBC cholinesterase. Consistent decreases in plasma cholinesterase may not have been observed in rats and dogs because they were treated with lower doses of diisopropyl methylphosphonate. These data do suggest that at the doses tested, the main effect of diisopropyl methylphosphonate appears to be on the red blood cells, and not on acetylcholinesterase inhibition. In the absence of other systemic effects, it may be speculated that the neurotoxic effects of diisopropyl methylphosphonate may be the cause of death

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in those animals receiving sufficient amounts of the compound. However, it is not expected that persons living near the RMA would experience these effects.

**Reproductive Effects.** No studies were located regarding reproductive effects in humans after inhalation, oral, or dermal exposure or in animals after inhalation or dermal exposure to diisopropyl methylphosphonate.

No toxicologically important dose-related reproductive responses were noted in a single-generation reproductive study in which male and female rats received diisopropyl methylphosphonate in the drinking water (0.009 or 0.9 mg/kg/day) for 10 weeks (Hardisty et al. 1977), in a three-generation reproductive study (with 2 matings per generation) in which male and female rats received diisopropyl methylphosphonate in feed at doses of 30 or 300 mg/kg/day (Hart 1980), or in reproductive studies in which mink received feed containing diisopropyl methylphosphonate at doses of 11, 37, or 95 mg/kg/day (Aulerich et al. 1979) or up to 262 mg/kg/day (males) or 330 mg/kg/day (females) (Bucci et al. 1997). However, in the Bucci et al. (1997) study, there was a significant increase in ovarian follicles in high-dose F<sub>1</sub> females, the only treated animals examined, compared to controls. Because there was no difference in the actual breeding outcome (number of offspring) in F<sub>0</sub> or F<sub>1</sub> females, the study authors concluded that there was insufficient evidence to suggest that the increase in ovarian follicles represented an adverse effect. However, the authors also reported that it is possible that these data represent a disrupted follicular maturation process. Because only the high-dose animals in this study were examined for this end point, a NOAEL could not be established for it.

As discussed in Section 2.2.2.1, there is some confusion regarding the actual doses the animals were exposed to in the Hardisty et al. (1977) study. Therefore, results from Hardisty et al. (1977) are considered inappropriate for human health risk assessment. A study in Mallard ducks revealed no changes in gonad weight or maternal death in animals given up to 10,000 ppm diisopropyl methylphosphonate in their feed beginning 10 weeks prior to egg production and terminating 10 weeks after egg production reached 50% (Aulerich et al. 1979). Doses (mg/kg/day) could not be calculated for this study. Reproductive effects are not expected to occur in persons living near the RMA who are exposed to diisopropyl methylphosphonate.

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**Developmental Effects.** No studies were located regarding developmental effects in humans after inhalation, oral, or dermal exposure or in animals after inhalation or dermal exposure to diisopropyl methylphosphonate.

Diisopropyl methylphosphonate was administered in the diet at doses of 10, 30, and 300 mg/kg/day to pregnant female rats on day 6 through day 15 of gestation. Observations of the uterine contents indicated no compound-related effects. Gross examination revealed no abnormalities of the internal organs. There was a slight increase in the abnormal-normal fetus ratio in the 300-mg/kg/day group; however, the effect was not statistically significant. Further, the changes noted did not suggest a specific area of involvement. The study author concluded that the administration of diisopropyl methylphosphonate in the diet at these concentrations produced no effect on development (Hart 1980). Up to 10,000 ppm diisopropyl methylphosphonate, given in the feed of Mallard ducks for 10 weeks prior to egg production through 10 weeks after egg production reached 50%, produced no increases in incubation parameters, shell thickness, number of cracked shells, developmental abnormalities, or chick 14-day survival (Aulerich et al. 1979). Doses (mg/kg/day) could not be calculated for this study. Developmental effects are not expected to occur after exposure to low levels of diisopropyl methylphosphonate near the RMA.

**Genotoxic Effects.** No studies were located regarding genotoxic effects in humans after inhalation, oral, or dermal exposure or in animals after inhalation or dermal exposure to diisopropyl methylphosphonate.

In a micronucleus assay using male B6C3F<sub>1</sub> mice dosed with 0, 250, 500, or 1,000 mg/kg diisopropyl methylphosphonate, a small but significant increase in micronuclei were observed at mid- and high-dose levels (DOD 1991a). However, the maximum response was found to be within the laboratory historical control limits. The assay was repeated and the increase in micronuclei was not observed, therefore, it is believed that diisopropyl methylphosphonate did not cause micronuclei induction in this experiment. Diisopropyl methylphosphonate was also negative for the induction of micronuclei in the rat bone marrow after administration of up to 800 mg/kg (DOD 1991b).

Treatment of male B6C3F<sub>1</sub> mice with 1,000 mg/kg diisopropyl methylphosphonate for 13 days did not result in a significant increase in DNA damage to either liver parenchymal cells or leukocytes in a single cell assay (DOD 1991 f). However, the same assay performed using Fischer-344 rats showed DNA damage to leukocytes but not liver parenchymal cells (DOD 1991g). However, since no increase in damage in the positive controls was found, the liver cell results are questionable.

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The mutagenic potential of diisopropyl methylphosphonate was investigated using the Ames assay. The compound was obtained from two different sources and tested on *Salmonella typhimurium* strains TA-1535, TA-1537, TA-1538, TA-98, and TA-100, both with and without S-9 activation. The compound did not demonstrate mutagenic activity in any of the assays (Hart 1980). Diisopropyl methylphosphonate was also negative for gene mutation in *Saccharomyces cerevisiae* (Hart 1980).

No DNA damage or increases in sister chromatid exchange were evident using Chinese hamster ovary cells treated with diisopropyl methylphosphonate either with or without activation (DOD 1991c, 1991e). Exposure of Chinese hamster ovary cells to diisopropyl methylphosphonate did increase clastogenic damage at levels of 5 and 15  $\mu\text{L}/\text{mL}$  in the absence and presence of S-9 activation, respectively (DOD 1991d). Diisopropyl methylphosphonate tested negative in the L5178Y TK+/- mouse lymphoma mutagenesis assay with S-9 activation, but without activation equivocal results were observed (DOD 1991h).

The results of these genotoxicity studies are summarized in Table 2-3 and Table 2-4.

**Cancer.** No quantitative data for carcinogenic effects in humans or animals were located involving inhalation, oral, or dermal exposure to diisopropyl methylphosphonate.

## 2.6 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 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

TABLE 2-3. Genotoxicity of Diisopropyl Methylphosphonate *In Vivo*

Species (test system)	End point	Results	Reference
Mammalian cells:			
Mouse (bone marrow)	Micronucleus induction	-	DOD 1991a
Rat (bone marrow)	Micronucleus induction	-	DOD 1991b
Mouse (liver, leukocytes)	DNA damage	-	DOD 1991f
Rat (liver)	DNA damage	-	DOD 1991g
Rat (leukocytes)	DNA damage	+	DOD 1991g

- = negative result; + = positive result

TABLE 2-4. Genotoxicity of Diisopropyl Methylphosphonate *In Vitro*

Species (test system)	End point	Results		Reference
		With activation	Without activation	
Prokaryotic organisms: <i>Salmonella typhimurium</i>	Gene mutation	-	-	Hart 1980
Eukaryotic organisms: Fungi: <i>Saccharomyces cerevisiae</i>	Gene mutation	-	-	Hart 1980
Mammalian cells: Chinese hamster ovary	DNA damage	-	-	DOD 1991c
Chinese hamster ovary	Chromosome aberration	+	+	DOD 1991d
Chinese hamster ovary	Sister chromatid exchange	-	-	DOD 1991e
Mouse lymphoma (L5178Y TK+/-)	Gene mutation	-	±	DOD 1991h

-- = negative result; + = positive result; ± = equivocal result

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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 diisopropyl methylphosphonate are discussed in Section 2.6.1.

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 diisopropyl methylphosphonate are discussed in Section 2.6.2.

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 2.8, Populations That Are Unusually Susceptible.

### **2.6.1 Biomarkers Used to Identify or Quantify Exposure to Diisopropyl Methylphosphonate**

No studies were located regarding the parent compound as a biomarker of exposure in humans. Animal exposure to diisopropyl methylphosphonate can be determined by measuring the parent compound in urine. However, since less than 3% of the parent compound was excreted in the urine of mice and rats, concentrations of the parent compound may be below detection levels (Hart 1976). No accumulation of the parent compound in tissues was identified. Elimination of diisopropyl methylphosphonate from the body occurs rapidly. Excretion is primarily in the urine as the metabolite IMPA.

IMPA, the major metabolite of diisopropyl methylphosphonate, has been suggested as a possible biomarker of exposure for diisopropyl methylphosphonate. The excretion of IMPA is not unique to diisopropyl methylphosphonate exposure; IMPA is also a major metabolite of GB (Sarin) (Little et al. 1986). Thus,

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IMPA in the urine may be an accurate biomarker of recent diisopropyl methylphosphonate exposure. IMPA can be detected in the blood as well as in the urine. However, since IMPA is cleared from the blood rapidly, its detection may only be useful for monitoring recent exposure.

**2.6.2 Biomarkers Used to Characterize Effects Caused by Diisopropyl Methylphosphonate**

No biomarkers have been identified to characterize the effects associated with exposure to diisopropyl methylphosphonate. At high doses in animals, diisopropyl methylphosphonate affects RBC and the nervous system. However, because the effects are not unique to diisopropyl methylphosphonate, they would not serve as useful biomarkers of effects. No clinical signs or symptoms in humans have been positively linked to diisopropyl methylphosphonate exposure.

**2.7 INTERACTIONS WITH OTHER SUBSTANCES**

No studies were located regarding interactions with other substances in humans or animals after exposure to diisopropyl methylphosphonate. However, the potential for multiple chemical interactions does exist. Diisopropyl methylphosphonate has been identified in the RMA in the presence of many other chemicals (such as endrin, dieldrin, dicyclopentadiene, bicycloheptadiene, diethyl benzene, and diethyl disulfide). The nervous system is a target of many of these compounds found at the RMA, including diisopropyl methylphosphonate. Therefore, there is potential for interaction, and studies examining multiple exposures would be useful in predicting risk to humans. Workers at the RMA reported skin irritation after dermal exposure to diisopropyl methylphosphonate at concentrations around 11.3 ppm in water. However, several other chemicals were also in the area (NIOSH 1981). Therefore, it is not clear if diisopropyl methylphosphonate contributed to the effects.

**2.8 POPULATIONS THAT ARE UNUSUALLY SUSCEPTIBLE**

A susceptible population will exhibit a different or enhanced response to diisopropyl methylphosphonate than will most persons exposed to the same level of diisopropyl methylphosphonate 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 may result in reduced detoxification or excretion of diisopropyl methylphosphonate, or compromised function of organs affected by diisopropyl methylphosphonate.



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Populations who are at greater risk due to their unusually high exposure to diisopropyl methylphosphonate are discussed in Section 5.6, Populations With Potentially High Exposures.

No populations that are unusually susceptible to the toxic effects of diisopropyl methylphosphonate have been identified. At high doses, the RBC is an important target of diisopropyl methylphosphonate in animals. Therefore, persons with more fragile blood cells, possibly those at high altitudes, or individuals with sickle cell anemia may be at greater risk for the hematological effects of diisopropyl methylphosphonate following exposure at high doses. Persons with neuromuscular diseases such as myasthenia gravis or other degenerative conditions may also be at greater risk based on the neurological effects observed in animals.

## 2.9 METHODS FOR REDUCING TOXIC EFFECTS

This section will describe clinical practice and research concerning methods for reducing toxic effects of exposure to diisopropyl methylphosphonate. 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 diisopropyl methylphosphonate. 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 chemical substances. Bronstein and Currance 1988; Ellenhorn and Barceloux 1988; Ellenhorn and Barceloux 1997; Stutz and Ulin 1992. However, exposure to diisopropylmethyl phosphonate is rare; therefore, these texts do not provide information regarding overexposure that is specific to this chemical, but do provide generalized information on procedures to be followed after various types of exposure.

### 2.9.1 Reducing Peak Absorption Following Exposure

No studies were located for reducing absorption in humans or animals exposed to diisopropyl methylphosphonate. Standard methods such as cathartics or activated carbon could be used. However, exposure would have to be identified within 4-6 hours since diisopropyl methylphosphonate is rapidly absorbed from the gastrointestinal tract (Ellenhorn and Barceloux 1988).

Common methods for reducing dermal absorption of diisopropyl methylphosphonate include removing contaminated clothes and washing contacted skin with soap and water (Ellenhorn and Barceloux 1988). Following eye contact with diisopropyl methylphosphonate, eyes should be flushed with copious amounts of

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water. In rabbits, rinsing the eyes immediately after direct exposure to diisopropyl methylphosphonate has been shown to reduce effects (Hart 1976).

### 2.9.2 Reducing Body Burden

In studies of mice, rats, and dogs, diisopropyl methylphosphonate was rapidly absorbed into plasma (Hart 1976). The plasma data indicate that all three species rapidly absorbed diisopropyl methylphosphonate, although the exact rate was species specific. Although no studies were located regarding human absorption, diisopropyl methylphosphonate is also likely to be absorbed rapidly into the plasma of humans. The ability of porous polymeric sorbents, activated carbon, and dialysis to remove diisopropyl methylphosphonate from human plasma has been studied (McPhillips 1983). The grafted butyl-XAD-4 was found to be the most efficient sorbent for the removal of diisopropyl methylphosphonate from human plasma. Hemoperfusion of plasma over synthetic XAD-4 or butyl-XAD-4 sorbent resin was more efficient than dialysis/ultrafiltration for the removal of diisopropyl methylphosphonate from human plasma; the smaller surface of the packed resins provided less area to minimize damage to molecular constituents of the plasma. These methods are useful in reducing diisopropyl methylphosphonate concentrations in the plasma. However, since diisopropyl methylphosphonate and its metabolites are not retained by the body, the need for methods to reduce body burden is uncertain.

### 2.9.3 Interfering with the Mechanism of Action for Toxic Effects

The mechanism of the toxic action of diisopropyl methylphosphonate is unclear. Therefore, therapies that could interfere with its mechanism are unknown.

## 2.10 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 diisopropyl methylphosphonate 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 diisopropyl methylphosphonate.

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.

### **2.10.1 Existing Information on Health Effects of Diisopropyl Methylphosphonate**

The existing data on health effects of inhalation, oral, and dermal exposure of humans and animals to diisopropyl methylphosphonate are summarized in Figure 2-3. The purpose of this figure is to illustrate the existing information concerning the health effects of diisopropyl methylphosphonate. 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* (ATSDR 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.

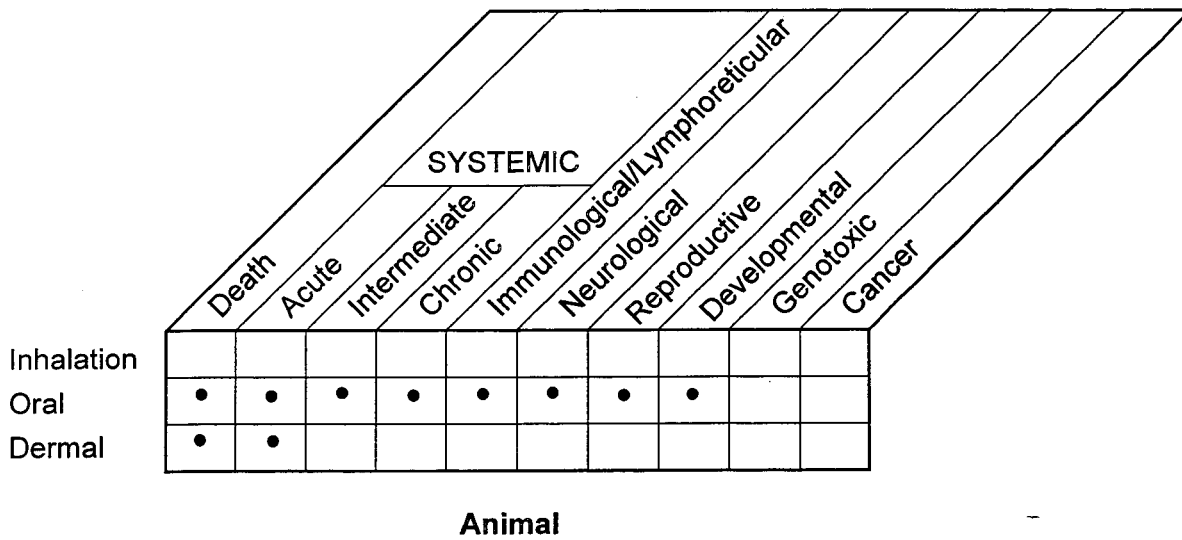
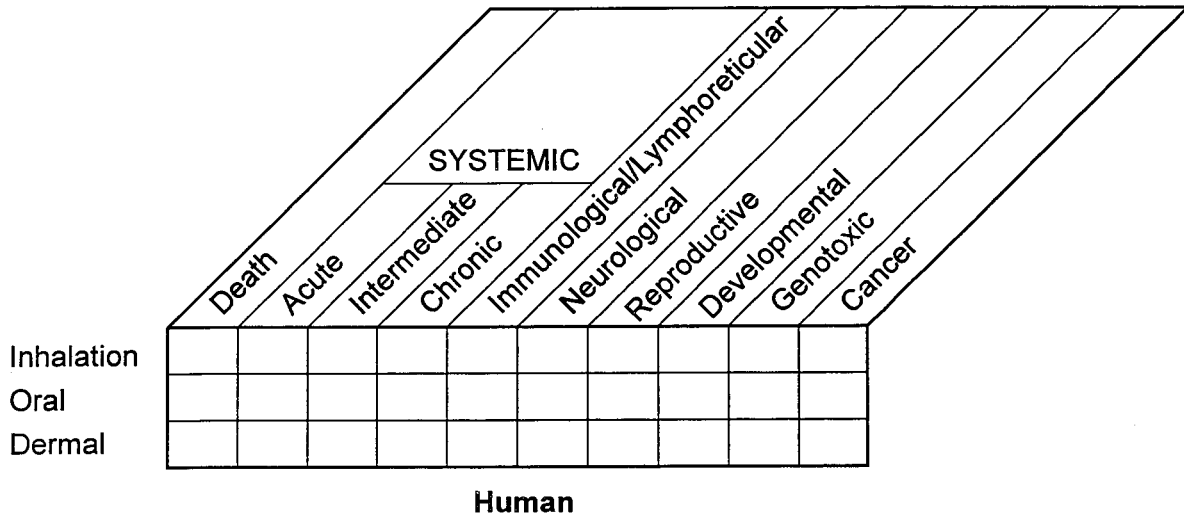
No studies were found on the health effects of diisopropyl methylphosphonate in humans following inhalation or oral exposure. The results of the one study that was located in which humans were exposed dermally to diisopropyl methylphosphonate was confounded by concurrent exposure to other chemicals. Limited animal data are available on the health effects of diisopropyl methylphosphonate following oral and dermal exposures.

