3. HEALTH EFFECTS

3.1 INTRODUCTION

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

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

3.2 DISCUSSION OF HEALTH EFFECTS BY ROUTE OF EXPOSURE

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

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

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

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

Levels of exposure associated with carcinogenic effects (Cancer Effect Levels, CELs) of heptachlor and heptachlor epoxide are indicated in Table 3-1 and Figure 3-1.

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.

3.2.1 Inhalation Exposure

3.2.1.1 Death

Limited information exists regarding exposure to heptachlor or heptachlor epoxide and mortality. One of the four reports available studied pesticide manufacturers (Wang and MacMahon 1979b), whereas the other three examined pesticide applicators (Blair et al. 1983; MacMahon et al. 1988; Shindell and Associates 1981; Wang and MacMahon 1979a). Exposure data were not available in any of these studies and none of them provided specific information for heptachlor or heptachlor epoxide. An occupational mortality study on workers employed in the manufacture of heptachlor and other chlorinated hydrocarbon pesticides for at least 3 months between 1952 and 1979 revealed no pattern of disease or medical condition that indicated that persons were at greater risk of adverse outcome than the general population (Shindell and Associates 1981). The only significant finding observed in workers employed at two facilities manufacturing chlordane or heptachlor and endrin was an excess of deaths from cerebrovascular disease, which was unrelated to duration of exposure or latency, and occurred exclusively after termination of employment (Wang and MacMahon 1979b). In professional pesticide applicators at three U.S. companies who were employed for 3 months or longer between 1967 and 1976, only deaths due to bladder cancer were significantly elevated in the applicators as a whole (Wang and MacMahon 1979a). In a separate analysis conducted for persons ever holding jobs as “termite control operators,” a group

more likely to be exposed to chlordane and heptachlor, bladder cancer was elevated in both the termite control operators and the group comprising the rest of the applicators. However, since confidence intervals were not provided, it is unknown whether this increase was statistically significant. A follow-up analysis of the same cohort extending to the end of 1984 reported that deaths due to cancer of the lung was the only outcome significantly elevated in the group as a whole (MacMahon et al. 1988). Separate analyses, as done in the earlier study, revealed that deaths due to lung cancer were not significantly elevated in the group with the highest likelihood of exposure to chlordane and heptachlor. In the last study of licensed male pesticide applicators in Florida, the standardized mortality ratio (SMR) for all causes of death was 103, but increased SMRs, although not statistically significant, were seen for leukemia, cancers of the brain, and lung cancer (Blair et al. 1983). The increased SMRs for brain and leukemia were based on small numbers. Mortality from lung cancer was related to years licensed, but could not be attributed to any specific pesticide due to lack of information on frequency and intensity of exposures. Furthermore, information on smoking was not available. In conclusion, the information available is insufficient to determine whether there is an association between exposure of workers to heptachlor or heptachlor epoxide and mortality.

No studies were located regarding death in animals after inhalation exposure to heptachlor or heptachlor epoxide.

3.2.1.2 Systemic Effects

No studies were located regarding respiratory, cardiovascular, gastrointestinal, musculoskeletal, hepatic, renal, dermal, or ocular effects in humans or animals after inhalation exposure to heptachlor or heptachlor epoxide.

Hematological Effects. The available data on potential hematological effects following inhalation exposure to heptachlor or heptachlor epoxide are limited to a case-control study of exterminators, gardeners, and agricultural workers exposed to several organochlorine pesticides (heptachlor among them) (Wang and Grufferman 1981). No dose-dependent causal relationship between exposure to several organochlorine pesticides and deaths from aplastic anemia was found.

No studies were located regarding hematological effects in animals after inhalation exposure to heptachlor or heptachlor epoxide.

No studies were located regarding the following effects in humans or animals after inhalation exposure to heptachlor or heptachlor epoxide:

3.2.1.3 Immunological and Lymphoreticular Effects

3.2.1.4 Neurological Effects

3.2.1.5 Reproductive Effects

3.2.1.6 Developmental Effects

3.2.1.7 Cancer

Several studies have examined the possible association between cancer and environmental and/or occupational exposure to chlordane and heptachlor (Epstein and Ozonoff 1987; MacMahon et al. 1988; Shindell and Associates 1981; Wang and MacMahon 1979a, 1979b). Several occupational cohorts showed that workers who were involved in the manufacture of chlordane and heptachlor did not have a significant increase in death from any type of cancer (MacMahon et al. 1988; Shindell and Associates 1981; Wang and MacMahon 1979a, 1979b). These occupational studies are presumed to reflect primarily inhalation exposure, with some concomitant dermal exposure. Among workers at pesticide manufacturing facilities, the SMR for bladder cancer was of borderline statistical significance (Wang and MacMahon 1979a). A follow-up study identified an increase in lung cancer, but the SMR for deaths from lung cancer in the group with the highest chance of exposure was not significant (MacMahon et al. 1988). No information on cigarette smoking was obtained from the participants. A retrospective mortality study conducted on male workers engaged in chlordane, heptachlor, and endrin manufacture for at least 3 months also showed a slight excess of lung cancer compared to the general U.S. population, but the increase was not statistically significant (Wang and MacMahon 1979b). Leukemia was associated with exposure to chlordane and heptachlor following home termiticide use (Epstein and Ozonoff 1987). An increased risk of prostate cancer was found among Hispanic farm workers potentially exposed to heptachlor (Mills and Yang 2003). Among the workers employed in counties with the highest heptachlor use, the odds ratio (adjusted for age, surrogates for exposure initiation, and duration) was 2.01 (95% confidence interval of 1.12–3.60).