### **2.10.2 Identification of Data Needs**

The following are topical sections that identify gaps in the present state of knowledge concerning the toxicology of diisopropyl methylphosphonate. Each of the sections identifies specific areas in which additional data may be helpful to gain a greater understanding of the toxicity of diisopropyl methylphosphonate as well as of the biochemical mechanisms of toxicity.

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**FIGURE 2-3. Existing Information on Health Effects of Diisopropyl Methylphosphonate**



• Existing Studies

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**Acute-Duration Exposure.** Acute oral LD<sub>50</sub> data are available for mice and rats (Hart 1976) and for ducks (Aulerich et al. 1979). Acute oral toxicity studies, including histopathological observations, are available for ducks, mice, rats, dogs, and mink (Aulerich et al. 1979; Hardisty et al. 1977; Hart 1976, 1980). Limited acute dermal toxicity are available for rats (Hart 1976). These data suggest a relatively low toxicity. However, a clear relationship between dose and effect has not been elucidated. Inhalation data of any kind were not identified, and dermal data were very limited.

**Intermediate-Duration Exposure.** No human or animal data were located concerning the toxicity of diisopropyl methylphosphonate after exposure via inhalation. Animal data are available for intermediate exposures by ingestion. Aulerich et al. (1979) reported deaths in mink treated with diisopropyl methylphosphonate. However, it is not clear if the deaths were truly treatment related. A more recent study by Bucci et al. (1997) did not find any treatment-related death in mink due to diisopropyl methylphosphonate after intermediate-duration exposure. Bucci et al (1992, 1994) found effects consistent with shortened RBC survival in mink treated with diisopropyl methylphosphonate in the diet at 747 mg/kg/day for males and 908 mg/kg/day for females, with no significant effects at lower doses. Plasma cholinesterase activity was significantly decreased at 345 mg/kg/day for males and 455 mg/kg/day for females, but not at lower doses. No change in RBC cholinesterase activity was observed. A change in plasma but not RBC cholinesterase is not considered adverse. Decreased weight gain was observed in ducks fed diisopropyl methylphosphonate at high doses. Additional intermediate-duration studies in rats and dogs did not find evidence of toxicity (Hardisty et al. 1977; Hart 1976, 1980), and a NOAEL in a dog study was used to derive an intermediateduration oral MRL (Hart 1980).

**Chronic-Duration Exposure and Cancer.** No studies were located regarding the health effects of chronic-duration exposure to diisopropyl methylphosphonate by inhalation or dermal route of exposure. There was no treatment-related lethality, changes in body weight, or other effects observed in mink fed 57 mg/kg/day diisopropyl methylphosphonate compared to controls for up to 13 months in a two-generation reproductive study (Bucci et al. 1997) although effects were observed at higher doses. However, F<sub>0</sub> females consumed almost 50% more feed than F<sub>1</sub> females, which was not completely explained by the study authors. This NOAEL was used to derive an chronic-duration oral MRL. Additional data are needed regarding all routes of exposures in various species to determine what, if any, toxicity exists. Future studies should focus on the understanding of the toxicity and the mechanism of toxicity of diisopropyl methylphosphonate. The design and protocol should be to study DIMP toxicity in an integrated manner, not to focus on a single

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endpoint. In addition, better understanding of the metabolism of diisopropyl methylphosphonate in longterm studies will also be helpful in elucidating its mechanism of action.

**Genotoxicity.** Several *in vitro* (DOD 1991c, 1991d, 1991e, 1991h; Hart 1980) and *in vivo* (DOD 1991a, 1991b, 1991f, 1991g) studies addressed the genotoxicity of diisopropyl methylphosphonate. Further *in vivo* studies may be useful to clarify any questions concerning the genotoxicity of the compound since equivocal results were sometimes observed.

**Reproductive Toxicity.** Significant amounts of work have been conducted in the reproductive toxicity of diisopropyl methylphosphonate (Aulerich et al. 1979; Bucci et al. 1997; Hart 1980). It seems fairly clear that diisopropyl methylphosphonate does not have reproductive effects, at least in the range of the doses tested. In addition, as discussed in Section 2.2.2.1, there is some confusion regarding the actual doses that the animals were exposed to in Hardisty et al. (1977). In addition, in the two-generation reproductive study by Bucci et al. (1997), an increase in ovarian follicles was found in high-dose F<sub>1</sub> animals compared to controls. The biological significance of these data are not clear, since there was no effect observed on breeding outcome. Nonetheless, further study is needed to determine whether this effect represents disrupted follicle maturation and to identify a NOAEL/LOAEL for this end point.

**Developmental Toxicity.** A single rat study, which found no adverse effects, addressed the developmental effects of diisopropyl methylphosphonate (Hart 1980). Additional data that utilize a greater range of doses in a few different species may be helpful.

**Immunotoxicity.** Limited data that addressed the immunological and lymphoreticular effects were available. No adverse responses were noted in the spleen of male or female rats (histopathological examination) (Army 1978) or in the spleen of mink (spleen weight, gross and histopathological examination) (Aulerich et al. 1979; Bucci et al. 1994, 1997). Additional data concerning various exposure pathways and a range of doses may be useful to corroborate the negative findings.

**Neurotoxicity.** The primary neurological concern with organophosphorous compounds is the potential for acetylcholinesterase inhibition. Plasma and erythrocyte cholinesterase measurements have been made in rats and mink (Bucci et al. 1992, 1994, 1997; Hart 1976, 1980). Even when large decreases (61%) in plasma cholinesterase were demonstrated, there were no decreases in erythrocyte cholinesterase nor were there any clinical signs of acetylcholinesterase inhibition. The data strongly suggest that diisopropyl methylphos-

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phosphate is not an inhibitor of acetylcholinesterase. Additional chronic studies in which higher doses are used may be useful in determining the toxicological importance of long-term plasma cholinesterase depression. Measurements of acetylcholinesterase at lethal doses of diisopropyl methylphosphonate may also illuminate its mechanism of action. In addition, quantitation of the distribution of DIMP and its metabolites (IMPA and MPA) among different brain areas and its correlation with the degree of cholinesterase inhibition found in a given brain area may help to understand its mechanism of action.

**Epidemiological and Human Dosimetry Studies.** No epidemiological studies were located.

However, prior to conducting any large-scale studies, animal studies are needed to determine what, if any, chronic health effects occur after exposure to diisopropyl methylphosphonate. In addition, appropriate cohorts would be very difficult to identify.

**Biomarkers of Exposure and Effect**

**Exposure.** Few studies were found regarding the measurement of diisopropyl methylphosphonate or its metabolites as indicators of exposure. IMPA in urine or plasma has been suggested as a biomarker of acute exposure. It would be useful to more fully explore urinary excretion of IMPA to determine dose relationships and its utility as a bioindicator of diisopropyl methylphosphonate exposure.

**Effect.** No biomarkers have been identified to characterize the effects associated with exposure to diisopropyl methylphosphonate. Toxicity data for diisopropyl methylphosphonate are limited. No specific clinical signs or symptoms in humans have been positively linked to diisopropyl methylphosphonate exposure. Further research is needed to identify the mechanism of neurological effects and the clinical symptoms of diisopropyl methylphosphonate exposure.

**Absorption, Distribution, Metabolism, and Excretion.** There are no data available on the absorption, distribution, metabolism, or excretion of diisopropyl methylphosphonate in humans. Limited animal data suggest that diisopropyl methylphosphonate is absorbed following oral and dermal exposure. Fat tissues do not appear to concentrate diisopropyl methylphosphonate or its metabolites to any significant extent. Nearly complete metabolism of diisopropyl methylphosphonate can be inferred based on the identification and quantification of its urinary metabolites; however, at high doses the metabolism of diisopropyl methylphosphonate appears to be saturated. Animal studies have indicated that the urine is the principal excretory route for removal of diisopropyl methylphosphonate after oral and dermal administration.

## 2. HEALTH EFFECTS

Because in most of the animal toxicity studies administration of diisopropyl methylphosphonate is in food, a pharmacokinetic study with the compound in food would be especially useful. It could help determine if the metabolism of diisopropyl methylphosphonate becomes saturated when given in the diet and if the levels of saturation are similar to those that result in significant adverse effects.

**Comparative Toxicokinetics.** There are no data on the kinetics of diisopropyl methylphosphonate in humans. Studies in animals suggest that metabolism and urinary metabolite profiles are qualitatively similar among species. Additional studies would be useful in understanding the differences in metabolic rates in species and in determining which animal species is the most appropriate model for human exposure.

**Interactions With Other Substances.** Little information is available regarding the interactions of diisopropyl methylphosphonate with other substances. As diisopropyl methylphosphonate has been identified at the RMA in the presence of many other chemicals, further studies of mixed chemical interactions should be done using chemicals found at the RMA to determine if the response to diisopropyl methylphosphonate is altered after multichemical exposure. Although the synergistic action of diisopropyl methylphosphonate and multiple pesticides has been suggested by one study (NIOSH 1981), the potential interaction of this substance with commonly-used antidotes for organophosphate poisoning (atropine and pralidoxime) might be explored. Subtle differences in the structure of organophosphates can result in different effects as well as different interactions with antidotes.

**Methods for Reducing Toxic Effects.** Little information is available regarding reducing the toxic effects of diisopropyl methylphosphonate following exposure. Recommended treatments include general hygienic procedures for rapid decontamination. The ability of porous polymeric sorbents, activated carbon, and dialysis to remove diisopropyl methylphosphonate from human plasma has been studied. However, since diisopropyl methylphosphonate and its metabolites are not retained by the body, the need for methods to reduce body burden is uncertain.

### 2.10.3 On-going Studies

No on-going studies on diisopropyl methylphosphonate were located.



### **3. CHEMICAL AND PHYSICAL INFORMATION**

#### **3.1 CHEMICAL IDENTITY**

Information regarding the chemical identity of diisopropyl methylphosphonate is located in Table 3-1.

#### **3.2 PHYSICAL AND CHEMICAL PROPERTIES**

Information regarding the physical and chemical properties of diisopropyl methylphosphonate is located in Table 3-2.

## 3. CHEMICAL AND PHYSICAL INFORMATION

**TABLE 3-1. Chemical Identity of Diisopropyl Methylphosphonate**

Characteristic	Information	Reference
Chemical name	Diisopropyl methylphosphonate	EPA 1989
Synonym(s)	DIMP; diisopropyl methanephosphonate; phosphonic acid; methyl-, bis-(1-methyl-ethyl) ester; phosphonic acid, methyl-diisopropyl ester	Dacre and Rosenblatt 1987
Registered trade name(s)	Not a commercial substance	
Chemical formula	C <sub>7</sub> H <sub>17</sub> O <sub>3</sub> P	Dacre and Rosenblatt 1987
Chemical structure	$  \begin{array}{ccccc}  \text{H}_3\text{C} & & \text{O} & & \text{CH}_3 \\  & \backslash &    & / & \\  & \text{CH-O-P-O-CH} & & & \\  & / &   & \backslash & \\  \text{H}_3\text{C} & & \text{CH}_3 & & \text{CH}_3  \end{array}  $	Dacre and Rosenblatt 1987
Identification numbers:		
CAS registry	1445-75-6	Dacre and Rosenblatt 1987
NIOSH RTECS	SZ9090000	Dacre and Rosenblatt 1987
EPA hazardous waste	No data	
OHM/TADS	No data	
DOT/UN/NA/IMCO shipping	No data	
HSDB	6864	HSDB 1994
NCI	No data	

CAS = Chemical Abstracts Services; DOT/UN/NA/IMCO = Department of Transportation/United Nations/North America/International Maritime Dangerous Goods Code; EPA = Environmental Protection Agency; HSDB = Hazardous Substances Data Bank; NCI = National Cancer Institute; NIOSH = National Institute for Occupational Safety and Health; OHM/TADS = Oil and Hazardous Materials/Technical Assistance Data System; RTECS = Registry of Toxic Effects of Chemicals Substances

## 3. CHEMICAL AND PHYSICAL INFORMATION

**TABLE 3-2. Physical and Chemical Properties of Diisopropyl Methylphosphonate**

Property	Information	Reference
Molecular weight	180.18	Dacre and Rosenblatt 1987
	180.21	HSDB 1994
Color	Colorless	EPA 1989
Physical state	Liquid	EPA 1989
Melting point	No data	
Boiling point	174°C	EPA 1989
Density:		
At 25°C	0.976 g/mL	EPA 1989
Odor	No data	
Odor threshold	No data	
Solubility:		
Water at 0°C	80 g/L	Bucci et al. 1997
Water at 25°C	1-2 g/L	Dacre and Rosenblatt 1987
Water at 80°C	11 g/L	Dacre and Rosenblatt 1987
Partition coefficients:		
Log K <sub>ow</sub>	No data	
Log K <sub>oc</sub>	No data	
Vapor pressure:		
Temperature not specified	10 <sup>-4</sup> -1 mmHg	Rosenblatt et al. 1975a, 1975b
At 10°C	13 (estimated)	Dacre and Rosenblatt 1987
At 100°C	34 (estimated)	Dacre and Rosenblatt 1987
Henry's law constant	No data	
Autoignition temperature	No data	
Flashpoint	No data	
Flammability limits	No data	
Conversion factors	No data	
Explosive limits	No data	



## 4. PRODUCTION, IMPORT, USE, AND DISPOSAL

### 4.1 PRODUCTION

Diisopropyl methylphosphonate is an organophosphate compound that was first produced in the United States as a by-product of the manufacture of the nerve gas isopropyl methylphosphonofluoridate (GB, or Sarin) (ATSDR 1996; EPA 1989; Robson 1977, 1981). It is not a nerve gas and is not a metabolite or degradation product (Roberts et al. 1995). Diisopropyl methylphosphonate constitutes approximately 2-3% of the crude GB product, but it is neither a metabolite nor a degradation product of GB (EPA 1989; Rosenblatt et al. 1975b). Diisopropyl methylphosphonate is not normally produced except for its use in research. One method of producing diisopropyl methylphosphonate is to combine triisopropyl phosphite and methyl iodide. The mixture is then boiled, refluxed, and distilled, yielding diisopropyl methylphosphonate and isopropyl iodide (Ford-Moore and Perry 1951). Diisopropyl methylphosphonate may also be prepared from sodium isopropyl methylphosphonate by a reaction at 270° C, but a portion of the resulting diisopropyl methylphosphonate is converted to trimethylphosphine oxide at this temperature (EPA 1989).

The only actual production of the diisopropyl methylphosphonate-containing GB nerve gas occurred during the years 1953-1957 at the RMA near Denver, Colorado. Beginning in 1957, the RMA became a storage site for unused chemical warfare agents. Then, beginning in 1973, the GB manufacturing facility was used to detoxify the stored GB nerve agent (Robson 1977, 1981). Diisopropyl methylphosphonate and other industrial effluents have been released at the RMA as a result of both the manufacturing and detoxification activities conducted at the site (ATSDR 1996; Robson 1977, 1981). Actual volumes of diisopropyl methylphosphonate at the RMA from GB manufacture and detoxification are unknown. No chemical agents are currently stored at the RMA (EPA 1989). Diisopropyl methylphosphonate is not known to occur naturally in the environment, and it does not appear to have any current uses that would result in its further production (EPA 1989).

### 4.2 IMPORT/EXPORT

No information could be found regarding import or export volumes of diisopropyl methylphosphonate.

## 4. PRODUCTION, IMPORT/EXPORT, USE, AND DISPOSAL

**4.3 USE**

There are no known commercial uses of diisopropyl methylphosphonate. The military has used diisopropyl methylphosphonate as a chemical simulant in training and in experiments to determine the toxicity, persistence, and fate of GB nerve gas in the environment (Van Voris et al. 1987) and the movement and permeation of GB nerve gas through porous media and protective clothing (Jenkins et al. 1994; Pal et al. 1993; Vo-Dinh and Pal 1992). Simulants such as diisopropyl methylphosphonate were used because they possess some properties which resemble those of more hazardous compounds. The simulants can be used to mimic and study specific attributes of the more hazardous compounds when training personnel and developing chemical defense material (Van Voris et al. 1987). Because diisopropyl methylphosphonate does not appear to be a cholinesterase inhibitor, it was used as a simulant for the more toxic GB (Sarin) nerve agent. No information was found concerning the approximate amounts of diisopropyl methylphosphonate used as a simulant.

**4.4 DISPOSAL**

The Department of Defense Authorization Act of 1986 (Public Law 99-145) directed the Secretary of Defense to destroy all lethal unitary chemical munitions and agents by September 30, 1994, including organophosphate nerve agents such as GB. The act was amended in 1988 (Public Law 100-456) to allow for complete disposal of chemical munitions by April 1997.

One method for disposing of diisopropyl methylphosphonate is microwave decomposition, in which nearly 100% decomposition of diisopropyl methylphosphonate is obtained in a microwave plasma discharge (Bailin et al. 1975). Another method for disposing of diisopropyl methylphosphonate is ultraviolet and infrared laser-induced photodestruction, resulting in the production of simple hydrocarbon gases in a phosphorous-bearing acid (Radziemski 1981). Chemical oxidation coupled with ultraviolet radiation catalyzation can be used to successfully remove diisopropyl methylphosphonate from industrial effluents (Zappi et al. 1990). Granular activated carbon has been shown to significantly reduce the concentration of diisopropyl methylphosphonate in contaminated groundwater (Calgon 1977).

Diisopropyl methylphosphonate has not been shown to be amenable to biodegradation. Little if any degradation by indigenous bacteria occurred in soil "dosed" with radiolabelled diisopropyl methylphosphonate (Williams et al. 1989). Biodegradation by aquatic microorganisms has been shown to be

## 4. PRODUCTION, IMPORT/EXPORT, USE, AND DISPOSAL

equally ineffective (Spangord et al. 1979; Van Voris et al. 1987), rendering it inappropriate as a means for the disposal of diisopropyl methylphosphonate.