It is difficult to determine from these studies whether or not exposure to heptachlor or heptachlor epoxide causes cancer. Although some studies suggest an association between exposure and cancer, other studies have not found significant associations, and there were many limitations among the studies. Limitations include the lack of quantitative exposure information, concomitant exposure to other chemicals, lack of

control measures for confounding factors, lack of information on diet, smoking habits, and liver function, and lack of complete occupational history, including other potential causal factors such as genetic disposition or immunologic disorders.

No studies were located regarding cancer in animals after inhalation exposure to heptachlor or heptachlor epoxide.

3.2.2 Oral Exposure

3.2.2.1 Death

No studies were located regarding death in humans after oral exposure to heptachlor or heptachlor epoxide.

Acute oral LD_{50} values for heptachlor in rodents (rats, mice, hamsters, and guinea pigs) and rabbits range from 40 to 2,302 mg/kg (purity ranging from unspecified to 99.9%) (Ben-Dyke et al. 1970; Berman et al. 1995; Eisler 1968; Gaines 1969; Gak et al. 1976; Lehman 1951; Podowski et al. 1979; Sperling et al. 1972; Sun 1972). The differences in the purity of the administered heptachlor may have influenced lethality. Pure heptachlor appears to be more lethal than technical-grade heptachlor (Berman et al. 1995; Podowski et al. 1979). Acute oral LD_{50} values for heptachlor epoxide in rodents (rats and mice) and rabbits range from 39 to 144 mg/kg (Eisler 1968; Podowski et al. 1979; Sperling et al. 1972).

Heptachlor can be converted to its photoisomer, photoheptachlor, in the presence of sunlight or ultraviolet light. This photolysis can take place on plant leaves. Photoheptachlor was found to be more toxic to rats than heptachlor or heptachlor epoxide; the LD_{50} for photoheptachlor was 3.8 mg/kg (Podowski et al. 1979).

The results of the non-LD_{50} studies indicate that the lethal dose decreases with duration of exposure. A single gavage dose of up to 129 mg/kg technical-grade heptachlor (73% heptachlor, 26% chlordane) did not result in mortality in rats (Berman et al. 1995). However, repeated doses of 23 or 69 mg/kg/day of technical-grade heptachlor resulted in 100% mortality after 6 or 3 doses, respectively (Berman et al. 1995; Moser et al. 1995). Increases in mortality were also observed in male rats exposed to 30 mg/kg/day and female rats exposed to 15 mg/kg/day technical-grade heptachlor (73% heptachlor, 22% chlordane, 5% nonachlor) in the diet for 6 weeks, followed by a 2-week period of observation (NCI 1977). No deaths were observed at 14 mg/kg/day in males and 7.6 mg/kg/day in females.

Similar lethal doses were observed in mice and mink exposed to heptachlor in the diet for intermediate durations. The lethal doses were 14 mg/kg/day for mice fed diets containing technical-grade heptachlor (73% heptachlor, 22% chlordane, 5% nonachlor) for 6 weeks, followed by a 2-week period of observation (NCI 1977) and 6.19 mg/kg/day for mink fed technical-grade heptachlor (72% heptachlor) for 28 days (Aulerich et al. 1990) or 1.7 mg/kg/day for 181 days (Crum et al. 1993). No alterations in mortality were observed in mice exposed to doses of 5.2 mg/kg/day or in mink at doses of 5.67 mg/kg/day. In chronic-duration studies, no statistically significant alterations in survival were observed in male or female mice exposed to 2.4 or 3.0 mg/kg/day, respectively, technical-grade heptachlor in the diet for 80 weeks (NCI 1977).

All reliable LD_{50} values and all reliable LOAEL values for death in each species and duration category are recorded in Table 3-1 and plotted in Figure 3-1.

3.2.2.2 Systemic Effects

No studies were located regarding respiratory, musculoskeletal, or dermal effects in humans or animals after oral exposure to heptachlor or heptachlor epoxide.

The highest NOAEL values and all reliable LOAEL values for systemic effects in each species and duration category are recorded in Table 3-1 and plotted in Figure 3-1.

Cardiovascular Effects. The information regarding cardiovascular effects in humans associated with heptachlor and heptachlor epoxide exposure is limited to a study that found statistically higher heptachlor epoxide serum levels among individuals with moderate to severe arterosclerosis (Pines et al. 1986). This report cannot be construed as showing a causal relationship between heptachlor epoxide exposure and arteriosclerosis because no adjustments for other risk factors were made.

Animal data are limited to a study that found increases in relative heart weight (statistical significance not reported) in female rats exposed to heptachlor in the diet 5 days/week for 4 weeks (Enan et al. 1982). The investigators noted that 10 mg/kg heptachlor was added to the diet; it is not known if this is a dietary concentration or dose. The biological significance of this effect is not known, particularly since no information on body weight changes or food intake were provided and a histological examination was not performed.

Table 3-1 Levels of Significant Exposure to Heptachlor and Heptachlor Epoxide - Oral

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference Chemical Form	Comments
ACUTE EXPOSURE								
Death								
1	Rat (Fischer- 344)	once (GO)				230 F (LD50)	Berman et al. 1995 heptachlor	
2	Rat (Sherman)	1 d 1 x/d (GO)				100[b] M (LD50) 162 F (LD50)	Gaines 1969 heptachlor	
3	Rat (NS)	once (GO)				105 (LD50)	Gak et al. 1976 heptachlor	
4	Rat (Fischer- 344)	Gd 6-15 (GO)				12 F (38% mortality in pregnant rats)	Narotsky et al. 1995 heptachlor	
5	Rat (NS)	once				71 M (LD50)	Podowski et al. 1979 heptachlor	
6	Rat (NS)	once				60 M (LD50)	Podowski et al. 1979 heptachlor epoxide	
7	Mouse (NS)	once (GO)				70 (LD50)	Gak et al. 1976 heptachlor	
8	Hamster (NS)	once (GO)				100 (LD50)	Gak et al. 1976 heptachlor	

Table 3-1 Levels of Significant Exposure to Heptachlor and Heptachlor Epoxide - Oral (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference Chemical Form	Comments
Systemic								
9	Rat (Fischer- 344)	14 d 1 x/d (GO)	Hepatic	2 F	7 F (hepatocytomegaly)		Berman et al. 1995 heptachlor	
10	Rat (Wistar)	once (GO)	Hepatic		60 F (increased serum ALT and aldolase levels, decreased liver ALT and aldolase levels, vacuolated cells, pyknotic nuclei)		Krampl 1971 heptachlor	
11	Rat (Wistar)	3, 7, or 14 d (GO)	Hepatic		7 F (decreased liver ALT and aldolase levels, increased serum ALT and aldolase levels; monocellular necrosis and vacuolar dystrophy)		Krampl 1971 heptachlor	
12	Rat (Wistar)	14 d (F)	Resp	5			Pelikan 1971 heptachlor	
			Cardio	5				
			Gastro		5 (slightly hyperemic stomach and intestinal wall)			
			Hepatic	5				
			Renal	5				
			Bd Wt	5				

Table 3-1 Levels of Significant Exposure to Heptachlor and Heptachlor Epoxide - Oral (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference Chemical Form	Comments
13	Mouse (albino)	11 d (W)	Endocr			87 F (atrophy in adrenal cortex)	Akay et al. 1982 heptachlor	
Immuno/ Lymphoret								
14	Rat (Fischer- 344)	once (GO)		69 F	129 F (necrotic lymphocytes in spleen and thymus)		Berman et al. 1995 heptachlor	Limited to examination of spleen and thymus.
15	Rat (Fischer- 344)	14 d 1 x/d (GO)		69 F			Berman et al. 1995 heptachlor	Limited to examination of spleen and thymus.
16	Rat (Wistar)	14 d (F)		5			Pelikan 1971 heptachlor	Limited to examination of spleen.
Neurological								
17	Rat (Fischer- 344)	once (GO)			7 F (excitability)		Moser et al. 1995 heptachlor	
18	Rat (Fischer- 344)	14 d 1 x/d (GO)		2 F	7 F (increased arousal)		Moser et al. 1995 heptachlor	
Reproductive								
19	Rat (Wistar)	14 d (GO)				1.8[c] F (decreased fertility; increased resorptions)	Amita Rani and Krishnakumari 1995 heptachlor	
20	Mouse (CD-1)	1 d 1 x/d (G)		15 M			Arnold et al. 1977 heptachlor	

Table 3-1 Levels of Significant Exposure to Heptachlor and Heptachlor Epoxide - Oral (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference Chemical Form	Comments
21	Mouse (ICR)	5 d (G)		8 M			Epstein et al. 1972 heptachlor epoxide	
22	Mouse (ICR)	5 d (G)		10 M			Epstein et al. 1972 heptachlor	
Developmental								
23	Rat (Fischer- 344)	Gd 6-19 (GO)			4.5 (decreased pup body weight at pnd 6)		Narotsky and Kavlock 1995 heptachlor	
24	Rat (Fischer- 344)	Gd 6-15 (GO)		5.1 F	6.8 F (decreased pup body weight)	9 F (pup mortality)	Narotsky et al. 1995 heptachlor	
25	Rat (Sprague-Dawley)	Gd 10-21 (GO)			4.2 F (decreased righting reflex in pups)	8.4 F (decreased pup survival)	Purkerson-Parker et al. 2001b heptachlor	
INTERMEDIATE EXPOSURE								
Death								
26	Rat (Osborne-Mendel)	6 wk (F)				30 M (2/5 male rats died) 15 F[b] (4/5 female rats died)	NCI 1977 heptachlor	
27	Mouse (B6C3F1)	6 wk (F)				14 M (5/5 males died)	NCI 1977 heptachlor	
28	Mink (NS)	28 d (F)				6.2 M (3/8 died)	Aulerich et al. 1990 heptachlor	

Table 3-1 Levels of Significant Exposure to Heptachlor and Heptachlor Epoxide - Oral (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference Chemical Form	Comments
29	Mink (NS)	181 d (F)				1.7 F (67% mortality)	Crum et al. 1993 heptachlor	
Systemic								
30	Rat (albino)	21 d (G)	Endocr	1 M			Akhtar et al. 1996 heptachlor	Measured thyroid hormone levels.
			Bd Wt	1 M				
31	Rat (Wistar)	28 d (F)	Resp	5			Pelikan 1971 heptachlor	
			Cardio	5				
			Gastro		5 (thin mucous secretion covering the stomach and intestine mucosa)			
			Hepatic		5 (steatosis, 21-23% increased relative liver weight)			
			Renal	5				
			Bd Wt	5				

Table 3-1 Levels of Significant Exposure to Heptachlor and Heptachlor Epoxide - Oral (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference Chemical Form	Comments
32	Mouse (NS)	10 wk (F)	Hepatic		9.3 (hepatitis, necrosis, granuloma, congestion)		Akay and Alp 1981 heptachlor	No incidence data or statistical analysis reported.
			Renal		37 (granuloma)			
			Bd Wt		9.3 (unspecified decrease in body weight)			
33	Mouse (albino)	26 d (W)	Endocr			87 F (lipid accumulation and extensive degeneration and fibrosis in adrenal cortex)	Akay et al. 1982 heptachlor	
34	Mouse (DDY)	180 d ad lib (W)	Hepatic		6.9 M (increased serum ALT activity levels and liver weight)		Izushi and Ogata 1990 heptachlor	No histological examination.
			Bd Wt	6.9 M				
			Metab	6.9 M				
35	Mouse (DDY)	92 d 2 x/wk (GO)	Hepatic		10 M (increased serum ALT, alkaline phosphatase, and triglyceride levels, liver triglyceride levels, and liver weight)		Izushi and Ogata 1990 heptachlor	No histological examination.
			Bd Wt	10 M				
			Metab	10 M				