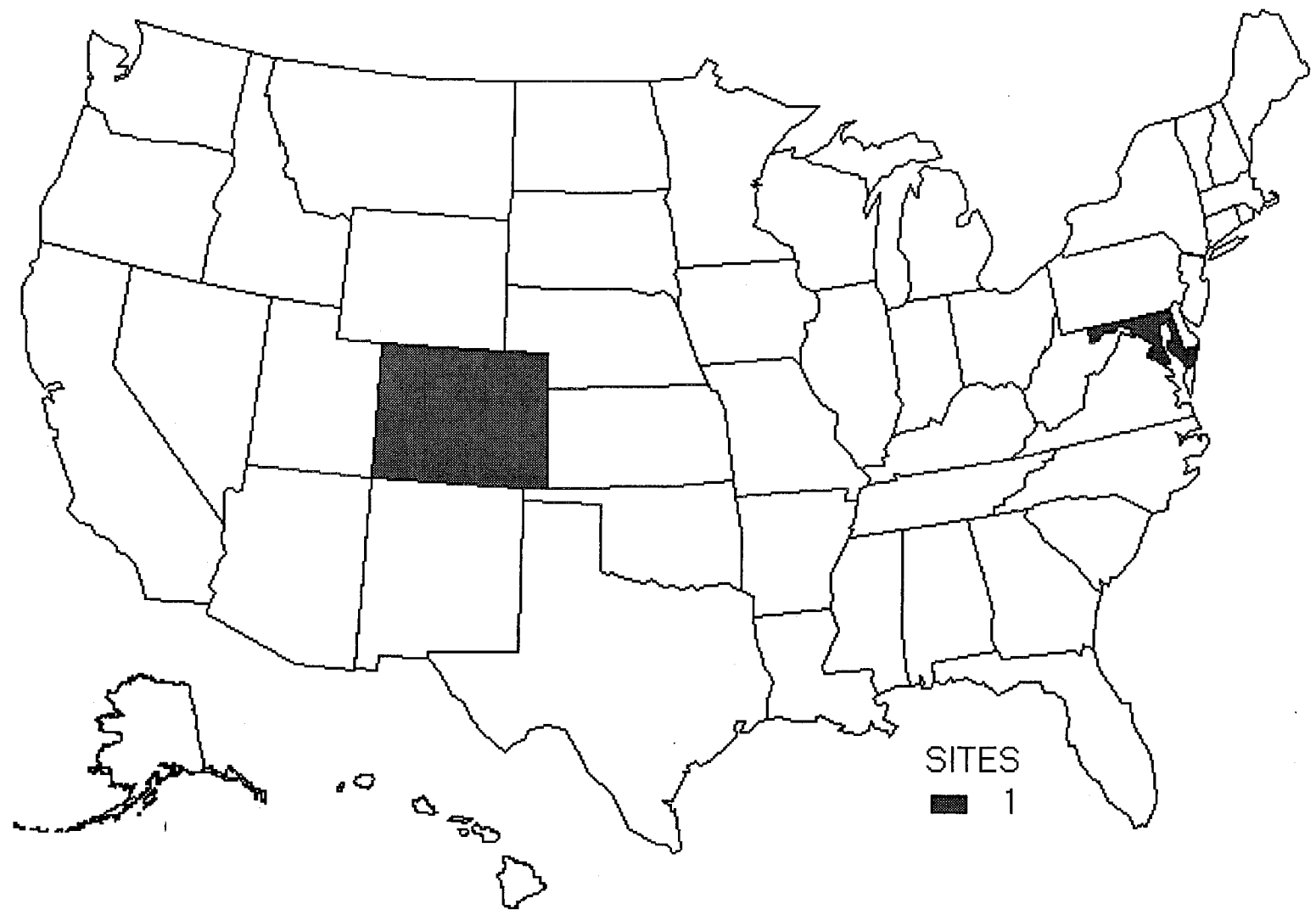
## 5. POTENTIAL FOR HUMAN EXPOSURE

### 5.1 OVERVIEW

Diisopropyl methylphosphonate has been identified at 2 of the 1,416 hazardous waste sites that have been proposed for inclusion on the EPA National Priorities List (NPL) (HazDat 1997). One site, identified in Figure 5-1, is the RMA located near Denver, Colorado. As diisopropyl methylphosphonate was never made commercially, the EPA neither expects nor routinely looks for this chemical at hazardous waste sites unless site history indicates it might be present.

Diisopropyl methylphosphonate was released at the RMA near Denver, Colorado, during 1953-1957 as a byproduct in the effluent from the manufacture of the nerve agent GB (Sarin). It is not a nerve gas and is not a metabolite or degradation product (Roberts et al. 1995). Contamination was confined primarily to the groundwater and, to a lesser extent, surface water and soil. Because of its low volatility, diisopropyl methylphosphonate is not likely to contaminate the atmosphere, and diisopropyl methylphosphonate does not undergo any significant photolysis (direct or indirect) in water. Biotransformation of diisopropyl methylphosphonate has not been detected in natural waters. Biotransformation of diisopropyl methylphosphonate is very slow in soil and is temperature dependent. Diisopropyl methylphosphonate is relatively mobile in soil. The presence of a nonreactive carbon-phosphorous (C-P) bond contributes to the environmental persistence of diisopropyl methylphosphonate. Bioconcentration has been demonstrated in plants, primarily in leaves. Consequently, it is possible that diisopropyl methylphosphonate may enter the food chain via animal feed. Exposure of the general population to diisopropyl methylphosphonate is expected to be low. If exposure does occur, it is most likely to arise from drinking or showering with diisopropyl methylphosphonate-contaminated water at or near the RMA, or through ingestion of fruits and vegetables that have been irrigated with such contaminated water.

Figure 5-1. Frequency of NPL Sites With Diisopropyl Methylphosphonate Contamination\*



\*Derived from HazDat 1998

## 5.2 RELEASES TO THE ENVIRONMENT

Releases of diisopropyl methylphosphonate are not required to be reported under SARA Section 313; consequently, there are no data for these compounds in the 1995 Toxics Release Inventory (TRI) (TRI95 1997).

### 5.2.1 Air

No specific information was found on the releases of diisopropyl methylphosphonate to air. However, diisopropyl methylphosphonate is unlikely to be an atmospheric contaminant because of its very low volatility (Rosenblatt et al. 1975b). There is the possibility that exposure to airborne diisopropyl methylphosphonate could occur if the compound was sorbed to soil and subsequently entrained by wind.

### 5.2.2 Water

Diisopropyl methylphosphonate was discharged in industrial effluent during the manufacture of the nerve agent GB (Sarin) during 1952-1957 at the RMA (EPA 1989; Robson 1977). Prior to 1956, the effluent from the production of Sarin was disposed of in five unlined surface ponds. Effluent percolated through the bottom of these ponds, entered the groundwater in a shallow alluvial aquifer, and resulted in groundwater contamination. In 1974, chemical analyses of the groundwater revealed the presence of diisopropyl methylphosphonate in a 73-km<sup>2</sup> area to the northwest of the arsenal and within 1.6 km (1 mile) of municipal water wells supplying the city of Brighton, Colorado (Robson 1977, 1981). The concentration of diisopropyl methylphosphonate in the groundwater ranged from 0.5 µg/L (the analytical detection limit) to 44,000 µg/L near one of the abandoned waste disposal ponds.

Groundwater samples collected over the last 10 years from monitoring and private water wells in the offpost area of RMA have provided data regarding the extent of diisopropyl methylphosphonate. The data indicate that diisopropyl methylphosphonate in the alluvial aquifer extends from the RMA northwest to the South Platte River. In the 1995 sampling event, diisopropyl methylphosphonate concentrations ranged from less than 0.392 µg/L, the detection limit, to just over 600 µg/L in this area. The highest reported concentrations were present upstream of the Peoria Street Treatment Plant. The lowest or non-detect concentrations are

## 5. POTENTIAL FOR HUMAN EXPOSURE

closer to the South Platte River. The treatment facility was installed in 1993 to intercept the contaminated groundwater north of RMA.

In 1996, chemical analyses of groundwater revealed the presence of DIMP in a 19.4 km<sup>2</sup> area located north and northwest of RMA, and within 11 km of South Adams County Water and Sanitation District (SACWSD) municipal water wells supplying the city of Commerce City (Harding Lawson Associates 1996).

DIMP contamination levels in the groundwater of Adams County around the RMA are decreasing due to treatment by the Army. The highest DIMP level in a well in 1997 was 1,500 µg/L. This well is in the vicinity of the offpost groundwater treatment system, operated by the Army, and will be intercepted and treated by the system. Downstream of the system, the highest diisopropyl methylphosphonate level was 660 µg/L in 1997. The Onpost Record of Decision in June 1996 provided that well owners with wells contaminated above 0.392 ppb could be connected to a municipal water supply. All well owners that accepted will be connected to the municipal supply by the summer of 1998. The Record of Decision also provides for wells to be connected in the future if diisopropyl methylphosphonate concentrations exceed values promulgated in the Colorado Basic Standards for Groundwater. Offpost surface water studies have not been conducted in 1998. In 1997, surface water near the RMA contained approximately less than the analytical reporting limit of 0.2 µg/L to 7.7 µg/L diisopropyl methylphosphonate.

### 5.2.3 Soil

No specific information was found on the releases of diisopropyl methylphosphonate to soil. However, diisopropyl methylphosphonate has been detected in soils at the RMA (Williams et al. 1989). Concentrations of diisopropyl methylphosphonate in five surface soil samples at the RMA ranged from <0.05 to 0.24 mg/kg (Williams et al. 1989). Concentrations of diisopropyl methylphosphonate in surface soil samples measured during the Remedial Investigation/Feasibility Study (RI/FS) at RMA ranged from less than the analytical limit of 0.114 mg/kg to 11 mg/kg (Ebasco Services 1991). The most recent sampling conducted in 1995 indicated the concentrations of diisopropyl methylphosphonate in the onpost surface soil samples were less than the analytical reporting limit of 0.342 mg/kg (D.P. Associates 1995).

### 5.3 ENVIRONMENTAL FATE

#### 5.3.1 Transport and Partitioning

Diisopropyl methylphosphonate is slightly soluble in water (0.1-0.3 g/L at 25°) and has been demonstrated in laboratory studies to quickly diffuse between the surface microlayer into the water column after deposition as aerosols on fresh water (Van Voris et al. 1987). The solubility of diisopropyl methylphosphonate was 80 g/L (8%) and remained in solution even when the temperature was lowered to freezing (Bucci et al. 1997). In addition, there was no significant loss of diisopropyl methylphosphonate from the water column to the atmosphere, in either the presence or absence of a light wind over the water surface. Human exposure resulting from the vaporization of diisopropyl methylphosphonate from surface water is considered insignificant (EPA 1989).

Laboratory studies on the deposition of diisopropyl methylphosphonate to soils suggest that soil acts as a sink for this chemical (Van Voris et al. 1987). Deposition velocities of diisopropyl methylphosphonate for soil were approximately 0.87-1.2 cm/second when mass loading rates ranged from 300 to 3,000  $\mu\text{g}/\text{cm}^2$ . Results of soil lysimeter studies indicate that the movement of diisopropyl methylphosphonate in soil occurs with irrigation water flow (O'Donovan and Woodward 1977a, 1977b). Moreover, the movement of diisopropyl methylphosphonate through various types of soil depends on the volume of irrigation water applied to their surfaces. Radioactive tracer experiments indicate that evaporation of diisopropyl methylphosphonate from dry soil is not significant (O'Donovan and Woodward 1977a, 1977b). Dry soils containing diisopropyl methylphosphonate retained over 95% of the initial radioactivity after approximately 250 hours of treatment. The moist soil samples retained somewhat less activity (approximately 78%).

Bioconcentration of diisopropyl methylphosphonate by various plants was studied in soil and in hydroponic cultures (O'Donovan and Woodward 1977a, 1977b). The hydroponic studies used corn, beans, radish, wheat, tomato, carrot, sugar beet, meadow fescue, rose, and juniper. The soil studies included carrot, wheat, alfalfa, sugar beet, and bean. The concentrations of diisopropyl methylphosphonate in the irrigation water used in the soil tests were 0, 1, 8, 20, 50, 100, 300, 500, 700, and 1,000 ppm. Hydroponic cultures contained 0, 1, 10, 100, and 1,000 ppm diisopropyl methylphosphonate. Results of the hydroponic studies demonstrated that bioconcentration occurred in most plants, with peak bioconcentration occurring primarily in the 1st month of growth. In general, the leaves of the plants displayed the highest concentration of diisopropyl methylphosphonate, which was up to 20 times greater than concentrations contained in the solutions. Other

plant parts typically did not concentrate diisopropyl methylphosphonate or did so in a very limited manner. Bioconcentration also occurred in plants when diisopropyl methylphosphonate was added to soil. When measured at 65 days after exposure, the concentration of diisopropyl methylphosphonate in plant tissues (in particular, the leaves) generally increased with increasing diisopropyl methylphosphonate concentrations in the irrigation solution.

### **5.3.2 Transformation and Degradation**

#### **5.3.2.1 Air**

No information was found on the transformation of diisopropyl methylphosphonate in the atmosphere. Based on the results of environmental fate studies of diisopropyl methylphosphonate in distilled water and natural water, photolysis (either direct or indirect) is not important in the transformation of diisopropyl methylphosphonate in aquatic systems (Spanggard et al. 1979). The ultraviolet and infrared laser-induced photodegradation of diisopropyl methylphosphonate in both the vapor or liquid phase has been demonstrated (Radziemski 1981). Light hydrocarbon gases were the principal decomposition products. Hydrogen, carbon monoxide (CO), carbon dioxide (CO<sub>2</sub>), and water were also detected.

#### **5.3.2.2 Water**

Diisopropyl methylphosphonate does not undergo direct or indirect photolysis in aquatic systems, as demonstrated by the stability of the compound in distilled water or in a natural water sample after 232 hours of reaction time with the mercury lamp filtered to exclude all wavelengths below 290 nm (Spanggard et al. 1979).

Biodegradation of diisopropyl methylphosphonate was not observed in acclimated cultures of bog water obtained from the RMA, even after the addition of extra carbon sources and an incubation period of up to 12 weeks (Spanggard et al. 1979). In addition, no biodegradation of diisopropyl methylphosphonate was observed in waters obtained from the Palo Alto sewage plant aeration tank or from a pond near Searsville Lake in Woodside, California.

### 5.3.2.3 Sediment and Soil

The biodegradation of diisopropyl methylphosphonate by microorganisms was studied in soil obtained from the RMA (Spanggord et al. 1979). Diisopropyl methylphosphonate was degraded to CO<sub>2</sub> by soil microorganisms, but the biodegradation was very slow, with only 13.4% of the original activity being evolved as <sup>14</sup>CO<sub>2</sub> after 34 weeks of incubation. It was determined that soil biodegradation was not a function of the concentration of diisopropyl methylphosphonate. However, temperature can affect the soil biodegradation of diisopropyl methylphosphonate with transformation being very slow at 25° C (15-20%) and almost completely halted at 10° C (<0.1 %) (Spanggord et al. 1979). A half-life in excess of 2 years was predicted for diisopropyl methylphosphonate in soil. In another study of the microbial degradation of diisopropyl methylphosphonate in soil, <sup>14</sup>C-diisopropyl methylphosphonate was converted to <sup>14</sup>CO<sub>2</sub>, to the extent of 6.4% and 6.0% in soils, uncontaminated and contaminated, respectively, obtained from the RMA (Williams et al. 1989). Results of a mass balance study revealed that the majority of the <sup>14</sup>C-diisopropyl methylphosphonate was recovered as the undegraded parent compound.

## 5.4 LEVELS MONITORED OR ESTIMATED IN THE ENVIRONMENT

### 5.4.1 Air

No information was found on levels monitored or estimated in the air. However, because of its very low volatility, diisopropyl methylphosphonate is unlikely to be an atmospheric contaminant (EPA 1989). A rate constant for diisopropyl methylphosphonate has not been determined.

### 5.4.2 Water

There are no measurements of the actual concentrations of diisopropyl methylphosphonate in groundwater at the RMA during the years of active production of the nerve gas Sarin (i.e., 1953-1957) (EPA 1989). The first actual measurements of the concentration of diisopropyl methylphosphonate in the groundwater on the arsenal and surrounding property to the north and west were made in 1974 (Robson 1981). The concentrations of diisopropyl methylphosphonate in the groundwater ranged from 0.5 µg/L (analytical detection limit) to as much as 44,000 µg/L near the abandoned waste disposal ponds. Diisopropyl methylphosphonate was discharged into a lined reservoir at the RMA in 1956 and was still present 20 years later in concentrations of about 400,000 µg/L (Robson 1977).

## 5. POTENTIAL FOR HUMAN EXPOSURE

During the most recent comprehensive groundwater sampling at RMA in 1995, diisopropyl methylphosphonate concentrations in groundwater were reported to range from less than 0.110 µg/L to 965 µg/L. The highest concentrations were reported near the areas of prior waste disposal. In 1997, offpost groundwater near the RMA contained less than the analytical reporting limit of 0.2 µg/L to 1,500 µg/L diisopropyl methylphosphonate (USGS 1998). In 1997, offpost surface water near the RMA contained less than the analytical reporting limit of 0.2 µg/L to 7.7 µg/L diisopropyl methylphosphonate.

In 1997, concentrations of diisopropyl methylphosphonate in onpost groundwater samples at the RMA ranged from less than the analytical reporting limit of 0.2 µg/L to 1,500 µg/L (USGS 1998). Concentrations of diisopropyl methylphosphonate in onpost surface water samples at the RMA ranged from less than the analytical reporting limit of 0.2 µg/L to 0.581 µg/L.

#### 5.4.3 Sediment and Soil

The concentrations of diisopropyl methylphosphonate in 5 surface soil samples obtained at the RMA ranged from <0.05 to 0.24 mg/kg (Williams et al. 1989).

The most recent onpost surface soil sampling was conducted in 1995. Concentrations of diisopropyl methylphosphonate in surface soil samples measured in 1995 at RMA were less than the analytical reporting limit of 0.342 mg/kg, (D.P. Associates 1995). The most recent onpost subsurface soil sampling occurred in 1993. Concentrations of Diisopropyl methylphosphonate in 1993 at RMA ranged from less than the analytical reporting limit of 0.114 mg/kg to 0.864 mg/kg (Harding Lawson Associates 1994).

The most recent onpost sediment sampling occurred in 1990. All concentrations of diisopropyl methylphosphonate in sediment samples measured during the RI at RMA were less than the analytical reporting limits, which ranged from 0.05 mg/kg to 1.0 mg/kg (Ebasco Services Inc. 1992). Diisopropyl methylphosphonate has not been detected in sediment samples near the RMA. The most recent sampling for diisopropyl methylphosphonate occurred in 1990 and all the results were less than the analytical reporting limit of 1.0 mg/kg (Harding Lawson Associates 1992).



## 5. POTENTIAL FOR HUMAN EXPOSURE

**5.4.4 Other Environmental Media**

No information was located regarding concentrations of diisopropyl methylphosphonate in other environmental media.

**5.5 GENERAL POPULATION AND OCCUPATIONAL EXPOSURE**

Exposure of the general population to diisopropyl methylphosphonate is expected to be highly unlikely and to occur at extremely low levels, but data are insufficient for exposure estimates. Diisopropyl methylphosphonate has been detected in the groundwater and, to a lesser extent, in the surface water and soil at or near the RMA. If exposure of the general population to diisopropyl methylphosphonate were to occur, water would be the most likely source.

Although workers in facilities manufacturing diisopropyl methylphosphonate may have been exposed to the chemical during the active years of production, no occupational monitoring studies for diisopropyl methylphosphonate were located.