Table 3-1 Levels of Significant Exposure to Heptachlor and Heptachlor Epoxide - Oral (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference Chemical Form	Comments
36	Mink (NS)	28 d (F)	Bd Wt	3.1 M	5.7 M (22% decrease in body weight)		Aulerich et al. 1990 heptachlor	
Immuno/ Lymphoret								
37	Rat (Wistar)	28 d (F)		5			Pelikan 1971 heptachlor	Limited to examination of spleen.
38	Mouse (NS)	10 wk (F)		19	37 (splenic fibrosis)		Akay and Alp 1981 heptachlor	No incidence data or statistical analysis reported.
39	Mink (NS)	28 d (F)		5.7 M	6.2 M (49% decrease in spleen/brain weight)		Aulerich et al. 1990 heptachlor	
Neurological								
40	Mouse (NS)	10 wk (F)		9.3		19 F (difficulty standing, walking, and righting)	Akay and Alp 1981 heptachlor	
41	Mink (NS)	28 d (F)		5.7 M	6.2 M (clinical signs of hyperexcitability and incoordination)		Aulerich et al. 1990 heptachlor	
42	Mink (NS)	181 d (F)		1 F		1.7 F (hyperexcitability and seizures)	Crum et al. 1993 heptachlor	

Table 3-1 Levels of Significant Exposure to Heptachlor and Heptachlor Epoxide - Oral (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference Chemical Form	Comments
Reproductive								
43	Rat (Wistar)	70 d (GO)			0.65 M (decreased epididymal sperm count)	0.65 M (increased resorptions)	Amita Rani and Krishnakumari 1995 heptachlor	
44	Mouse (NS)	10 wk (F)				9.3 (100% infertility)	Akay and Alp 1981 heptachlor	
45	Mink (NS)	181 d (F)		1.7 F			Crum et al. 1993 heptachlor	Sperm motility or morphology.
Developmental								
46	Rat (Sprague-Dawley)	daily Gd 8-21, Ld 0-21 (GO)				5 F (increased pup mortality and decreased birth weights)	Lawson and Luderer 2004 heptachlor	
47	Rat (Sprague-Dawley)	Gd 12- pnd 7; pups exposed from pnd 7-21 or pnd 7-42 (GO)			0.03 (impaired spatial memory)		Moser et al. 2001 heptachlor	
48	Rat (Sprague-Dawley)	Gd 10- Ld 7; pups exposed on days 7-21 or 42 (GO)		0.3	3 (decrease in righting reflex)	8.4 (decreased pup survival)	Purkerson-Parker et al. 2001b heptachlor	
49	Rat (Sprague-Dawley)	Gd 12- pnd 71; pups exposed to day 42 (GO)			0.03[d] (suppression of immune response to sheep RBC in offspring)		Smialowicz et al. 2001 heptachlor	

Table 3-1 Levels of Significant Exposure to Heptachlor and Heptachlor Epoxide - Oral (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference Chemical Form	Comments
50	Mink (NS)	181 d (F)		1		1.7 (increased stillbirths and decreased kit survival)	Crum et al. 1993 heptachlor	
CHRONIC EXPOSURE								
Cancer								
51	Mouse (B6C3F1)	80 wk (F)				2.4 M (hepatocellular carcinoma)	NCI 1977 heptachlor	

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

b Differences in levels of health effects and cancer effects between male and females are not indicated in Figure 3-1. Where such differences exist, only the levels of effect for the most sensitive gender are presented.

c Used to derive an acute oral minimal risk level (MRL) of 0.0006 mg/kg/day; dose divided by an uncertainty factor of 1000 (10 for use of a LOAEL, 10 for extrapolation from animals to humans, and 10 for human variability) and a modifying factor of 3 for the use of a serious endpoint.

d Used to derive an intermediate oral minimal risk level (MRL) of 0.0001 mg/kg/day; dose divided by an uncertainty factor of 300 (3 for use of a minimal LOAEL, 10 for extrapolation from animals to humans, and 10 for human variability).

ALT = alanine aminotransferase; Bd Wt = body weight; Cardio = cardiovascular; d = day(s); Endocr = endocrine; (F) = feed; F = Female; (G) = gavage; Gastro = gastrointestinal; Gd = gestational day; (GO) = gavage in oil; Ld = lactation day; LD50 = lethal dose, 50% kill; LOAEL = lowest-observed-adverse-effect level; M = male; Metab = metabolic; NOAEL = no-observed-adverse-effect level; NS = not specified; pnd = post-natal day; RBC = red blood cell(s); Resp = respiratory; x = time(s); (W) = drinking water; wk = week(s)

Figure 3-1 Levels of Significant Exposure to Heptachlor and Heptachlor Epoxide - Oral

Acute (≤14 days)