**5.6 POPULATIONS WITH POTENTIALLY HIGH EXPOSURES**

Populations living near the RMA may be at risk of exposure to diisopropyl methylphosphonate. Individuals with diisopropyl methylphosphonate in their domestic wells are receiving their drinking water from alternative sources. According to the On-post Record of Decision (June 1996), well owners with wells contaminated above 0.392 µg/L of diisopropyl methylphosphonate could be connected to a municipal water supply.

**5.7 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 diisopropyl methylphosphonate is available. Where adequate information is not available, ATSDR, in conjunction with the 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 diisopropyl methylphosphonate.

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.

### 5.7.1 Identification of Data Needs

**Physical and Chemical Properties.** The physical and chemical properties of diisopropyl methylphosphonate are sufficiently well defined to allow assessment of its fate following release to the environment (EPA 1989; HSDB 1994; Robson 1981; Rosenblatt et al. 1975b). The solubility of diisopropyl methylphosphonate under various conditions is not well defined. No additional information is needed.

**Production, import/Export, Use, Release, and Disposal.** The risk for exposure of the general population to substantial levels of diisopropyl methylphosphonate is quite low. GB (Sarin) and diisopropyl methylphosphonate have not been produced in the United States since 1957, and there is no indication that U.S. production of these chemicals will resume (EPA 1989). No information exists regarding the import or export of diisopropyl methylphosphonate. Diisopropyl methylphosphonate has no known commercial uses, but has been used by the military as a simulant for chemical warfare agents (Van Voris et al. 1987).

Methods for disposing of diisopropyl methylphosphonate include microwave decomposition, ultraviolet and infrared laser-induced photodestruction, chemical oxidation coupled with ultraviolet radiation catalyzation, and adsorption using granular activated carbon (Bailin et al. 1975; Calgon 1977; Radziemski 1981; Zappi et al. 1990). However, no information is available regarding the amounts of diisopropyl methylphosphonate disposed of by each method.

**Environmental Fate.** The environmental fate of diisopropyl methylphosphonate has been investigated in several studies (O'Donovan and Woodward 1977a, B977b; Radziemski 1981; Robson 1977, 1981; Spanngord et al. 1979; Van Voris et al. 1987; Williams et al. 1989). This chemical is relatively unreactive and degrades slowly in environmental media. Diisopropyl methylphosphonate is a by-product of production at the RMA and is confined to the RMA and surrounding areas. Municipal water will be supplied to consenting drinking water well owners within the DIMP plume. Thus, potential exposure of the general

## 5. POTENTIAL FOR HUMAN EXPOSURE

population is low. Additional monitoring studies of the contaminated plumes will be useful to assess the transport of this chemical from the RMA.

**Bioavailability from Environmental Media.** Oral absorption of DIMP from water was found to be rapid in both the rats and minks (Bucci et al. 1992). Dermal absorption of DIMP in swine was reported to be between 3 and 7% through 7 days (Snodgrass and Metkler 1992). However, no studies were located regarding the bioavailability of diisopropyl methylphosphonate from other environmental media. Studies that investigate the bioavailability of diisopropyl methylphosphonate from soils would be useful in estimating exposure of persons who live near the RMA.

**Food Chain Bioaccumulation.** Bioconcentration of diisopropyl methylphosphonate occurs primarily in the leaves of plants (O'Donovan and Woodward 1977a, 1977b). However, DIMP also bioconcentrates in the edible root portions of radishes and carrots, and in the fruit of tomato plants at lower levels. Exposure may occur through the ingestion of fruits and vegetables that have been irrigated with DIMP contaminated water. Additional studies are needed to assess the potential for bioconcentration in plants. While it is possible that diisopropyl methylphosphonate may enter the food chain via animal feed, DIMP is rapidly changed to IMPA by animals that eat it. Therefore, it is unlikely that DIMP will be bioaccumulated in animals and be present further up the food chain.

**Exposure Levels in Environmental Media.** Monitoring data were not located for diisopropyl methylphosphonate in ambient air. This chemical has been detected in surface and groundwater receiving effluent from the RMA (ATSDR 1996). It has also been detected in soil at the RMA. Since this chemical is not expected to be prevalent in the environment and exposure of the general population is not expected to be of concern, monitoring of ambient environmental media does not appear to be required. However, monitoring of environmental media such as groundwater and soil in the vicinity of the RMA has been conducted to help determine the potential for exposure.

**Exposure Levels in Humans.** This information is necessary for assessing the need to conduct health studies on these populations. No occupational studies were located regarding exposure levels of diisopropyl methylphosphonate in humans. Workers conducting wildlife toxicity studies at the RMA reported effects that could be associated with exposure to diisopropyl methylphosphonate; however, the workers were also exposed to a number of other toxic chemicals. One study that attempted to determine levels of diisopropyl methylphosphonate or its metabolite IMPA in the urine of residents of communities adjacent to the RMA was

## 5. POTENTIAL FOR HUMAN EXPOSURE

unsuccessful because urine components may have reduced sensitivity to target analytes by producing interferences (ATSDR 1996). Additional monitoring of populations at the RMA or living in the vicinity of the RMA would help assess the effects of diisopropyl methylphosphonate on the general population.

**Exposure Registries.** No exposure registries for diisopropyl methylphosphonate were located. This substance is not currently one of the compounds for which a subregistry has been established in the National Exposure Registry.

### 5.7.2 On-going Studies

ATSDR, EPA, the state of Colorado, and Shelf Oil are developing a medical monitoring plan for the remediation of the RMA. Exactly what chemicals will be monitored has not yet been determined.

## 6. ANALYTICAL METHODS

The purpose of this chapter is to describe the analytical methods that are available for detecting, measuring, and/or monitoring diisopropyl methylphosphonate, its metabolites, and other biomarkers of exposure and effect to diisopropyl methylphosphonate. The intent is not to provide an exhaustive list of analytical methods. Rather, the intention is to identify well-established methods that are used as the standard methods of analysis. Many of the analytical methods used for environmental samples are the methods approved by federal agencies and organizations such as EPA and the National Institute for Occupational Safety and Health (NIOSH). Other methods presented in this chapter are those that are approved by groups such as the Association of Official Analytical Chemists (AOAC) and the American Public Health Association (APHA). Additionally, analytical methods may be included that modify previously used methods to obtain lower detection limits and/or to improve accuracy and precision.

### 6.1 BIOLOGICAL SAMPLES

Methods exist for measuring diisopropyl methylphosphonate and its metabolites in biological materials. Metabolites of diisopropyl methylphosphonate have been analyzed in plasma, urine, and fecal samples of mice and rats (Weiss et al. 1994). Cold acetonitrile is added to each sample, with fecal samples first being homogenized with distilled water. Each sample is then centrifuged, and the supernatant is separated and fractionated by reverse phase, high performance liquid chromatography and thin layer chromatography. Gas chromatography (GC), combined with detection by mass spectrometry (MS), and proton nuclear magnetic resonance are then used to identify the fractions (Weiss et al. 1994). Attempts to develop verifiable analytical techniques for the detection of IMPA, the major metabolite of diisopropyl methylphosphonate, were unsuccessful in one study (ATSDR 1996).

GC, utilizing flame ionization detection (FID), has been used to measure diisopropyl methylphosphonate in meat, grain, or milk (Caton et al. 1994). Sample preparation steps include homogenization, filtration, dialysis, and extraction on a solid sorbent. Two common solid phase extractants, Tenax GC and octadecylsilane bonded silica gel (C18 Silica), were compared by Caton et al. (1994). They reported 70% recovery when using Tenax GC and 85% recovery when using C18 Silica. Sensitivity was not reported. Equilibrium experiments indicate that 8-10 mg of Tenax GC are required to achieve maximum recovery of each  $\mu\text{g}$  of diisopropyl methylphosphonate (Caton et al. 1994). By extrapolating these results to a complex

## 6. ANALYTICAL METHODS

mixture sample in which several chemical species would be competing for sorption sites, the study authors estimated that 0.1-0.5 mg of Tenax GC should be used for each mg of plant or animal tissue. An alternative approach to measure diisopropyl methylphosphonate in meat or milk is based upon thermal desorption into an ion trap mass spectrometer (TDITMS) (Buchanan et al. 1995). Preparation of beef tissue samples requires combination of centrifugation, filtration, and absorption on solid sorbents. Milk samples did not require centrifugation and filtration. Overall recovery for diisopropyl methylphosphonate from the sorbent Tenax was found to be in the range of 20-50% (Buchanan et al. 1995). Small initial sample sizes are required to reach detection limits of 50-100 ppb, suggesting that it may be possible to extract small tissue samples from livestock by needle biopsy. This would eliminate the requirement of animal sacrifice for analysis. Further, the time needed for sample preparation and analysis using TDITMS is shorter than conventional EPA methods, allowing for a greater number of samples to be included in the analysis. A summary of the analytical methods for various biological media is presented in Table 6-1.

## 6.2 ENVIRONMENTAL SAMPLES

Methods exist for determining levels of diisopropyl methylphosphonate in air, soil, and water. These methods include separation by GC coupled with FID and flame photometric detection (FPD), determination by infrared and Raman spectroscopy, separation by ionization mass spectrometry, determination utilizing piezoelectric crystals, and determination by gas-sensitive microsensors. Table 6-2 summarizes the methods that have been used to analyze environmental samples for diisopropyl methylphosphonate.

Methods used for analyzing diisopropyl methylphosphonate in air include real-time analyses utilizing interdigitated gate electrode-field effect transistors (IGEFET), coated and uncoated piezoelectric crystal detectors, gas chromatographs, and mass spectrometers. The IGEFET method utilizes electron-beam evaporated thin films of copper phthalocyanine (CuPc) to obtain a microelectronic integrated circuit sensor that can selectively detect parts-per-billion (ppb) concentration levels of diisopropyl methylphosphonate (Kolesar and Wiseman 1989; Kolesar et al. 1992). At specific frequencies, the interaction between adsorbed diisopropyl methylphosphonate and the thin film of CuPc produces a distinct change in the film's electrical resistance and reactance, allowing the IGEFET to selectively identify diisopropyl methylphosphonate.

A number of studies (Kristoff and Guilbault 1983; Milanko et al. 1992) have investigated the use of coated and uncoated piezoelectric crystals in the detection and analysis of diisopropyl methylphosphonate in air samples. Piezoelectric crystals have a natural resonant frequency of oscillation that can be utilized to detect

TABLE 6-1. Analytical Methods for Determining Diisopropyl Methylphosphonate in Biological Samples

Sample matrix	Preparation method	Analytical method	Sample detection limit	Percent recovery	Reference
Plasma	Add cold acetonitrile, centrifuge, separate supernatant, fractionate by reverse-phase HPLC and TLC	GC/MS and PNMR	No data	No data	Weiss et al. 1994
Urine	Add cold acetonitrile, centrifuge, separate supernatant, fractionate by reverse-phase HPLC and TLC	GC/MS and PNMR	No data	No data	Weiss et al. 1994
Fecal matter	Homogenize with distilled water, add cold acetonitrile, centrifuge, separate supernatant, fractionate by reverse-phase HPLC and TLC	GC/MS and PNMR	No data	No data	Weiss et al. 1994
Beef muscle	Homogenize, filter, remove macromolecular constituents by dialysis, isolate remaining constituents on solid sorbent	GC/FID or ITMS	No data	70-85%	Caton et al. 1994

TABLE 6-1 (continued)

Sample matrix	Preparation method	Analytical method	Sample detection limit	Percent recovery	Reference
Beef muscle	Homogenize, dilute with distilled water, centrifuge and filter to remove water-insoluble particulates; extract onto solid-phase sorbents	ITMS	50–100 ppb	20–50%	Buchanan et al. 1995
Milk	Homogenize, dilute with distilled water, extract onto solid-phase sorbents	ITMS	50–100 ppb	20–50%	Buchanan et al. 1995
Milk	Homogenize, filter, remove macromolecular constituents by dialysis, isolate remaining constituents on solid sorbent	GC/FID or ITMS	No data	70–85%	Caton et al. 1994
Plant	Homogenize, filter, remove macromolecular constituents by dialysis, isolate remaining constituents on solid sorbent	GC/FID or ITMS	No data	70–85%	Caton et al. 1994

FID = flame ionization detection; GC = gas chromatography; HPLC = high performance liquid chromatography; ITMS = ion trap mass spectrometry; MS = mass spectrometry; PNMR = proton nuclear magnetic resonance; TLC = thin-layer chromatography



TABLE 6-2. Analytical Methods for Determining Diisopropyl Methylphosphonate in Environmental Samples

Sample matrix	Preparation method	Analytical method	Sample detection limit	Percent recovery	Reference
Air	None (real time)	IGEFET	No data	No data	Kolesar et al. 1992
Air	None (real time)	Uncoated piezoelectric crystal detector	0.5 ng	No data	Kristoff and Guilbault 1983
Air	None (real time)	Coated piezoelectric crystal detector	No data	No data	Milanko et al. 1992
Air	None (real time)	GC/FPD	No data	No data	Sass and Parker 1980
Air	None (real time)	MIMS	300 ppt	No data	Syage et al. 1988
Soil	Extract with distilled water, filter, pass through C <sub>18</sub> sorbent, elute with chloroform, add internal standard	GC/FID	No data	66±7%	Tornes et al. 1991
Soil	Extract using ultrasonic vibrations	GC/FID/FPD/MS	No data	No data	Sawyer et al. 1992
Soil	Extract with ethyl acetate, filter or separate, dry, concentrate	GC/AFID	No data	No data	Spanggord et al. 1979
Water	Extract with distilled water, filter, pass through C <sub>18</sub> sorbent, elute with chloroform, add internal standard	GC/FID	No data	87±10%	Tornes et al. 1991
Water	Extract with methylene	GC/MS	No data	No data	Calgon 1977

TABLE 6-2 (continued)

Sample matrix	Preparation method	Analytical method	Sample detection limit	Percent recovery	Reference
Water	chloride, filter through anhydrous sodium sulfate, evaporate, concentrate				
Water	Extract with methylene chloride, separate, dry, concentrate	GC/FPD	9.05 µg/L	No data	Fasano et al. 1982
Water	Extract with ethyl acetate, filter or separate, dry, concentrate	GC/AFID	No data	No data	Spangord et al. 1979

AFID = alkali-flame ionization detection; FID = flame ionization detection; FPD = flame photometric detection; GC = gas chromatography; IGEFET = interdigitated gate electrode field-effect transistor; ITMS = ion trap mass spectrometry; MIMS = multiphoton ionization mass spectrometry; MS = mass spectrometry

## 6. ANALYTICAL METHODS

chemical contaminants such as diisopropyl methylphosphonate. A thin coating of an appropriate substance is applied to a crystal. The film selectively adsorbs the diisopropyl methylphosphonate, which increases the mass of the piezoelectric crystal, thereby decreasing the oscillation frequency (Brothers 1990). Although piezoelectric crystals are very sensitive, there is a major problem with this sensitivity. The chemically sensitive coating must be extremely selective in its adsorption of diisopropyl methylphosphonate. Otherwise, other contaminants may also adsorb to the crystal, altering its mass and causing the piezoelectric sensor to be unable to measure the diisopropyl methylphosphonate (Brothers 1990). Piezoelectric crystal coatings include Triton X-100 (polyethylene glycol *p*-(1,1,3,3-tetramethylbutyl)phenyl ether), Aerosol-OT (sodium dioctylsulphosuccinate), dibutyl phthalate, and collodion (Milanko et al. 1992). Kristoff and Guilbault (1983) investigated the diisopropyl methylphosphonate detection ability of uncoated piezoelectric crystals with various metal electrodes (gold, silver, and nickel). They found that sensitivity increased and selectivity decreased in the following order: gold, silver, nickel. Sensitivity was found to decrease with increasing temperature (20-50° C) for the gold-plated crystal. Water vapor at 50% relative humidity was not a serious interference. The smallest weight of diisopropyl methylphosphonate detected at 10 µg L<sup>-1</sup> was approximately 0.5 ng (Kristoff and Guilbault 1983).

Gas chromatography with a flame photometric detector (Sass and Parker 1980) and multiphoton ionization mass spectrometry (MI/MS) (Syage et al. 1988) have also been used to analyze diisopropyl methylphosphonate in air samples.

Soil samples of diisopropyl methylphosphonate are detected and quantified mainly by GC, FID, FPD, and MS (Sawyer et al. 1992; Spanggord et al. 1979; Tomes et al. 1991). A number of different preparation methods exist. In a study designed to determine chemical warfare agents in samples from a battlefield environment, a solid-phase extraction method was described that extracts soil samples by shaking them in distilled water (Tomes et al. 1991). The extract is then filtered to remove particulate matter and immediately passed through a cartridge containing 200 mg C<sub>18</sub> sorbent wetted with methanol and water. The diisopropyl methylphosphonate is then eluted from the sample with chloroform, an internal standard is added, and the sample is analyzed by GC using a 30m x 0.242 mm I.D. capillary column and FID (Tomes et al. 1991). Percent recovery was 66 ± 7% from soil samples contaminated with 1 mg of diisopropyl methylphosphonate; no sample detection limit was given. Sawyer et al. (1992) described a method in which extraction is accomplished using ultrasonic vibrations prior to analysis by GC using a 15-m column and FID, FPD, and MS. No sample detection limits or percent recovery data were reported. In a study on the environmental fate of diisopropyl methylphosphonate at the RMA, Spanggord et al. (1979) described a method in which soil

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samples are extracted with three volumes of ethyl acetate, filtered or separated, dried over anhydrous  $\text{Na}_2\text{SO}_4$ , and concentrated using rotary evaporation. Triethyl phosphate (TEP) is added as an internal standard, and the sample is analyzed by GC using a 50-m SP2100 glass capillary column and alkali-flame ionization detection (AFID). Retention time for diisopropyl methylphosphonate was 9.82 minutes at a flow rate of 0.5 mL/minute  $\text{N}_2$  at 110° C isothermal. Although a detection limit was not specified, a detection level of 0.05 ppm was reported for soil extracts (Spanggord et al. 1979).

The most common method for analyzing water samples for diisopropyl methylphosphonate is GC with FID or FPD. One method described by Rosenblatt et al. (1975b) involves extraction of diisopropyl methylphosphonate from water with chloroform prior to GC analysis using a 5-foot-long, 0.25-inch-diameter glass chromatographic column filled with OV-17/Reoplexon 400 CRG. A 1-ppm solution of diisopropyl methylphosphonate in chloroform was used as a standard (Rosenblatt et al. 1975b). Methods described by Tornes et al. (1991) and Spanggord et al. (1979) for analysis of diisopropyl methylphosphonate in soils are also applicable to diisopropyl methylphosphonate analysis in water samples. Tornes et al. (1991) reported an  $87 \pm 10\%$  recovery rate from 30-mL water samples contaminated with 1 mg of diisopropyl methylphosphonate. Fasano et al. (1982) described a method for the quantification of diisopropyl methylphosphonate in water in the presence of dimethyl methylphosphonate (DMMP) and trimethyl phosphate (TMP). Samples are extracted with methylene chloride, dried, concentrated, and analyzed using a 10-foot-long, 2-mm-diameter I.D. glass GC column packed with 10% Carbowax 20M on 100/120 mesh Supelcoport. A sample detection limit of 9.05  $\mu\text{g/L}$  was reported for diisopropyl methylphosphonate in water in the presence of 10 mg/L of DMMP (Fasano et al. 1982).