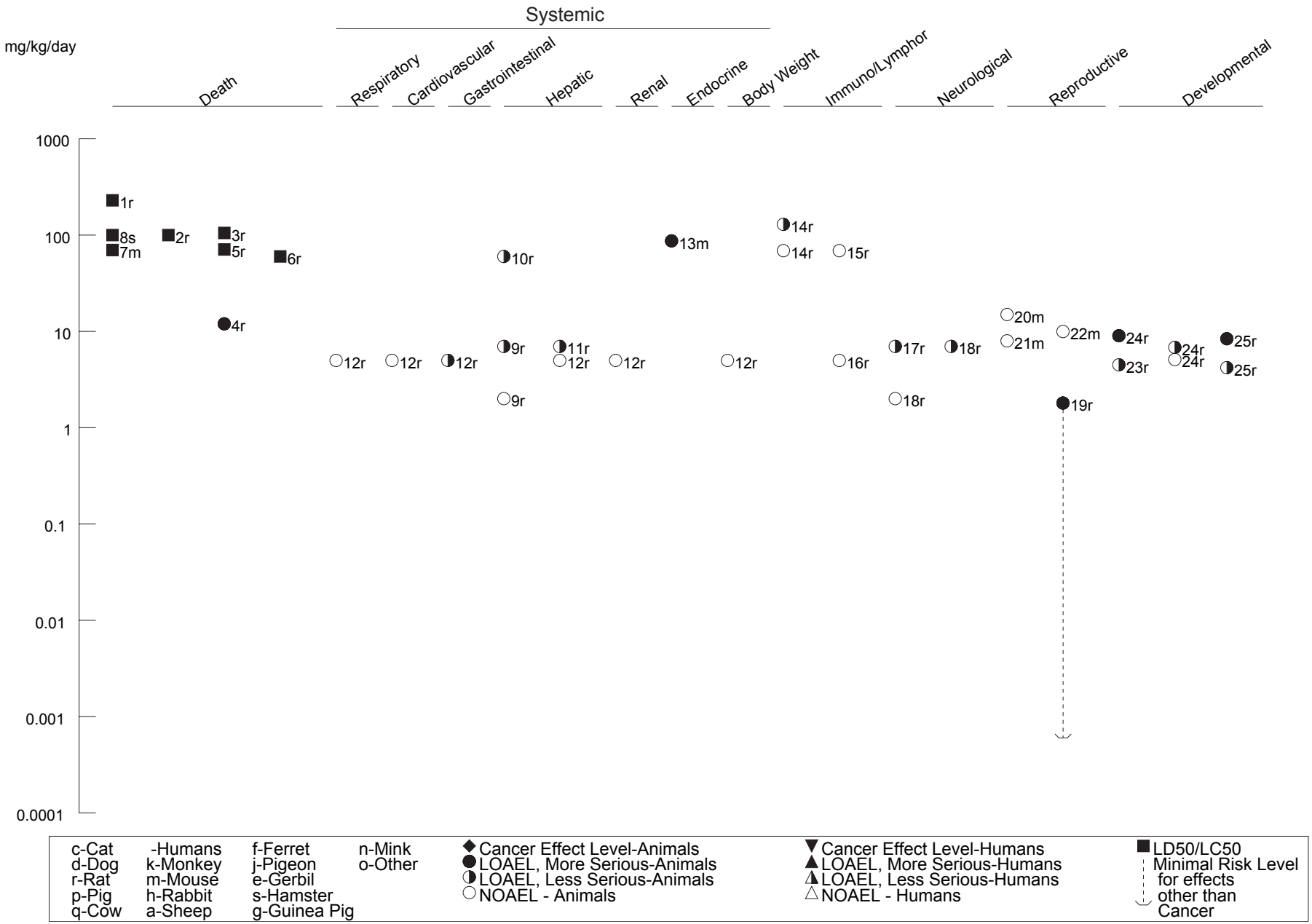


Figure 3-1 Levels of Significant Exposure to Heptachlor and Heptachlor Epoxide - Oral *(Continued)*

Intermediate (15-364 days)

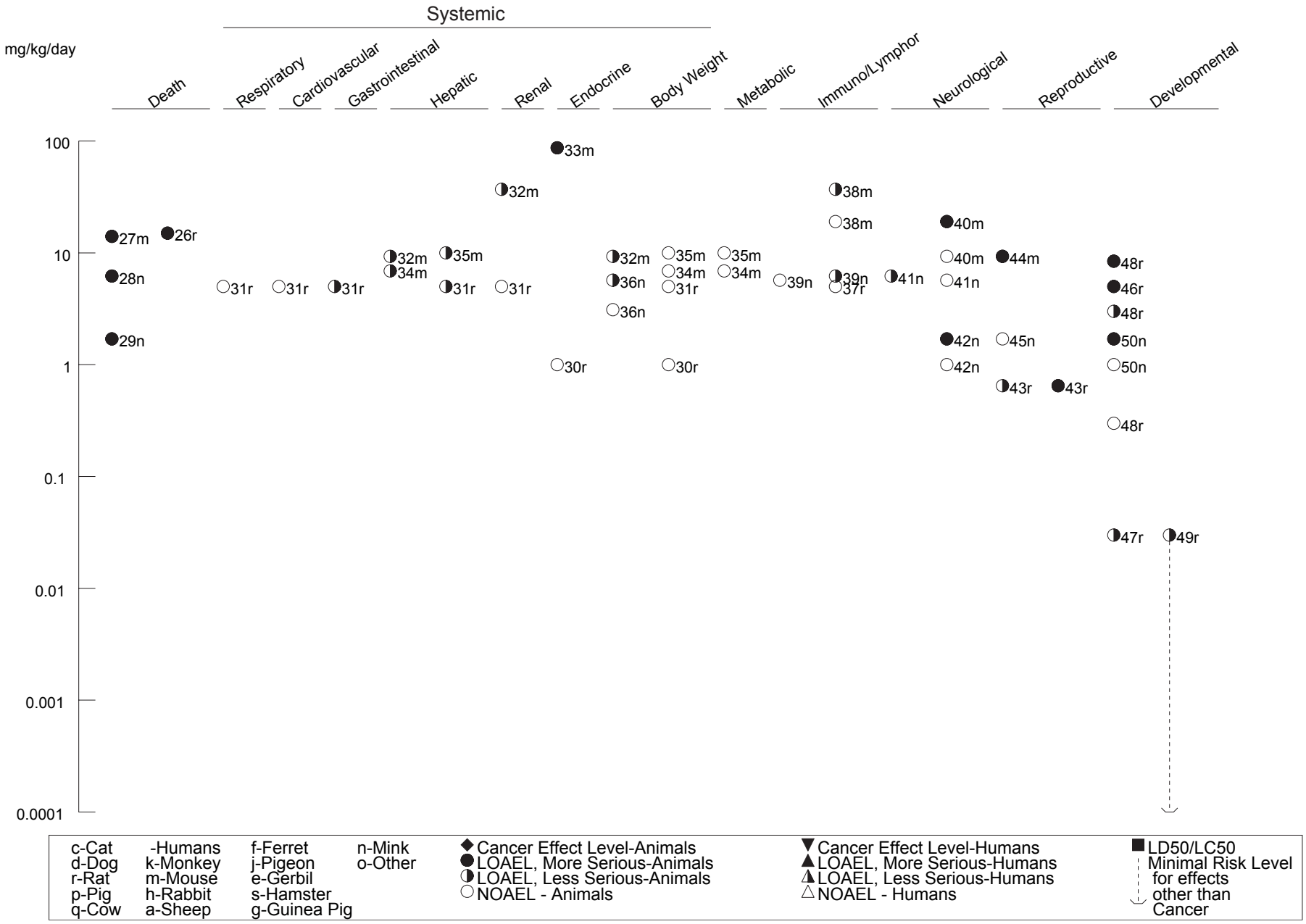
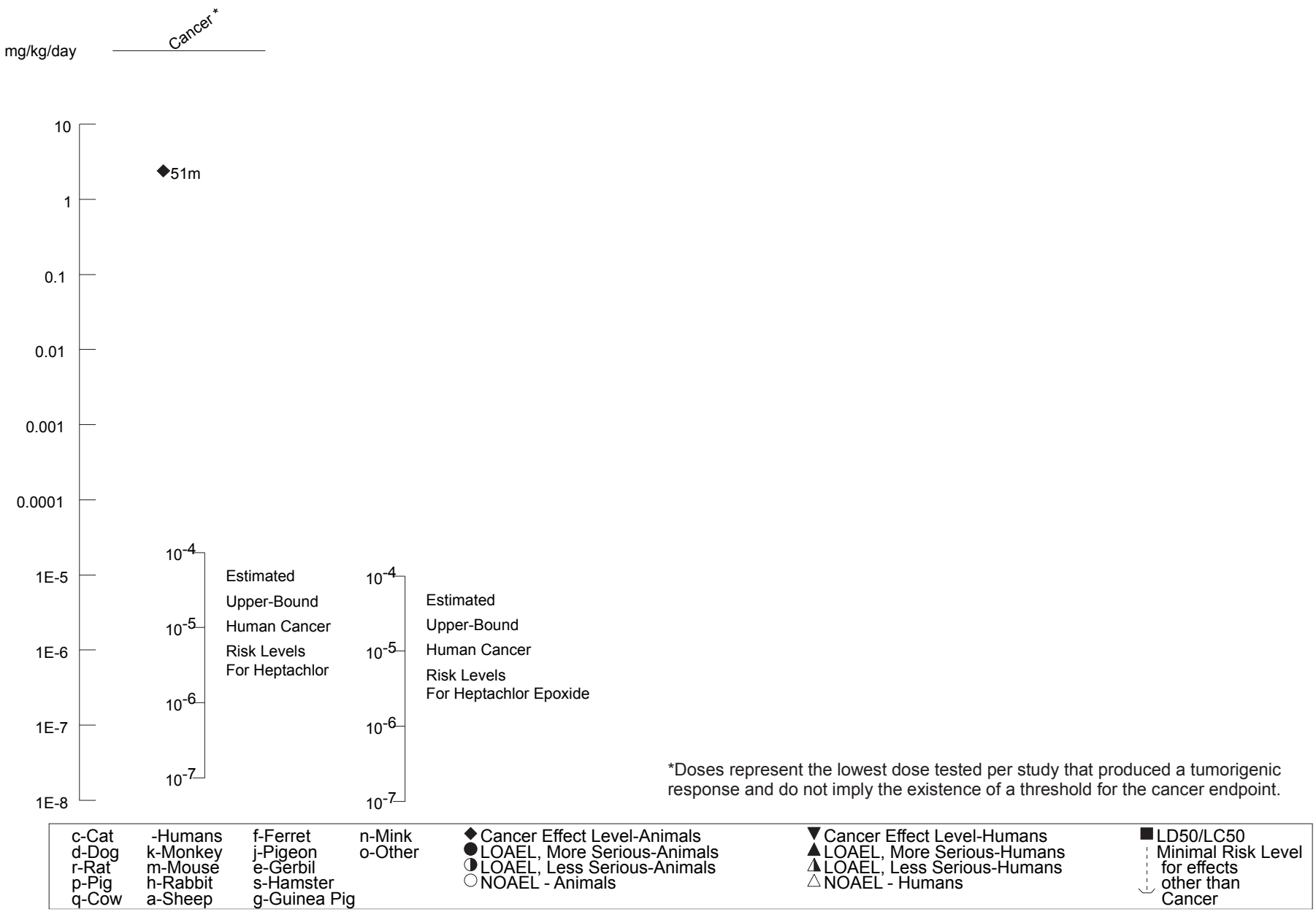