### 6.3 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 diisopropyl methylphosphonate is available. Where adequate information is not available, ATSDR, in conjunction with the 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 diisopropyl methylphosphonate.

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

## 6. ANALYTICAL METHODS

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.

### 6.3.1 Identification of Data Needs

#### Methods for Determining Biomarkers of Exposure and Effect

**Exposure.** No biomarkers of exposure were identified that were specific to diisopropyl methylphosphonate. Although standard procedures exist for identifying diisopropyl methylphosphonate's primary metabolite (IMPA) in plasma, urine, and feces (Weiss et al 1994), the detection of IMPA is not unique to diisopropyl methylphosphonate exposure. IMPA is also a major metabolite of GB (Sarin) (Little et al. 1986). In addition, IMPA is cleared from the body rapidly, making it a useful indicator for recent exposure only.

**Effect.** No biomarkers of effect were identified that were specific to diisopropyl methylphosphonate. No specific target organs of diisopropyl methylphosphonate are known, and no toxic effects in humans can be positively linked to diisopropyl methylphosphonate exposure.

#### Methods for Determining Parent Compounds and Degradation Products in Environmental

**Media.** Methods exist to detect diisopropyl methylphosphonate and its degradation products in air (Kolesar et al. 1992; Kristoff and Guilbault 1983; Milanko et al. 1992; Sass and Parker 1980; Syage et al. 1988), soil (Sawyer et al. 1992; Spanggord et al. 1979; Tomes et al. 1991), and water (Calgon 1977; Fasano et al. 1982; Spanggord et al. 1979; Tomes et al. 1991). These methods are relatively sensitive and reliable and have varying degrees of selectivity.

### 6.3.2 On-going Studies

No on-going analytical methods studies were located.



## 7. REGULATIONS AND ADVISORIES

The national and state regulations and guidelines regarding diisopropyl methylphosphonate in water and other media are summarized in Table 7-1. No international regulations or guidelines were located regarding diisopropyl methylphosphonate. No NTP or IARC cancer classifications have been reported for diisopropyl methylphosphonate (IARC 1997). No regulations or guidelines were located regarding diisopropyl methylphosphonate in air.

ATSDR has derived an intermediate-duration oral MRL of 0.8 mg/kg/day based on the lack of effects including hematological effects in dogs treated with diisopropyl methylphosphonate in the diet at a dose of 75 mg/kg/day for 90 days (Hart 1980). The NOAEL was divided by an uncertainty factor of 100 to extrapolate from animals to humans and to account for human variability. ATSDR also has derived a chronic-duration oral MRL of 0.6 mg/kg/day based on the lack of effects, including hematological effects, in mink treated with 57 mg/kg/day of diisopropyl methylphosphonate for 13 months (Bucci et al. 1997). In this derivation, the NOAEL was divided by an uncertainty factor of 100 to extrapolate from animals to humans and for human variability. EPA has determined an RfD for diisopropyl methylphosphonate of 0.08 mg/kg/day with an uncertainty factor of 1,000 (10 for extrapolation of dose levels from laboratory animals to humans, 10 for uncertainty in the threshold for sensitive humans, and 10 for uncertainty in the effect of duration when extrapolating from subchronic to chronic exposure), based on the same 90-day dog feeding study (IRIS 1995). Based on this RfD and on the assumptions that a 70-kg person drinks 2 L of water/day and that 20% of the diisopropyl methylphosphonate exposure would occur through drinking water, EPA has recommended a lifetime drinking water health advisory of 0.6 mg/L (EPA 1996).

In contrast to federal government agencies, the state of Colorado has chosen a more conservative approach. Based on the Aulerich et al. (1979) study, it calculated a safe oral intake of 0.001 mg/kg/day based on a LOAEL of 11 mg/kg/day for death of female mink and an uncertainty factor of 10,000 (10 each for interspecies variation, intraspecies variation, less than lifetime exposure, and extrapolation from a LOAEL). Using the assumptions of a 70-kg person drinking 2 L of water/day, with 20% of exposure resulting from drinking water, a groundwater and surface water standard of 0.008 mg/L has been derived (Colorado 1993a, 1993b). This standard is enforceable throughout the state of Colorado.

## 7. REGULATIONS AND ADVISORIES

**TABLE 7-1. Regulations and Guidelines Applicable to Diisopropyl Methylphosphonate**

Agency	Description	Information	References
<b><u>NATIONAL</u></b>			
Guidelines:			
a. Water:			
EPA ODW	Health Advisories:		EPA 1996
	1-day (child)	8 mg/L	
	10-day (child)	8 mg/L	
	Longer term (child)	8 mg/L	
	Longer term (adult)	30 mg/L	
	RfD	0.08 mg/kg/day	
	DWEL	3 mg/L	
	Lifetime	0.6 mg/L	
EPA	Carcinogenic classification	D <sup>a</sup>	EPA 1996
EPA	Drinking water criterion	26.3 mg/L	Dacre and Rosenblatt 1987
USAMBRDL	Drinking water	0.5 ppm (0.5 mg/L)	Burrows 1978
USAMBRDL	Water for recreation	5 ppm (5 mg/L)	Burrows 1978
	Water to protect aquatic life	12.5 ppm (12.5 mg/L)	
	Water for irrigation	20 ppm (20 mg/L)	
b. Food:			
USAMBRDL	Food	0.5 ppm (0.5 mg/L)	Burrows 1978
<b><u>STATE</u></b>			
Regulations and Guidelines:			
a. Water:			
Colorado	Groundwater, surface water standards	8 ppb (0.008 mg/L)	Colorado 1993a, 1993b

<sup>a</sup> EPA Group D: Not classifiable as to human carcinogenicity

EPA = Environmental Protection Agency; DWEL = Drinking Water Equivalent Level; ODW = Office of Drinking Water; RfD = reference dose; USAMBRDL = United States Army Medical Bioengineering Research and Development Laboratory



## 7. REGULATIONS AND ADVISORIES

The two-generation study in mink (Bucci 1997) has been completed and will hopefully help resolve the differences between the federal agencies' recommendation and those of the state of Colorado.



## 8. REFERENCES

- \*Andersen ME, Krishnan K. 1994. Relating *in vitro* to *in vivo* exposures with physiologically based tissue dosimetry and tissue response models. In: Salem H, ed. Animal test alternatives: Refinement, reduction, replacement. New York, NY: Marcel Dekker, Inc., 9-25.
- \*Army. 1978. Toxicity study in rats dosed with diisopropyl methylphosphonate (DIMP) in their drinking water. Aberdeen Proving Ground, MD: Army Armament Research and Development Command. NTIS No. AD-A054-733.
- \*ATSDR. 1989. Decision guide for identifying substance-specific data needs related to toxicological profiles. Atlanta, GA: Agency for Toxic Substances and Disease Registry.
- \*ATSDR. 1996. The rocky mountain arsenal pilot exposure study; Part II: Analysis of exposure to diisopropylmethylphosphate, aldrin, dieldrin, endrin, isodrin and chlorophenylmethsulfone. Atlanta, GA: U.S. Department of Health and Human Services, Public Health Service, Agency for Toxic Substances and Disease Registry.
- \*Aulerich RJ, Coleman TH, Polin D, et al. 1979. Toxicology study of diisopropyl methylphosphonate and dicyclopentadiene in mallard ducks, bobwhite quail, and mink. East Lansing, MI: Michigan State University, Department of Poultry Science. NTIS No. AD-A087-257.
- \*Bailin LJ, Sibert ME, Jonas LA, et al. 1975. Microwave decomposition of toxic vapor simulants. Environmental Science and Technology 9(3):254-258.
- Balog PP, Stanford TB, Nordstrom RJ, et al. 1986. Feasibility assessment of piezoelectric crystals as chemical warfare agent sensors. Final report. AMD-TR-86-003. Columbus, OH: Batelle-Columbus Laboratories. NTIS No. AD-A174-065
- \*Barnes DG, Dourson M. 1988. Reference dose (RfD): Description and use in health risk estimates. Regul Toxicol Pharmacol 8:471-486.
- Bentley RE, Leblanc GA, Hollister TA, et al. 1976. Acute toxicity of diisopropylmethyl phosphonate and dicyclopentadiene to aquatic organisms. Wareham, MA: E.G. and G. Bionomics. NTIS No. AD-A037-750.
- \*Bronstein AC, Currance PL. 1988. Emergency care of hazardous materials exposure. St. Louis, MO: The C.V. Mosby Company.

- \*Brothers CP. 1990. Evaluation of an interdigitated gate electrode field-effect transistor for detecting organophosphorus compounds. Wright-Patterson AFB, OH: Air Force Institute of Technology. NTIS No. AD-A230-161.
- \*Bucci TJ, Mercieca MD, Perman V. 1997. Two-generation reproductive study in mink fed DIMP: Final report, study No. TP-001. Prepared by Pathology Associates International, Jefferson, AR.
- \*Bucci TJ, Parker RM, Wustenberg W. 1992. A 90-Day oral toxicity study and a 5-day metabolism study of diisopropyl methylphosphonate (DIMP) in mink. Jefferson, AR: Pathology Associates, Inc. NTIS No. AD-A257-397.
- \*Bucci TJ, Wustenberg W, Perman V, et al. 1994. Subchronic oral toxicity study of diisopropyl methylphosphonate in mink. *Fundam Appl Toxicol*22(2):220-230.
- \*Buchanan MV, Hettich RL, Xu JH. 1995. Low level detection of chemical agent simulants in meat and milk by ion trap mass spectrometry. *Journal of Hazardous Materials* 42:49-59.
- \*Burrows WD. 1978. Development of guidelines for contaminated soil and groundwater at U.S. Army installations. 4th Jt. Conf. Sens. Environ. Pollut., 80-82.
- Butler MA, Ricco AJ. 1992. Fibre-optic micro-mirror studies of the interaction of thin copper films with an organophosphonate. *Anal Chem* 64(17): 1851- 1854.
- \*Calabrese E. 1990. Health effects assessment of diisopropyl methylphosphonate. Prepared for State of Colorado, Department of Health, November 12, 1990.
- \*Calgon Corporation. 1977. Removal of trace organics from groundwater using granular activated carbons. Denver, CO: Calgon Corporation. Government Reports Announcements and Index (GRA&I) Issue 07. NTIS No. AD-A956-5 1316.
- Casazza JP, Felver ME, Veech RL. 1984. The metabolism of acetone in rats. *J Biol Chem* 259(1):23 1-236.
- \*Caton JE, Hazen M, Griest WH, et al. 1994. Rapid method for isolating targeted organic chemicals from biological matrixes. *Anal Lett* 27(2):35 1-362.
- \*Clewell HJ LII, Andersen ME. 1985. Risk assessment extrapolations and physiological modeling. *Toxicol Ind Health* 1(4):111-131.
- \*Colorado. 1993a. The basic standards for ground water 3.11.0. Colorado Department of Health, Water Quality Commission.
- \*Colorado. 1993b. The basic standards and methodologies for surface water 3.1.0. (SCCR 1002-8). Colorado Department of Health, Water Quality Commission
- \*Cysewski SJ, Palmer JS, Crookshank HR, et al. 1981. Toxicologic evaluation of diisopropyl methylphosphonate and dicyclopentadiene in cattle. *Arch Environ Contam Toxicol* 10(5):605-615.

Dacre JC. 1975. Fact sheet on DIMP Toxicity. U.S. Army Medical Bioengineering Research and Development Laboratory.

Dacre JC. 1984. Recommended interim criteria for three environmental polluting compounds of the RMA Technical Report 8302. U.S. Army Medical Bioengineering Research and Development Laboratory. Fort Detrick, Frederick, MD. NTIS No. AD-A154-82612.

Dacre JC, Hart ER. 1978. Mammalian toxicologic studies on diisopropyl methylphosphonate. In: Proceedings of the First International Congress on Toxicology. Toronto, ON, Canada, March 30 - April 2, 1977.

\*Dacre JC, Rosenblatt DH. 1987. Drinking water criteria for the groundwater pollutant diisopropyl methylphosphonate (DIMP). Fort Detrick, MD: Army Biomedical Research and Development Lab. NTIS No. AD-A1 86-562.

\*Department of Defense. 1991a. Cytogenetic testing on di-isopropylmethylphosphonate employing the rodent bone marrow micronucleus assay in male B6C3F<sub>1</sub>. Aberdeen Proving Grounds, MD: U.S. Army Environmental Hygiene Agency. Contract no. DAAD05-89C-0224.

\*Department of Defense. 1991b. Cytogenetic testing on di-isopropylmethylphosphonate employing the rodent bone marrow micronucleus assay in male Fischer 344 rats. Aberdeen Proving Grounds, MD: U.S. Army Environmental Hygiene Agency. Contract no. DAAD05-89C-0224.

\*Department of Defense. 1991c. In vitro DNA damage testing on di-isopropylmethylphosphonate employing the single cell gel assay in Chinese hamster ovary cells. Aberdeen Proving Grounds, MD: U.S. Army Environmental Hygiene Agency. Contract no. DAAD05-89C-0224.

\*Department of Defense. 1991d. In vitro genotoxicity testing of di-isopropylmethylphosphonate employing the chromosome aberration assay in Chinese hamster ovary cells. Aberdeen Proving Grounds, MD: U.S. Army Environmental Hygiene Agency. Contract no. DAAD05-89C-0224.

\*Department of Defense. 1991e. In vitro genotoxicity testing of di-isopropylmethylphosphonate employing the sister chromatid exchange assay in Chinese hamster ovary cells. Aberdeen Proving Grounds, MD: U.S. Army Environmental Hygiene Agency. Contract no. DAAD05-89C-0224.

\*Department of Defense. 1991f. In vivo DNA damage testing on di-isopropylmethylphosphonate employing the single cell gel assay in male B6C3F<sub>1</sub> mice. Aberdeen Proving Grounds, MD: U.S. Army Environmental Hygiene Agency. Contract no. DAAD05-89C-0224.

\*Department of Defense. 1991 g. In vivo DNA damage testing on di-isopropylmethylphosphonate employing the single cell gel assay in male Fischer 344 rats. Aberdeen Proving Grounds, MD: U.S. Army Environmental Hygiene Agency. Contract no. DAAD05-89C-0224.

\*Department of Defense. 1991 h. Mouse lymphoma mutagenesis assay on diisopropylmethylphosphonate and dimethylmethylphosphonate. Aberdeen Proving Grounds, MD: U.S. Army Environmental Hygiene Agency. Contract no. DAAD05-89C-0224.

- \*D.P. Associates. 1995. RMA Environmental Database.
- \*Ebasco Services Incorporated. 1991. Surficial soil program. Data Summary, Final Report , Version 3.1.
- \*Ebasco Services Incorporated. 1992. Remedial Investigation Summary Report, Final Version 3.2.
- \*Ellenhorn MJ, Barceloux DG. 1988. Medical toxicology. Diagnosis and treatment of human poisoning. Elsevier, New York.
- \*Environmental Science and Engineering, Inc. 1988. Final phase II data addendum, site 36-3: Insecticide pits. Version 3.1a
- \*EPA. 1989. Health advisory for diisopropyl methylphosphonate (DIMP). U.S. Environmental Protection Agency, Office of Drinking Water. NTIS No. PB90-273517.
- EPA. 1990. Interim methods for development of inhalation reference concentrations. Research Triangle Park, NC: U.S. Environmental Protection Agency. EPA/600/%88/066F.
- \*EPA. 1992. Health advisory for isopropyl methylphosphonic acid (IMPA). U.S. Environmental Protection Agency, Office of the Assistant Administrator for Water. NTIS No. PB92-232149.
- \*EPA. 1996. Drinking water regulations and health advisories. Washington, DC: U.S. Environmental Protection Agency, Office of Water. February 1996. EPA 822/R-96-001.
- \*Fasano R, Randel M, Sadowski L, et al. 1982. Analytical methods development for dimethyl methylphosphonate, diisopropyl methylphosphonate, and trimethyl phosphate. Cambridge, MA: Arthur D. Little, Inc. NTIS No. AD-A120-863.
- \*Ford-Moore AH, Perry BJ. 195 1. Diisopropyl methylphosphonate. Org Syntheses 3 1:33-35.
- Foster Wheeler Environmental Corporation. 1995. Feasibility Study, Soil Volume refinement program, geotechnical sampling in the new toxic storage yard, section 31, Rocky Mountain Arsenal. Final Report. Version 2.0.
- \*FSTRAC. 1990. Summary of state and federal drinking water standards and guidelines. Chemical Communication Subcommittee, Federal-State Toxicology and Regulatory Alliance Committee. Washington, DC: U.S. Environmental Protection Agency, Office of Water. EPA 570/R-90-019.
- Geary RS, Wall CM, Miller MA, et al. 1994. Partition coefficient measurements of diisopropyl methylphosphonate (DIMP) and trichloroethylene in rats using microdialysis and incorporated in physiologically-based pharmacokinetic (PBPK) modelling [Abstract]. Society of Toxicology 33rd Annual Meeting, Dallas, TX: 13-17 March, 1994. Paper No. 82.
- Griest WH, Ramsey RS, Ho CH, et al. 1992. Supercritical fluid extraction of chemical warfare agent simulants from soil. J Chromatogr 600(2):273-277..

Guilbault GG, Das J. 1969. Chemisorption reactions of diisopropylmethyl phosphonate with transition metal salts. *Journal of Physical Chemistry* 73(7):2243-2247.

Guilbault GG, Scheide EP. 1970. Chemisorption reactions of diisopropyl methyl phosphonate with various metal salts and the effect of complex-ion formation on the phosphorus-oxygen stretching frequency. *Journal of Inorganic and Nuclear Chemistry* 32(9):2959-2962.

Guilbault G, Ngeh-Ngwainbi J, Foley P, et al. 1986. Use of protein coatings on piezoelectric crystals for assay of gaseous pollutants. *Anal Chem Symp Ser Electrochem Sens Anal* 25:335-341.

\*Harding Lawson Associates. 1994. Feasibility Study. Soils support program report; Rocky Mountain Arsenal, Commerce City, Colorado. Draft Final Versiou 2.

\*Harding Lawson Associates. 1992. Qffpost operable unit remedial investigation final addendum.