Figure 3-1 Levels of Significant Exposure to Heptachlor and Heptachlor Epoxide - Oral *(Continued)*

Chronic (≥365 days)



Gastrointestinal Effects. No studies were located regarding gastrointestinal effects in humans after oral exposure to heptachlor or heptachlor epoxide. Gross necropsy showed that the stomach and intestinal walls were slightly hyperemic in rats exposed to 5 mg/kg/day of heptachlor in the diet for 14 days (Pelikan 1971); after 28 days of exposure, the mucosa of the stomach and intestine was covered by a thin mucus secretion. No histological alterations were observed in gastrointestinal tissues at either duration. Ulceration and bloody mucus were observed in the stomachs of mink exposed to 1.7 or 3.1 mg/kg/day heptachlor in the diet for 181 days (Crum et al. 1993). These doses were also associated with mortality and pronounced neurological effects; the investigators also noted that the animals stopped eating 1–2 weeks prior to death.

Hematological Effects. No studies were located regarding hematological effects in humans after oral exposure to heptachlor or heptachlor epoxide; one animal study examining hematological end points was identified. A statistically significant increase in total leukocyte levels was observed in rats exposed to heptachlor in the diet for 1, 7, or 28 days (Enan et al. 1982). No alterations in erythrocyte levels were found. As noted previously, it is not known if the reported concentration of 10 mg/kg is a dietary concentration or dose.

Hepatic Effects. Very limited information is available regarding hepatic effects of heptachlor or heptachlor epoxide in humans. Evaluation of individuals exposed for an unspecified period of time to contaminated raw milk products from cattle fed heptachlor-contaminated feed revealed significantly elevated serum levels of heptachlor metabolites relative to national background levels from National Health and Nutrition Examination Survey (NHANES) II and to levels monitored in unexposed reference subjects (Stehr-Green et al. 1986, 1988). Compared to the reference subjects, no significant alterations in serum liver enzyme activity levels were found, and no hepatomegaly was detected by clinical examination. This information is insufficient to draw any meaningful conclusion regarding liver effects of heptachlor in humans.

A number of animal studies have reported liver effects following oral exposure to heptachlor. Although collectively, the studies indicate that the liver is a target of toxicity, interpretation of many of the individual studies is limited by the lack of statistical analysis and the incomplete descriptions of the observed effects (including incidence data). No histological alterations and a small increase (15–17%) in relative liver weight were observed in rats exposed to 5 mg/kg/day in the diet for 14 days (Pelikan 1971). In another acute exposure study, hepatocytomegaly was observed at 7 mg/kg/day in rats administered

heptachlor in corn oil via gavage for 14 days (Berman et al. 1995). Monocellular necrosis and vacuolar dystrophy (statistical significance not reported) were observed in rats administered 7 mg/kg/day heptachlor in corn oil for 3, 7, or 14 days (Krampl 1971); decreases in liver alanine aminotransferase and aldolase activity levels and increases in serum alanine aminotransferase and aldolase activity levels were also observed at this dose level. A single dose of 60 mg/kg heptachlor resulted in similar enzyme activity changes (Krampl 1971); minimal evidence of single monocellular necrosis with inflammatory reaction, vacuolated cells, and pyknotic nuclei were observed 72 hours after dosing. The investigator noted that the histological alterations paralleled the time course for the enzyme changes.

At longer durations, the severity of the liver effects appeared to increase. Steatosis was observed in rats exposed to 5 mg/kg/day in the diet for 28 days (Pelikan 1971) and hepatitis, necrosis, granuloma, and congestion were observed in mice fed diets containing 9.3 mg/kg/day heptachlor for 10 weeks (Akay and Alp 1981). As with acute exposure, intermediate exposure to heptachlor also resulted in alterations in serum enzyme levels; increases in serum aldolase, alanine aminotransferase, and alkaline phosphatase activity levels were observed at 6.9 mg/kg/day and higher (Izushi and Ogata 1990; Krampl 1971). Additionally, increases in serum and hepatic triglyceride levels were observed in mice administered 10 mg/kg heptachlor in olive oil twice weekly (2.9 mg/kg/day) for 92 days (Izushi and Ogata 1990), but not in mice fed 6.9 mg/kg/day in the diet for 180 days (Izushi and Ogata 1990). Liver effects have also been observed in non-rodent experimental animals. Ultrastructural changes indicative of liver cell damage were observed in a small number of pigs administered 2 mg/kg/day “heptachlorine” in the diet for 78 days (Dvorak and Halacka 1975; Halacka et al. 1974); no histological alterations were observed. Fatty liver was reported in mink exposed to 6.2 mg/kg/day heptachlor in the diet for 28 days (Aulerich et al. 1990) or 1.7 mg/kg/day in the diet for 181 days (Crum et al. 1993). In both studies, these doses were associated with increased mortality. One chronic exposure study reported no clear effects on liver function (BSP clearance) in rats exposed to 6 mg/kg/day for 18 months (Mestitzova 1967).