\*Hardisty JF, Pellerin RJ, Biskup RK, et al. 1977. Reproductive studies with diisopropylmethylphosphonate in rats. Aberdeen Proving Ground, MD: Chemical Systems Laboratory. NTIS No. AD-A040-454

\*Hart ER. 1976. Mammalian toxicological evaluation of DIMP and DCPD. Final Report. Kensington, MD: Litton Bionetics, Inc. NTIS No. AD-A058-323.

\*Hart ER. 1980. Mammalian toxicological evaluation of DIMP and DCPD (Phase II). Kensington, MD: Litton Bionetics, Inc. NTIS No. AD-A082-685.

\*HAZDAT. 1997. Agency for Toxic Substances and Disease Registry (ATSDR), Atlanta, GA.

Hlavay J, Guilbault GG. 1977. Applications of the piezoelectric crystal detector in analytical chemistry. *Anal Chem*49(13):1890-1898.

\*Hoskin FCG. 1956. Some observations concerning the biochemical inertness of methyl phosphonic and isopropyl methylphosphonic acids. *Can J Biochem Physiol*34:743-746.

\*HSDB. 1994. Hazardous substance data bank. Bethesda, MD: National Library of Medicine, National Toxicology Information Program. June 7, 1994.

\*IARC. 1997. Overall evaluations of carcinogenicity to humans. World Health Organization, Lyon, France.

\*IRIS. 1997. Integrated Risk Information Systems. U.S. Environmental Protection Agency, Office of Health and Environmental Assessment, Environmental Criteria and Assessment Office, Cincinnati, Ohio.

\*Ivie GW. 1980. Fate of diisopropyl methylphosphonate (DIMP) in a lactating cow. *Bull Environ Contam Toxicol* 24(1):40-48.

\*Jenkins RA, Buchanan MV, Merriweather R, et al. 1994. Movement of chemical warfare simulants through porous media. *Journal of Hazardous Materials* 37(2):303-325.

- \*Jones RE, Howell KS, Ringer RK. 1992. Effect of an environmental contaminant, diisopropyl methylphosphonate, on the blood pressure of the mallard. *Biomed Environ Sci* 5(4):3 14-320.
- Karayannis NM, Mikulski CM, Strocko MJ, et al. 197 la. Reactions between diisopropyl methylphosphonate and halides of divalent metal ions. *Z. Anorg Allg C&em* 384(3):267-279.
- Karayannis NM, Mikulski CM, Strocko MJ, et al. 197lb. Reactions of alkali metal iodides with diisopropyl methylphosphonate. *Inorg Chim Acta* 5(3):357-361.
- Karayannis NM, Owens C, Pytlewski LL, et al. 1969. Diisopropyl methylphosphonate complexes of metal perchlorates. *Journal of Inorganic and Nuclear Chemistry* 3 1(7):2059-2071.
- Karayannis NM, Owens C, Pytlewski LL, et al. 1970. Complexes of diisopropyl methylphosphonate with metal salts containing complex anionic groups. *Journal of Inorganic and Nuclear Chemistry* 32(1):83-90.
- Karayannis NM, Pytlewski LL, Owens C. 1980. Studies of adduct formation between diisopropyl methylphosphonate and various metal-salts and complexes. *Journal of Inorganic & Nuclear Chemistry* 42(5):675-682.
- Ketkar SW, Penn SM, Fite WL. 199 1. Influence of coexisting analytes in atmospheric pressure ionization mass spectrometry. *Anal Chem* 63(9):924-925.
- Koch P, Rumpel H, Sutter P, et al. 1989. The reaction of alkanephosphonic acid esters with metals. *Phosphorus, Sulfur, Silicon, and Related Elements* 44(1-2):75-85.
- \*Kolesar ES, Wiseman JM. 1989. Interdigitated gate electrode field effect transistor for the selective detection of nitrogen dioxide and diisopropyl methylphosphonate. *Anal Chem* 61(21):2355.
- Kolesar ES, Wiseman JM. 199 1. Selective detection of nitrogen-dioxide and diisopropyl methylphosphonate with an interdigitated gate electrode field-effect transistor (IGEFET). *Sensors and Actuators B - Chemical* 5(1-4):37-46.
- \*Kolesar ES, Brothers CP, Howe CP, et al. 1992. Integrated-circuit microsensor for selectively detecting nitrogen-dioxide and diisopropyl methylphosphonate. *Thin Solid Films* 220(1-2):30-37.
- Krikorian SE, Chorn TA, King JW. 1987. Determination of octanovwater partition coefficients of certain organophosphorus compounds using high-performance liqluid chromatography. *Quant Struct-Act Relat* 6(2):65-69.
- \*Krishnan K, Andersen ME. 1994. Physiologically based pharmacokinetic modeling in toxicology. In: Hayes W, ed. *Principles and methods of toxicology*. 3rd ed. New York, NY: Raven Press, Ltd., 149-188.
- \*Krishnan K, Andersen ME, Clewell HJ III, et al. 1994. Physiologically based pharmacokinetic modeling of chemical mixtures. In: Yang, RSA, ed. *Toxicology of chemical mixtures*. New York, NY: Academic Press, 399-437.



## 8. REFERENCES

\*Kristoff J, Guilbault GG. 1983. Application of uncoated piezoelectric crystals for the detection of an organic phosphonate. *Anal Chim Acta* 149:337-341.

Kuitunen ML, Hartonen K, Riekkola ML. 1991. Analysis of chemical warfare agents in soil samples by off-line supercritical fluid extraction and capillary gas chromatography. *Journal of Microcolumn Separation* 3(6):505-512.

\*Leung H-W. 1993. Physiologically-based pharmacokinetic modeling. In: Ballantyne B, Marrs T, Turner P, eds. *General and applied toxicology*. New York, NY: Stockton Press, 1:153-164.

\*Little PJ, Reynolds ML, Bowman ER, et al. 1986. Tissue disposition of (3H)sarin and its metabolites in mice. *Toxicol Appl Pharmacol* 83(3):412-419.

\*Little PJ, Scimeca JA, Martin BR. 1988. Distribution of (3H)diisopropyl fluorophosphate, (3H)soman, (3H)sarin, and their metabolites in mouse brain. *Drug Metab Dispos* 16(4):515-520.

Long SR, Christesen SD. 1989. Laser ionization studies of organophosphonates and phosphorus oxide radicals. *Journal of Physical Chemistry* 93(18):6625-6628.

\*McPhillips DM, Armer TA, Owen DR. 1983. Grafted synthetic sorbents for enhanced removal of toxic chemical agents from plasma. *J Biomed Mater Res* 17(6):993-1002.

Mikulski CM, Harris N, Iaconianni FJ, et al. 1980. Group-VI metal hexacarbonyl reactions with diisopropyl methylphosphonate. *Inorganic & Nuclear Chemistry* 16(2):79-89.

Mikulski CM, Pytlewski LL, Karayannis NM. 1977. WCL6 and MOCLS interactions with diisopropyl methylphosphonate. *Inorganic & Nuclear Chemistry* 13(3-4): 165-171.

\*Milanko OS, Milinkovic SA, Rajakovic LV. 1992. Improved methodology for testing and characterization of piezoelectric gas sensors. *Anal Chim Acta* 264(1):43-52.

\*NAS/NRC. 1989. *Biological markers in reproductive toxicology*. National Academy of Sciences/National Research Council. Washington, DC: National Academy Press, 15-35.

\*NIOSH. 1981. Health hazard evaluation report HETA 81-176-968, Rocky Mountain Arsenal, Basin F, Commerce City, Colorado. Cincinnati, OH: National Institute for Occupational Safety and Health. NTIS No. PB83-161257.

NTDB. 1997. National Trade Data Bank. U.S. Department of Commerce, Economics and Statistics Administration, Washington, D.C.

\*O'Donovan PA, Woodward JE. 1977a. Investigation of the soil translocation and phytotoxicity of DIMP and DCPD. Downey, CA: Aerojet Ordnance and Manufacturing Co. NTIS No. AD-A058-790.

\*O'Donovan PA, Woodward JE. 1977b. Investigations of the environmental fate and phytotoxicity of DIMP and DCPD. Downey, CA: Aerojet Ordnance Co. NTIS No. AD-A956-500.

## 8. REFERENCES

- Owen OE, Trapp VE, Skutches CL, et al. 1982. Acetone metabolism during diabetic ketoacidosis. *Diabetes* 31:242-248
- Owens, C. 1969. Adducts of metal halides and diisopropylmethylphosphonate: synthesis and properties. [Dissertation] Philadelphia, PA: Drexel Institute of Technology.
- Owens C, Karyannis NM, Pytlewski LL, et al. 1971. Infrared and proton nuclear magnetic resonance studies of adduct of tin(II) and (IV) and titanium(IV) halides with diisopropyl methylphosphonate. *Journal of Physical Chemistry* 75(5):637-641.
- Owens C, Pytlewski LL, Mikulski CM, et al. 1979. X-ray crystallographic, morphological and thermal-decomposition studies of 2-l adducts of diisopropyl methylphosphonate with stannous and stannic halides. *Journal of Inorganic & Nuclear Chemistry* 41(9):1261-1268.
- \*Pal T, Griffin GD, Miller GH, et al. 1993. Permeation measurements of chemical agent simulants through protective clothing materials. *Journal of Hazardous Materials* 33(1):123-141.
- \*Palmer JS, Cysewski SJ, Crockshank HR, et al. 1979. Toxicologic evaluation and fate of diisopropyl methylphosphonate (DIMP) and dicyclopentadiene (DCPD) in cattle. College Station, TX: Science and Technology Administration. NTIS No. AD-8093-673.
- Pappas AA, Ackerman BH, Olsen KM, et al. 1991. Isopropanol ingestion: A report of 6 episodes with isopropanol and acetone serum concentration time data. *Clin Toxicol* 21(1): 11-21.
- Purdon JG, Pagotto JG, Miller RK. 1989. Preparation, stability, and quantitative analysis by gas chromatography and gas chromatography-electron impact mass spectrometry of tert-butyldimethylsilyl derivatives of some alkylphosphonic and alkyl methylphosphonic acids. *J Chromatogr* 475:261-272.
- \*Radziemski LJ Jr. 1981. Laser-induced photodestruction of the organo-phosphates: DIMP and DMMP. *J Environ Sci Health* 16(3):337-361.
- Reichard GA, Skutches CL, Hoeldtke RD, et al. 1986. Acetone metabolism in humans during diabetic ketoacidosis. *Diabetes* 35:668-674.
- \*Roberts WC, Abernathy CO, Commons BJ. 1995. US drinking water health advisories; methodology and basis for some munitions chemicals. *Toxicol Ecotoxicol News* 2( 1):4-8.
- \*Robson SG. 1977. Digital model study of groundwater contamination by diisopropylmethylphosphonate (DIMP), RMA near Denver, Colorado. United States Geological Survey. NTIS No. AD-A956-502.
- \*Robson SG. 1981. Computer simulation of movement of DIMP-contaminated groundwater near the Rocky Mountain Arsenal, Colorado. ASTM Special Technical Publication 746:209-220.
- Rosenblatt DH, Miller TA, Dacre JC, et al. 1975a. Problem definition studies on potential environmental pollutants. I. Toxicology and ecological hazards of 16 substances at the RMA. Technical Report 7508. U.S. Army Medical Bioengineering Research and Development Laboratory, Fort Detrick, Frederick, MD.

- \*Rosenblatt DH, Miller TA, Dacre JC, et al. 1975b. Problem definition studies on potential environmental pollutants. II. Physical, chemical, toxicological, and biological properties of 16 substances. Technical Report 7509. U.S. Army Medical Bioengineering Research and Development Laboratory, Fort Detrick, Frederick, MD. NTIS No. AD-A030-428.
- \*Sass S, Parker GA. 1980. Structure-response relationship of gas chromatography-flame photometric detection to some organophosphorus compounds. *J Chromatogr* 189(3):331-349.
- Sass S, Fisher TL, Steger RJ, et al. 1982. Gas-chromatographic methods for the analysis of trace quantities of isopropyl methylphosphonofluoridate and associated compounds, in situ and in decontamination effluent. *J Chromatogr* 238(2):445-456.
- \*Sawyer TW, Weiss MT, D'Agostino PA, et al. 1992. Bioassay of organophosphate nerve agents in soil using neuronal tissue cultures. *J Appl Toxicol* 12(1):1-6.
- Scheide EP, Guilbault GG. 1972. Piezoelectric detectors for organophosphorus compounds and pesticides. *Anal Chem* 44(11):1764-1768.
- \*Schneider RR, Hunter DB. 1993. Nursing disease in mink: Clinical and postmortem findings. *Vet Pathol* 30(6):512-521.
- Shin JE. 1989. Evaluation of chemically-sensitive field-effect transistors for detection of organophosphorus compounds. Wright-Patterson AFB, OH: Air Force Institute of Technology. NTIS No. AD-A215-536.
- \*Snodgrass HL, Metker LM. 1992. Dermal penetration of C-14-labelled diisopropyl methylphosphonate in swine. *J Toxicol Environ Health* 36(4):367-376.
- Snyder AP, Harden CS. 1990. Determination of the fragmentation mechanisms of organophosphorus ions by water and deuterium oxide atmospheric-pressure ionization tandem mass spectrometry. II. Dialkylphosphonate ions. *Org Mass Spectrom* 25(6):301-308.
- \*Spangord RJ, Chou T, Mabey WR. 1979. Studies of environmental fates of DIMP and DCPD. Menlo Park, CA: SRI, International. NTIS No. AD-A078-236.
- St. John GA, Anbar M. 1974. Determination of subpicogram amounts of chemical agents in the atmosphere. EC-CR-74028. Menlo Park, CA: Stanford Research Institute. NTIS No. AD/A-000-886.
- \*Stutz DR, Ulin S. 1992. Basic and advanced life support. Hazardous materials injuries: A handbook for prehospital care, 3rd ed. Bradford Communications Corporation, Beltsville, MD.
- \*Syage JA, Pollard JE, Cohen RB. 1988. Ultrasensitive detection of atmospheric constituents-by supersonic molecular beam, multiphoton ionization mass spectroscopy. El Segundo, CA: Aerospace Corp. NTIS No. AD-A202-299.
- \*Tornes JA, Opstad AM, Johnsen BA. 1991. Use of solid-phase extraction in determination of chemical warfare agents. Part II. Determination of chemical warfare agents in samples from a battlefield environment. *International Journal of Environmental Analytical Chemistry* 44(4):227-232.

TR195. 1997. Toxic Chemical Release Inventory. U.S. Environmental Protection Agency. Office of Toxic Substances, Washington, DC.

USGS. 1995. Groundwater monitoring program evaluation report for water year 1995. Rocky Mountain Arsenal, Commerce City, Colorado. United States Geological Survey.

USGS. 1998. Surface water and groundwater monitoring programs. United States Geological Survey. Rocky Mountain Arsenal, Annual Data Summary, 1997 Water Year. Final Report.

\*Van Voris P, Cataldo DA, Ligothke MW, et al. 1987. Acute environmental toxicity and persistence of selected chemical agent simulants: diisopropyl fluorophosphate (DFP) and diisopropyl methylphosphonate (DIMP). NTIS No. AD-A181-309.

Verweij A, Boter I-IL, Degenhardt C. 1979. Chemical warfare agents: verification of compounds containing the phosphorus-methyl linkage in waste water. *Science* 204(4393):616-618.

\*Vo-Dinh T, Pal, T. 1992. Development of a fluorescence quenching technique to detect permeation of chemical agent simulants through protective clothing materials. *Appl Spectrosc* 46(4):677-681.

\*Weiss DJ, Geary RS, Wustenberg W, et al. 1994. Comparative metabolism of diisopropyl methylphosphonate in mink and rats. *Arch Environ Contam Toxicol* 27:420-425.

Weiss DJ, Wustenberg W, Bucci TJ, et al. 1992. Hematologic alterations in mink induced by diisopropyl methylphosphonate (DIMP) [Abstract]. *Toxicol Pathol* 20:646.

\*Williams RT, Ziegenfuss PS, Marks PJ. 1989. Installation restoration program environmental technology department. Biodegradation of DIMP, dieldrin, isodrin, DBCP, and PCPMSO in the RMA soils. West Chester, PA: Roy F. Weston, Inc. NTIS No. AD-A245-852.

Wils ERJ. 1990. Mass spectral data of precursors of chemical warfare agents. *Fresenius Journal of Analytical Chemistry* 338(1):22-27.

\*Zappi ME, Fleming EC, Thompson DW, et al. 1990. Treatability study of four contaminated areas at the RMA, Commerce City, Colorado using chemical oxidation with ultraviolet radiation catalyzation. Proceedings of the 7th National Conference on Hazardous Waste Materials, 405-409.

## 9. GLOSSARY

**Acute Exposure** -- Exposure to a chemical for a duration of 14 days or less, as specified in the Toxicological Profiles.

**Adsorption Coefficient ( $K_{oc}$ )** -- The ratio of the amount of a chemical adsorbed per unit weight of organic carbon in the soil or sediment to the concentration of the chemical in solution at equilibrium.

**Adsorption Ratio ( $K_d$ )** -- The amount of a chemical adsorbed by a sediment or soil (i.e., the solid phase) divided by the amount of chemical in the solution phase, which is in equilibrium with the solid phase, at a fixed solid/solution ratio. It is generally expressed in micrograms of chemical sorbed per gram of soil or sediment.

**Bioconcentration Factor (BCF)** -- The quotient of the concentration of a chemical in aquatic organisms at a specific time or during a discrete time period of exposure divided by the concentration in the surrounding water at the same time or during the same period.

**Cancer Effect Level (CEL)** -- The lowest dose of chemical in a study, or group of studies, that produces significant increases in the incidence of cancer (or tumors) between the exposed population and its appropriate control.

**Carcinogen** -- A chemical capable of inducing cancer.

**Ceiling Value** -- A concentration of a substance that should not be exceeded, even instantaneously.

**Chronic Exposure** -- Exposure to a chemical for 365 days caused death in humans or animals.

**Lethal Concentration<sub>(50)</sub>( $LC_{50}$ )** -- A calculated concentration of a chemical in air to which exposure for a specific length of time is expected to cause death in 50% of a defined experimental animal population.

**Lethal Dose<sub>(LO)</sub>( $LD_{LO}$ )** -- The lowest dose of a chemical introduced by a route other than inhalation that is expected to have caused death in humans or animals.

**Lethal Dose<sub>(50)</sub>( $LD_{50}$ )** -- The dose of a chemical which has been calculated to cause death in 50% of a defined experimental animal population.

**Lethal Time<sub>(50)</sub>( $LT_{50}$ )** -- A calculated period of time within which a specific concentration of a chemical is expected to cause death in 50% of a defined experimental animal population.

**Lowest-Observed-Adverse-Effect Level (LOAEL)** -- The lowest dose of chemical in a study, or group of studies, that produces statistically or biologically significant increases in frequency or severity of adverse effects between the exposed population and its appropriate control.

**Malformations** -- Permanent structural changes that may adversely affect survival, development, or function.

**Minimal Risk Level** -- An estimate of daily human exposure to a dose of a chemical that is likely to be without an appreciable risk of adverse noncancerous effects over a specified duration of exposure.

**Mutagen** -- A substance that causes mutations. A mutation is a change in the genetic material in a body cell. Mutations can lead to birth defects, miscarriages, or cancer.

**Neurotoxicity** -- The occurrence of adverse effects on the nervous system following exposure to chemical.

**No-Observed-Adverse-Effect Level (NOAEL)** - The dose of chemical at which there were no statistically or biologically significant increases in frequency or severity of adverse effects seen between the exposed population and its appropriate control. Effects may be produced at this dose, but they are not considered to be adverse.

**Octanol-Water Partition Coefficient ( $K_{ow}$ )** -- The equilibrium ratio of the concentrations of a chemical in n-octanol and water, in dilute solution.

**Permissible Exposure Limit (PEL)** -- An allowable exposure level in workplace air averaged over an 8-hour shift.

**$q_1^*$**  -- The upper-bound estimate of the low-dose slope of the dose-response curve as determined by the multistage procedure. The  $q_1^*$  can be used to calculate an estimate of carcinogenic potency, the incremental excess cancer risk per unit of exposure (usually  $\mu\text{g}/\text{L}$  for water,  $\text{mg}/\text{kg}/\text{day}$  for food, and  $\mu\text{g}/\text{m}^3$  for air).

**Reference Dose (RfD)** -- An estimate (with uncertainty spanning perhaps an order of magnitude) of the daily exposure of the human population to a potential hazard that is likely to be without risk of deleterious effects during a lifetime. The RfD is operationally derived from the NOEL (from animal and human studies) by a consistent application of uncertainty factors that reflect various types of data used to estimate RfDs and an additional modifying factor, which is based on a professional judgment of the entire database on the chemical. The RfDs are not applicable to nonthreshold effects such as cancer.

**Reportable Quantity (RQ)** -- The quantity of a hazardous substance that is considered reportable under CERCLA. Reportable quantities are (1) 1 pound or greater or (2) for selected substances, an amount established by regulation either under CERCLA or under Sect. 311 of the Clean Water Act. Quantities are measured over a 24 hour period.

**Reproductive Toxicity** -- The occurrence of adverse effects on the reproductive system that may result from exposure to a chemical. The toxicity may be directed to the reproductive organs and/or the related endocrine system. The manifestation of such toxicity may be noted as alterations in sexual behavior, fertility, pregnancy outcomes, or modifications in other functions that are dependent on the integrity of this system.

**Short-Term Exposure Limit (STEL)** -- The maximum concentration to which workers can be exposed for up to 15 min continually. No more than four excursions are allowed per day, and there must be at least 60 min between exposure periods. The daily TLV-TWA may not be exceeded.

**Target Organ Toxicity** -- This term covers a broad range of adverse effects on target organs or physiological systems (e.g., renal, cardiovascular) extending from those arising through a single limited exposure to those assumed over a lifetime of exposure to a chemical.

**Teratogen** -- A chemical that causes structural defects that affect the development of an organism.

**Threshold Limit Value (TLV)** -- A concentration of a substance to which most workers can be exposed without adverse effect. The TLV may be expressed as a TWA, as a STEL, or as a CL.

**Time-Weighted Average (TWA)** -- An allowable exposure concentration averaged over a normal 8-hour workday or 40-hour workweek.

**Toxic Dose (TD<sub>50</sub>)** -- A calculated dose of a chemical, introduced by a route other than inhalation, which is expected to cause a specific toxic effect in 50% of a defined experimental animal population.

**Uncertainty Factor (UF)** -- A factor used in operationally deriving the RfD from experimental data. UFs are intended to account for (1) the variation in sensitivity among the members of the human population, (2) the uncertainty in extrapolating animal data to the case of human, (3) the uncertainty in extrapolating from data obtained in a study that is of less than lifetime exposure, and (4) the uncertainty in using LOAEL data rather than NOAEL data. Usually each of these factors is set equal to 10.

## APPENDIX A

### ATSDR MINIMAL RISK LEVEL

The Comprehensive Environmental Response, Compensation, and Liability Act (CERCLA) [42 U.S.C. 9601 et seq.], as amended by the Super-fund Amendments and Reauthorization Act (SARA) [Pub. L. 99-499], requires that the Agency for Toxic Substances and Disease Registry (ATSDR) develop jointly with the US Environmental Protection Agency (EPA), in order of priority, a list of hazardous substances most commonly found at facilities on the CERCLA National Priorities List (NPL); prepare toxicological profiles for each substance included on the priority list of hazardous substances; and assure the initiation of a research program to fill identified data needs associated with the substances.

The toxicological profiles include an examination, summary, and interpretation of available toxicological information and epidemiologic evaluations of a hazardous substance. During the development of toxicological profiles, Minimal Risk Levels (MRLs) are derived when reliable and sufficient data exist to identify the target organ(s) of effect or the most sensitive health effect(s) for a specific duration for a given route of exposure. An MRL is an estimate of the daily human exposure to a hazardous substance that is likely to be without appreciable risk of adverse noncancer health effects over a specified duration of exposure. MRLs are based on noncancer health effects only and are not based on a consideration of cancer effects. These substance-specific estimates, which are intended to serve as screening levels, are used by ATSDR health assessors to identify contaminants and potential health effects that may be of concern at hazardous waste sites. It is important to note that MRLs are not intended to define clean-up or action levels.

MRLs are derived for hazardous substances using the no-observed-adverse-effect level uncertainty factor approach. They are below levels that might cause adverse health effects in the people most sensitive to such chemical-induced effects. MRLs are derived for acute (1-14 days), intermediate (15-364 days), and chronic (365 days and longer) durations and for the oral and inhalation routes of exposure. Currently, MRLs for the dermal route of exposure are not derived because ATSDR has not yet identified a method suitable for this route of exposure. MRLs are generally based on the most sensitive chemical-induced end point considered to be of relevance to humans. Serious health effects (such as irreparable damage to the liver or kidneys, or birth defects) are not used as a basis for establishing MRLs. Exposure to a level above the MRL does not mean that adverse health effects will occur.



## APPENDIX A

MRLs are intended only to serve as a screening tool to help public health professionals decide where to look more closely. They may also be viewed as a mechanism to identify those hazardous waste sites that are not expected to cause adverse health effects. Most MRLs contain a degree of uncertainty because of the lack of precise toxicological information on the people who might be most sensitive (e.g., infants, elderly, nutritionally or immunologically compromised) to the effects of hazardous substances. ATSDR uses a conservative (i.e., protective) approach to address this uncertainty consistent with the public health principle of prevention. Although human data are preferred, MRLs often must be based on animal studies because relevant human studies are lacking. In the absence of evidence to the contrary, ATSDR assumes that humans are more sensitive to the effects of hazardous substance than animals and that certain persons may be particularly sensitive. Thus, the resulting MRL may be as much as a hundredfold below levels that have been shown to be nontoxic in laboratory animals.

Proposed MRLs undergo a rigorous review process: Health Effects/MRL Workgroup reviews within the Division of Toxicology, expert panel peer reviews, and agencywide MRL Workgroup reviews, with participation from other federal agencies and comments from the public. They are subject to change as new information becomes available concomitant with updating the toxicological profiles. Thus, MRLs in the most recent toxicological profiles supersede previously published levels. For additional information regarding MRLs, please contact the Division of Toxicology, Agency for Toxic Substances and Disease Registry, 1600 Clifton Road, Mailstop E-29, Atlanta, Georgia 30333.

**MINIMAL RISK LEVEL WORKSHEET**

Chemical Name: Diisopropyl Methylphosphonate  
CAS Number: 144-75-6  
Date: March 1998  
Profile Status: Draft 2 - Postpublic Comment  
Route:  Inhalation  Oral  
Duration:  Acute  Intermediate  Chronic  
Graph Key: 27  
Species : Dog

Minimal Risk Level: 0.8 mg/kg/day

Reference: Hart 1980

Experimental design: Purebred beagles, four males and four females, received diisopropyl methylphosphonate in the diet at concentrations of 150, 1,500, and 3,000 ppm (4, 38, or 75 mg/kg/day) for 90 days. A control group of four males and four females was also maintained. At the outset of the study and at 48, and 13 weeks, hemograms and clinical chemistry parameters were determined for all of the dogs. The dogs were examined daily for general condition. Food consumption and body weights were determined weekly. At the termination of the 90-day study each dog was necropsied and selected tissues preserved and examined for histopathological changes.

Effects noted in study and corresponding doses:

The dogs appeared in good condition throughout the study. No hematological effects were ascribed to diisopropyl methylphosphonate. The authors concluded that the ingestion of diisopropyl methylphosphonate produced no toxic effects at the concentrations that the dogs received over the 90-day period of the study.

At the termination of the study a gross necropsy was performed on all of the dogs and no meaningful changes were observed. In addition, the liver, brain, thyroid, kidneys, adrenal glands, testes, ovaries, heart, and spleen were removed and weighed. No significant weight changes were noted. The liver, brain, thyroid, kidneys, adrenal glands, testes, ovaries, heart, spleen, spinal cord, lungs, pancreas stomach, small intestines, colon, urinary bladder, prostate, eyes with optic nerve, pituitary, marrow of the femur, rib junction, mesenteric lymph node, mammary tissue, skin, sciatic nerve, muscle, uterus, gall bladder, and any gross lesion of the 3000 ppm group and the control group were examined histologically and no clear or meaningful changes were noted.

Dose and end point used for MRL derivation:

NOAEL  LOAEL

75 mg/kg/day (3,000 ppm dietary concentration)

Uncertainty Factors used in MRL derivation:

[X] 10 for human variability

[X] 10 for extrapolation of from animals to humans

$$75 \text{ mg/kg/day} \div 100 = 0.75 \text{ mg/kg/day}$$

Was a conversion used from num in food or water to a mg/body weight dose? If so, explain:

The doses of diisopropyl methylphosphonate that the dogs received in the diet (1,500, 1,500, and 3,000 ppm) were calculated using the recommended reference value of 0.025 kg food/kg body weight/day as follows:

$$(3,000 \text{ ppm}) (0.025 \text{ kg/kg/day}) = 75 \text{ mg/kg/day}$$

Other additional studies or pertinent information which lend support to this MRL:

Although this study (Hart 1980) did not identify an effect level, the NOAEL in this study is below the lowest-Observable-effect level (LOAEL) found in all studies examining the toxicity of diisopropyl methylphosphonate. The LOAEL for diisopropyl methylphosphonate is 345 mg/kg/day for male mink, and 455 mg/kg/day for female mink (average 400 mg/kg/day), doses at which statistically significant decreases in plasma (butyrylcholinesterase) but not RBC cholinesterase (acetylcholinesterase) activity was observed (Bucci et al. 1994). No effects were observed at 63 mg/kg/day in males and 82 mg/kg/day in females (average 73 mg/kg/day). A decrease in plasma cholinesterase activity is considered to be a marker of exposure rather than a marker of effect, while decreases in RBC acetylcholinesterase activity is thought to reflect decreases in brain acetylcholinesterase activity and is considered adverse.

Adverse effects, shortened RBC survival, increased Heinz body formation, increased number of reticulocytes, and reduced blood cell counts were observed at 747 mg/kg/day in males and 907 mg/kg/day in females (average 827 mg/kg/day). Although not statistically significant, the number of Heinz bodies was increased relative to controls at 400 mg/kg/day. The observed effects are consistent with a direct effect on RBC decreasing survival of the cells.

Metabolism studies in mink (Bucci et al. 1992) indicate that diisopropyl methylphosphonate is readily absorbed, metabolized to isopropylmethyl phosphonate, and excreted in the urine. After comparison with a metabolism study in dogs (Hart 1976), Bucci et al. (1992) concluded that mink handled the compound in a similar manner to dogs providing support for the use of mink when estimating risk to humans following diisopropyl methylphosphonate exposure. The metabolism study in mink (Bucci et al 1992) suggested that at high doses ( $\geq 270$  mg/kg) the principal pathway of metabolism of diisopropyl methylphosphonate is saturable. In the metabolism study the animals were treated by gavage; therefore, the dose rates in the gavage and dietary dosing studies may not be directly comparable. Further metabolism studies using dietary treatment are required to determine if the metabolism of diisopropyl methylphosphonate was saturated at the high doses ( $\geq 827$  mg/kg/day) at which adverse effects on RBC were observed.

The Environmental Protection Agency derived a reference dose (RfD) of 0.08 mg/kg/day based on the same NOAEL from the Hart (1980) study. The RfD, however, utilized an additional uncertainty factor of 10 to extrapolate to chronic exposure.

**MINIMAL RISK LEVEL WORKSHEET**

Chemical Name: Diisopropyl Methylphosphonate

CAS Number: 144-75-6

Date: March 1998

Profile Status: Draft 2 - Postpublic Comment

Route: [ ] Inhalation [X] Oral

Duration: [ ] Acute [ ] Intermediate [X] Chronic

Graph Key: 45

Species : Mink

Minimal Risk Level: 0.6 mg/kg/day

Reference: Bucci et al. 1997

Experimental design: In a 2-generation reproductive study, Ranch Wild mink received 0, 16, 45, or 262 mg/kg/day (males) or 0, 20, 57, or 330 mg/kg/day (females) diisopropyl methylphosphonate in the diet. These dosages were calculated for F<sub>1</sub> animals by the study authors using actual concentrations in the feed of 0, 168, 490, and 2,774 ppm diisopropyl methylphosphonate. F<sub>1</sub> generation females were treated for up to 13 months, while other generations and F<sub>1</sub> males were treated for up to 8 months. Two groups of control animals were used. Animals were observed twice/day for clinical signs and were weighed weekly. In addition to standard examinations of body and organ weights, hematology, clinical chemistry, parental animals from both generations and representative kits underwent gross histopathological examination, which included reproductive organs in males and gross lesions and developmental defects in the kits. Reproductive parameters examined were live kits/litter, litter weight at birth and 28 days, and sex ratio. Ovarian follicles were also counted in high-dose females.

Effects noted in study and corresponding doses:

No effects were observed in F<sub>1</sub> females at 57 mg/kg/day that were attributable to diisopropyl methylphosphonate after 13 months of exposure. Although there was no significant difference in food consumption or body weight in either generation compared to controls, the F<sub>1</sub> females consumed almost 50% more feed than the F<sub>0</sub> females, but this may have resulted from feed wastage. There was no treatment related change in litter size, percentage live births, kit weight or sex distribution in either generation. However, at 330 mg/kg/day, Heinz body counts were increased in F<sub>1</sub> females after 6 and 13 months and high-dose males of this generation had increased Heinz body counts. There was a 31% decrease in plasma cholinesterase in animals fed 330 mg/kg/day for 13 months, but this is not considered to be biologically significant. There was also a significant increase in ovarian follicles among animals at this concentration (the only level examined). However, because treated dams of both generations produced as many offspring as the control animals, the biological significance of these findings is unclear.

This study is supported by intermediate-duration NOAELs for hematological effects of 75 mg/kg/day in dogs (Hart 1980) and 73 mg/kg/day in mink (Bucci et al. 1994). In the Bucci et al. (1997) mink study, the next highest level, 262 or 330 mg/kg/day, produced hematological changes that included increased Heinz body counts, reticulocytes, mean cell volume, and decreased red blood cell counts.

Dose and end point used for MRL derivation:

[x] NOAEL [ ] LOAEL

57 mg/kg/day (490 ppm dietary concentration)

Uncertainty Factors used in MRL derivation:

[X] 10 for human variability

[X] 10 for extrapolation of from animals to humans

57 mg/kg/day ÷ 100 = 0.57 mg/kg/day

Was a conversion used from ppm in food or water to a m&odv weight dose? If so, explain:

The doses were calculated by the study authors.

Other additional studies or pertinent information which lend support to this MRL:

The critical study identified a NOAEL that was accompanied by a LOAEL. This study is supported by intermediate-duration NOAELs for hematological effects of 75 mg/kg/day in a dog study (Hart 1980) and 73 mg/kg/day in a mink study (Bucci et al. 1994). In the Hart (1980) study, beagle dogs received 0, 4, 38, or 75 mg/kg/day diisopropyl methylphosphonate in the diet for 90 days. No toxic effects, including hematological effects, were noted over the study period. In the Bucci et al. (1997) 2-generation reproductive study in mink, animals were fed 0, 16, 45, or 262 mg/kg/day (males) or 0, 20, 57, or 330 mg/kg/day (females) diisopropyl methylphosphonate for up to 12 months. While no hematological effects were found at the low- or mid-dose in either sex, high-dose males had increased Heinz body counts and high-dose females had decreased RBC counts and increased MCV, reticulocytes, and Heinz bodies. No hematological changes were found in the F<sub>1</sub> kits after 11 weeks, but F<sub>2</sub> high-dose male F<sub>1</sub> kits had significantly decreased RBC counts at 6 weeks of age. High-dose F<sub>0</sub> females also had significant increases in absolute and relative spleen weight and spleen-to-brain weight ratio. There was a treatment-related hematopoietic cell proliferation apparent in the spleen of these animals; the spleen showed evidence of RBC replacement. The next highest level, 262 or 330 mg/kg/day in males and females, respectively, produced hematological changes that included increased Heinz body counts, reticulocytes, mean cell volume, and decreased red blood cell counts.

## APPENDIX B

### USER'S GUIDE

#### Chapter 1

##### Public Health Statement

This chapter of the profile is a health effects summary written in non-technical language. Its intended audience is the general public especially people living in the vicinity of a hazardous waste site or chemical release. If the Public Health Statement were removed from the rest of the document, it would still communicate to the lay public essential information about the chemical.

The major headings in the Public Health Statement are useful to find specific topics of concern. The topics are written in a question and answer format. The answer to each question includes a sentence that will direct the reader to chapters in the profile that will provide more information on the given topic.

#### Chapter 2

##### Tables and Figures for Levels of Significant Exposure (LSE)

Tables (2-1,2-2, and 2-3) and figures (2- 1 and 2-2) are used to summarize health effects and illustrate graphically levels of exposure associated with those effects. These levels cover health effects observed at increasing dose concentrations and durations, differences in response by species, minimal risk levels (MRLs) to humans for noncancer end points, and EPA's estimated range associated with an upper- bound individual lifetime cancer risk of 1 in 10,000 to 1 in 10,000,000. Use the LSE tables and figures for a quick review of the health effects and to locate data for a specific exposure scenario. The LSE tables and figures should always be used in conjunction with the text. All entries in these tables and figures represent studies that provide reliable, quantitative estimates of No-Observed-Adverse- Effect Levels (NOAELs), Lowest-Observed-Adverse-Effect Levels (LOAELs), or Cancer Effect Levels (CELs).

The legends presented below demonstrate the application of these tables and figures. Representative examples of LSE Table 2-1 and Figure 2-1 are shown. The numbers in the left column of the legends correspond to the numbers in the example table and figure.

#### LEGEND

##### See LSE Table 2-1

- (1) Route of Exposure One of the first considerations when reviewing the toxicity of a substance using these tables and figures should be the relevant and appropriate route of exposure. When sufficient data exists, three LSE tables and two LSE figures are presented in the document. The three LSE tables present data on the three principal routes of exposure, i.e., inhalation, oral, and dermal (LSE Table 2- 1, 2-2, and 2-3, respectively). LSE figures are limited to the inhalation (LSE Figure 2-1) and oral (LSE Figure 2-2) routes. Not all substances will have data on each route of exposure and will not therefore have all five of the tables and figures.
- (2) Exposure Period Three exposure periods - acute (less than 15 days), intermediate (15-364 days), and chronic (365 days or more) are presented within each relevant route of exposure. In this example, an inhalation study of intermediate exposure duration is reported. For quick reference to health effects occurring from a known length of exposure, locate the applicable exposure period within the LSE table and figure.

- (3) Health Effect The major categories of health effects included in LSE tables and figures are death, systemic, immunological, neurological, developmental, reproductive, and cancer. NOAELs and LOAELs can be reported in the tables and figures for all effects but cancer. Systemic effects are further defined in the “System” column of the LSE table (see key number 18).
- (4) Key to Figure Each key number in the LSE table links study information to one or more data points using the same key number in the corresponding LSE figure. In this example, the study represented by key number 18 has been used to derive a NOAEL and a Less Serious LOAEL (also see the 2 “18r” data points in Figure 2-1).
- (5) Species The test species, whether animal or human, are identified in this column. Section 2.5, “Relevance to Public Health,” covers the relevance of animal data to human toxicity and Section 2.3, “Toxicokinetics,” contains any available information on comparative toxicokinetics. Although NOAELs and LOAELs are species specific, the levels are extrapolated to equivalent human doses to derive an MRL.
- (6) Exposure Frequency/Duration The duration of the study and the weekly and daily exposure regimen are provided in this column. This permits comparison of NOAELs and LOAELs from different studies. In this case (key number 18), rats were exposed to 1,1,2,2-tetrachloroethane via inhalation for 6 hours per day, 5 days per week, for 3 weeks. For a more complete review of the dosing regimen refer to the appropriate sections of the text or the original reference paper, i.e., Nitschke et al. 1981.
- (7) System This column further defines the systemic effects. These systems include: respiratory, cardiovascular, gastrointestinal, hematological, musculoskeletal, hepatic, renal, and dermal/ocular. “Other” refers to any systemic effect (e.g., a decrease in body weight) not covered in these systems. In the example of key number 18, 1 systemic effect (respiratory) was investigated.
- (8) NOAEL A No-Observed-Adverse-Effect Level (NOAEL) is the highest exposure level at which no harmful effects were seen in the organ system studied. Key number 18 reports a NOAEL of 3 ppm for the respiratory system which was used to derive an intermediate exposure, inhalation MRL of 0.005 ppm (see footnote “b”).
- (9) LOAEL A Lowest-Observed-Adverse-Effect Level (LOAEL) is the lowest dose used in the study that caused a harmful health effect. LOAELs have been classified into “Less Serious” and “Serious” effects. These distinctions help readers identify the levels of exposure at which adverse health effects first appear and the gradation of effects with increasing dose. A brief description of the specific endpoint used to quantify the adverse effect accompanies the LOAEL. The respiratory effect reported in key number 18 (hyperplasia) is a Less serious LOAEL of 10 ppm. MRLs are not derived from Serious LOAELs.
- (10) Reference The complete reference citation is given in chapter 8 of the profile.
- (11) CEL A Cancer Effect Level (CEL) is the lowest exposure level associated with the onset of carcinogenesis in experimental or epidemiologic studies. CELs are always considered serious effects. The LSE tables and figures do not contain NOAELs for cancer, but the text may report doses not causing measurable cancer increases.

- (12) Footnotes Explanations of abbreviations or reference notes for data in the LSE tables are found in the footnotes. Footnote “b” indicates the NOAEL of 3 ppm in key number 18 was used to derive an MRL of 0.005 ppm.

## LEGEND

### See Figure 2-1

LSE figures graphically illustrate the data presented in the corresponding LSE tables. Figures help the reader quickly compare health effects according to exposure concentrations for particular exposure periods.

- (13) Exposure Period The same exposure periods appear as in the LSE table. In this example, health effects observed within the intermediate and chronic exposure periods are illustrated.
- (14) Health Effect These are the categories of health effects for which reliable quantitative data exists. The same health effects appear in the LSE table.
- (15) Levels of Exposure concentrations or doses for each health effect in the LSE tables are graphically displayed in the LSE figures. Exposure concentration or dose is measured on the log scale “y” axis. Inhalation exposure is reported in mg/m<sup>3</sup> or ppm and oral exposure is reported in mg/kg/day.
- (16) NOAEL In this example, 18r NOAEL is the critical endpoint for which an intermediate inhalation exposure MRL is based. As you can see from the LSE figure key, the open-circle symbol indicates to a NOAEL for the test species-rat. The key number 18 corresponds to the entry in the LSE table. The dashed descending arrow indicates the extrapolation from the exposure level of 3 ppm (see entry 18 in the Table) to the MRL of 0.005 ppm (see footnote “b” in the LSE table).
- (17) CEL Key number 38r is 1 of 3 studies for which Cancer Effect Levels were derived. The diamond symbol refers to a Cancer Effect Level for the test species-mouse. The number 38 corresponds to the entry in the LSE table.
- (18) Estimated Upper-Bound Human Cancer Risk Levels This is the range associated with the upper-bound for lifetime cancer risk of 1 in 10,000 to 1 in 10,000,000 These risk levels are derived from the EPA’s Human Health Assessment Group’s upper-bound estimates of the slope of the cancer dose response curve at low dose levels (q<sub>1</sub>\*).
- (19) Key to LSE Figure The Key explains the abbreviations and symbols used in the figure.



**SAMPLE**

1 →

**TABLE 2-1. Levels of Significant Exposure to [Chemical x] – Inhalation**

Key to figure <sup>a</sup>	Species	Exposure frequency/ duration	System	NOAEL (ppm)	LOAEL (effect)		Reference
					Less serious (ppm)		
<b>INTERMEDIATE EXPOSURE</b>							
2 →	5	6	7	8	9	10	
3 →	Systemic	↓	↓	↓	↓	↓	↓
4 →	18	Rat	13 wk 5d/wk 6hr/d	Resp	3 <sup>b</sup>	10 (hyperplasia)	Nitschke et al. 1981
<b>CHRONIC EXPOSURE</b>							
						11	
	Cancer					↓	
38	Rat	18 mo 5d/wk 7hr/d				20	(CEL, multiple organs) Wong et al. 1982
39	Rat	89–104 wk 5d/wk 6hr/d				10	(CEL, lung tumors, nasal tumors) NTP 1982
40	Mouse	79–103 wk 5d/wk 6hr/d				10	(CEL, lung tumors, hemangiosarcomas) NTP 1982

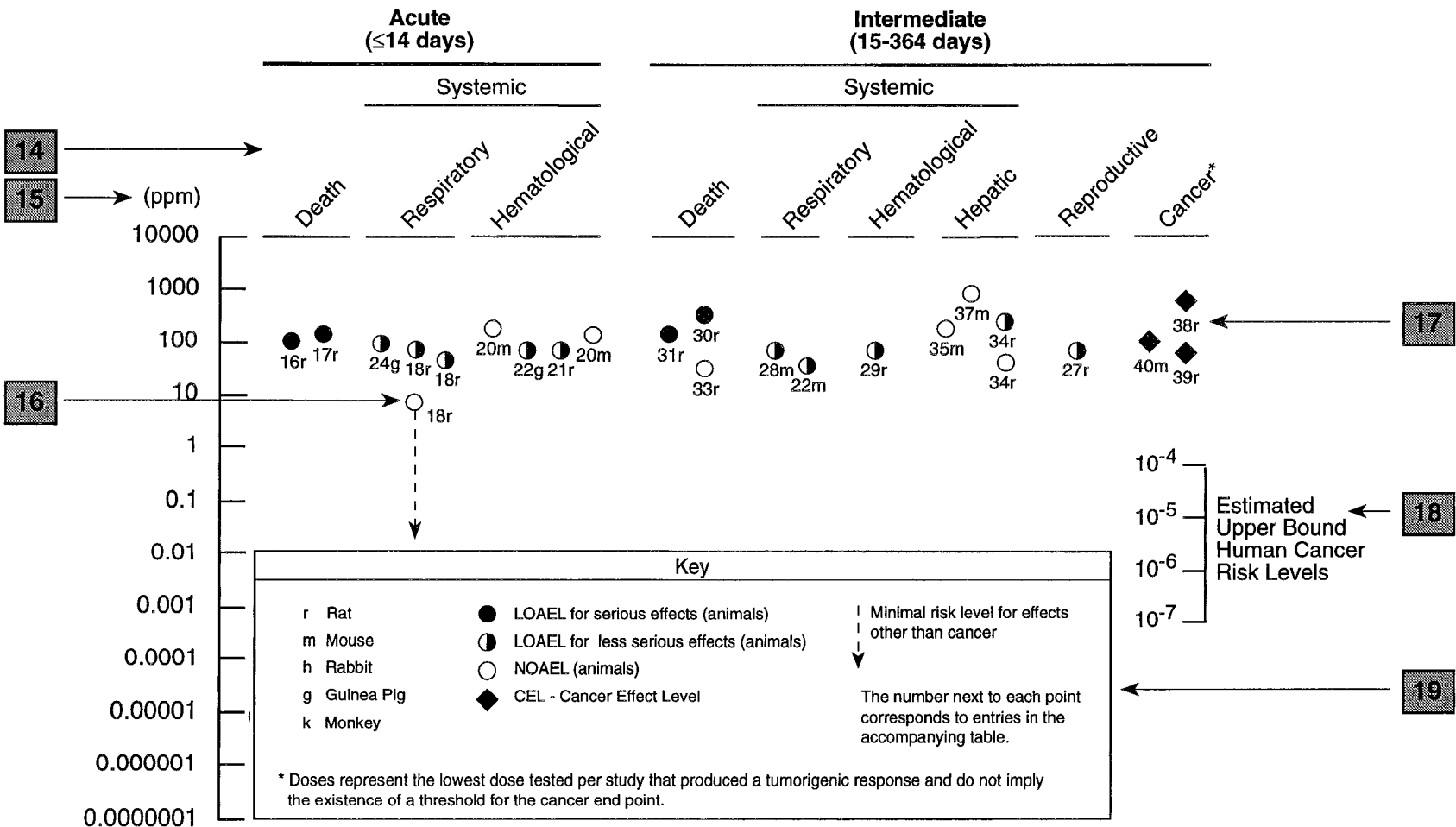
12 →

<sup>a</sup> The number corresponds to entries in Figure 2-1.

<sup>b</sup> an uncertainty factor of 100 (10 for extrapolation from animal to humans, 10 for human variability).

**SAMPLE**

**13** → **Figure 2-1. Levels of Significant Exposure to [Chemical X] – Inhalation**



## Chapter 2 (Section 2.5)

### Relevance to Public Health

The Relevance to Public Health section provides a health effects summary based on evaluations of existing toxicologic, epidemiologic, and toxicokinetic information. This summary is designed to present interpretive, weight-of-evidence discussions for human health end points by addressing the following questions.

- 1 . What effects are known to occur in humans?
- 2 . What effects observed in animals are likely to be of concern to humans?
- 3 . What exposure conditions are likely to be of concern to humans, especially around hazardous waste sites?

The section covers end points in the same order they appear within the Discussion of Health Effects by Route of Exposure section, by route (inhalation, oral, dermal) and within route by effect. Human data are presented first, then animal data. Both are organized by duration (acute, intermediate, chronic). In vitro data and data from parenteral routes (intramuscular, intravenous, subcutaneous, etc.) are also considered in this section. If data are located in the scientific literature, a table of genotoxicity information is included.

The carcinogenic potential of the profiled substance is qualitatively evaluated, when appropriate, using existing toxicokinetic, genotoxic, and carcinogenic data. ATSDR does not currently assess cancer potency or perform cancer risk assessments. Minimal risk levels (MRLs) for noncancer end points (if derived) and the end points from which they were derived are indicated and discussed.

Limitations to existing scientific literature that prevent a satisfactory evaluation of the relevance to public health are identified in the Data Needs section.

### Interpretation of Minimal Risk Levels

Where sufficient toxicologic information is available, we have derived minimal risk levels (MRLs) for inhalation and oral routes of entry at each duration of exposure (acute, intermediate, and chronic). These MRLs are not meant to support regulatory action; but to acquaint health professionals with exposure levels at which adverse health effects are not expected to occur in humans. They should help physicians and public health officials determine the safety of a community living near a chemical emission, given the concentration of a contaminant in air or the estimated daily dose in water. MRLs are based largely on toxicological studies in animals and on reports of human occupational exposure.

MRL users should be familiar with the toxicologic information on which the number is based. Chapter 2.5, "Relevance to Public Health," contains basic information known about the substance. Other sections such as 2.7, "Interactions with Other Substances," and 2.8, "Populations that are Unusually Susceptible" provide important supplemental information.

MRL users should also understand the MRL derivation methodology. MRLs are derived using a modified version of the risk assessment methodology the Environmental Protection Agency (EPA) provides (Barnes and Dourson 1988) to determine reference doses for lifetime exposure (RfDs).

To derive an MRL, ATSDR generally selects the most sensitive endpoint which, in its best judgement, represents the most sensitive human health effect for a given exposure route and duration. ATSDR cannot

make this judgement or derive an MRL unless information (quantitative or qualitative) is available for all potential systemic, neurological, and developmental effects. If this information and reliable quantitative data on the chosen endpoint are available, ATSDR derives an MRL using the most sensitive species (when information from multiple species is available) with the highest NOAEL that does not exceed any adverse effect levels. When a NOAEL is not available, a lowest-observed-adverse-effect level (LOAEL) can be used to derive an MRL, and an uncertainty factor (UF) of 10 must be employed. Additional uncertainty factors of 10 must be used both for human variability to protect sensitive subpopulations (people who are most susceptible to the health effects caused by the substance) and for interspecies variability (extrapolation from animals to humans). In deriving an MRL, these individual uncertainty factors are multiplied together. The product is then divided into the inhalation concentration or oral dosage selected from the study. Uncertainty factors used in developing a substance-specific MRL are provided in the footnotes of the LSE Tables.



**APPENDIX C****ACRONYMS, ABBREVIATIONS, AND SYMBOLS**

ACGIH	American Conference of Governmental Industrial Hygienists
ADME	Absorption, Distribution, Metabolism, and Excretion
AML	acute myeloid leukemia
atm	atmosphere
ATSDR	Agency for Toxic Substances and Disease Registry
BCF	bioconcentration factor
BEI	Biological Exposure Index
BSC	Board of Scientific Counselors
C	Centigrade
CDC	Centers for Disease Control
CEL	Cancer Effect Level
CERCLA	Comprehensive Environmental Response, Compensation, and Liability Act
CFR	Code of Federal Regulations
Ci	curie
CLP	Contract Laboratory Program
cm	centimeter
CML	chronic myeloid leukemia
CNS	central nervous system
d	day
DHEW	Department of Health, Education, and Welfare
DHHS	Department of Health and Human Services
DOL	Department of Labor
ECG	electrocardiogram
EEG	electroencephalogram
EPA	Environmental Protection Agency
EKG	see ECG
F	Fahrenheit
F <sub>1</sub>	first filial generation
FAO	Food and Agricultural Organization of the United Nations
FEMA	Federal Emergency Management Agency
FIFRA	Federal Insecticide, Fungicide, and Rodenticide Act
fpm	feet per minute
ft	foot
FR	<i>Federal Register</i>
g	gram
GC	gas chromatography
gen	generation
HPLC	high-performance liquid chromatography
hr	hour
IDLH	Immediately Dangerous to Life and Health
IARC	International Agency for Research on Cancer
ILO	International Labor Organization
in	inch
K <sub>d</sub>	adsorption ratio

## APPENDIX C

kg	kilogram
kkg	metric ton
$K_{oc}$	organic carbon partition coefficient
$K_{ow}$	octanol-water partition coefficient
L	liter
LC	liquid chromatography
$LC_{Lo}$	lethal concentration, low
$LC_{50}$	lethal concentration, 50% kill
$LD_{Lo}$	lethal dose, low
$LD_{50}$	lethal dose, 50% kill
LOAEL	lowest-observed-adverse-effect level
LSE	Levels of Significant Exposure
m	meter
MA	<u>trans,trans</u> -muconic acid
mCi	millicurie
mg	milligram
min	minute
mL	milliliter
mm	millimeter
mm Hg	millimeters of mercury
mmol	millimole
mo	month
mppcf	millions of particles per cubic foot
MRL	Minimal Risk Level
MS	mass spectrometry
NCE	normochromatic erythrocytes
NIEHS	National Institute of Environmental Health Sciences
NIOSH	National Institute for Occupational Safety and Health
NIOSHTIC	NIOSH's Computerized Information Retrieval System
ng	nanogram
nm	nanometer
NHANES	National Health and Nutrition Examination Survey
nmol	nanomole
NOAEL	no-observed-adverse-effect level
NOES	National Occupational Exposure Survey
NOHS	National Occupational Hazard Survey
NPL	National Priorities List
NRC	National Research Council
NTIS	National Technical Information Service
NTP	National Toxicology Program
OSHA	Occupational Safety and Health Administration
PEL	permissible exposure limit
PCE	polychromatic erythrocytes
pg	picogram
pmol	picomole
PHS	Public Health Service
PMR	proportionate mortality ratio
ppb	parts per billion
ppm	parts per million

## APPENDIX C

ppt	parts per trillion
REL	recommended exposure limit
RfD	Reference Dose
RTECS	Registry of Toxic Effects of Chemical Substances
sec	second
SCE	sister chromatid exchange
SIC	Standard Industrial Classification
SMR	standard mortality ratio
STEL	short term exposure limit
STORET	STORAGE and RETRIEVAL
TLV	threshold limit value
TSCA	Toxic Substances Control Act
TRI	Toxics Release Inventory
TWA	time-weighted average
UMDNJ	University of Medicine and Dentistry New Jersey
U.S.	United States
UF	uncertainty factor
yr	year
WHO	World Health Organization
wk	week
>	greater than
≥	greater than or equal to
=	equal to
<	less than
≤	less than or equal to
%	percent
α	alpha
β	beta
δ	delta
γ	gamma
μm	micrometer
μg	microgram