Renal Effects. No studies were located regarding renal effects in humans after oral exposure to heptachlor or heptachlor epoxide. Several studies have examined the potential of heptachlor to induce renal effects in animals. No histological alterations were observed in the kidneys from rats exposed to 5 mg/kg/day heptachlor in the diet for 14 or 28 days (Pelikan 1971) or from rats exposed to 6 mg/kg/day for 18 months (Mestitzova 1967). Increased blood urea levels were observed after 7 or 28 days in rats exposed to 10 mg/kg heptachlor in the diet (Enan et al. 1982); an increase and decrease in relative kidney weight were observed after 7 or 28 days, respectively. As noted previously, interpretation of the results of this study is limited by the poor reporting of the dose. Granulomas were observed in the kidneys of mice

that received 37 mg heptachlor/day for 10 weeks (Akay and Alp 1981); however, no incidence data or statistical analysis was reported, limiting the interpretation of the results. A significant decrease in kidney-to-brain-weight ratio and granulation and discoloration of kidneys were reported in minks fed 6.2 mg/kg/day of heptachlor daily for 28 days (Aulerich et al. 1990); the incidence of kidney damage was not reported.

Endocrine Effects. There is limited information on the potential of heptachlor to induce endocrine effects. Cortical atrophy and slight hypertrophy in the zona glomerulosa of the adrenal gland was observed in mice exposed to 87 mg/kg/day for 11 days (Akay et al. 1982). When the exposure was continued for 26 days, heavy lipid accumulation, congestion, cell degeneration, and extensive fibrosis were observed in the adrenal cortex (Akay et al. 1982). The interpretation of these findings is limited by the poor reporting of the study and the lack of incidence data. One other study examined endocrine end points; no alterations in thyroxine, triiodothyronine, or thyroid stimulating hormone levels were observed in rats administered 1 mg/kg/day via gavage for 21 days (Akhtar et al. 1996).

Ocular Effects. No studies were located regarding ocular effects in humans after oral exposure to heptachlor or heptachlor epoxide. Lens cataracts were observed in 22% of adult rats exposed to 6 mg/kg/day heptachlor in the diet for 4.5–9.5 months; cataracts were not observed in the controls (Mestitzova 1967). As discussed in the developmental toxicity section, cataracts were also observed in the F_1 and F_2 offspring. Other studies have not reported this finding among adults, although no studies were specifically designed to assess this end point. Additionally, Narotsky and Kavlock (1995) did not find increases in cataracts in the offspring of rats administered up to 6 mg/kg/day heptachlor via gavage on gestational days 6–19.

Body Weight Effects. No studies were located regarding body weight effects in humans after oral exposure to heptachlor or heptachlor epoxide. The available animal data do not suggest that oral exposure to heptachlor adversely affects body weight gain in the absence of decreases in food consumption. No alterations in body weight were observed in rats or mice exposed daily to doses as high as 1 and 6.9 mg/kg/day, respectively, for intermediate durations (Akhtar et al. 1996; Izushi and Ogata 1990) or mice administered 10 mg/kg 2 times/week for 92 days (Izushi and Ogata 1990). Decreases in body weight gain were observed in mink exposed to heptachlor in the diet at doses of 5.7 mg/kg/day for 28 days (Aulerich et al. 1990) or 1.7 mg/kg/day for 181 days, which included pregnancy and lactation periods (Crum et al. 1993); significant decreases in food consumption were observed in both studies.

Metabolic Effects. No studies were located regarding metabolic effects in humans after oral exposure to heptachlor or heptachlor epoxide. Studies in animals suggest that exposure to heptachlor may disrupt carbohydrate metabolism. Significant decreases in liver glycogen levels, increases in liver and kidney gluconeogenic enzymes, and increases in blood glucose levels were observed in rats administered a single dose of 200 mg/kg heptachlor (Kacew and Singhal 1973). Decreases in liver glycogen levels and increases in blood glucose were also observed in rats exposed to 10 mg/kg for 1, 7, or 28 days (Enan et al. 1982). The investigators noted that 10 mg/kg was added to the diet; it is not known if this is the dietary concentration (dose would be approximately 0.9 mg/kg/day) or dose. No alterations in blood glucose levels were observed in mice administered 6.9 mg/kg/day heptachlor in the diet for 180 days or 10 mg/kg via gavage 2 times/week (Izushi and Ogata 1990).

3.2.2.3 Immunological and Lymphoreticular Effects

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

No studies were located that specifically investigated the effects on the immune system of oral exposure to heptachlor or heptachlor epoxide in adult animals. However, several studies have found alterations in the lymphoreticular system. Necrotic lymphocytes were observed in the spleen and thymus of rats administered a single dose of 129 mg/kg heptachlor via gavage (Berman et al. 1995); the investigators noted that the effect may have been secondary to generalized toxicity. Fibrosis was observed in the spleens of mice exposed to 37 mg/kg/day heptachlor in the diet for 10 weeks (Akay and Alp 1981). Enlarged and hyperemic spleens were observed in rats exposed to 5 mg/kg/day heptachlor in the diet for 14 or 28 days (Pelikan 1971); however, no apparent alterations in relative spleen weight or histological alterations were observed and the gross changes were not considered to be biologically significant. A decrease in spleen-to-brain-weight ratio was reported in minks receiving 6.2 mg/kg/day heptachlor in the diet for 28 days (Aulerich et al. 1990); this dose was also associated with mortality, weight loss, and decreased food consumption.

The highest LOAEL values for lymphoreticular effects in each species following intermediate exposure are recorded in Table 3-1 and plotted in Figure 3-1.

3.2.2.4 Neurological Effects

No studies were located regarding neurological effects in humans after oral exposure to heptachlor or heptachlor epoxide.

Several studies in animals have reported adverse neurological effects shortly after exposure to heptachlor. At lethal doses, tremors and convulsions were observed in rats (Lehman 1951). Hyperexcitability, incoordination, and seizures were also observed in mink exposed to lethal doses (1.7 or 6.2 mg/kg/day) for intermediate durations (Aulerich et al. 1990; Crum et al. 1993). At nonlethal doses, alterations in a number of functional observational battery tests indicative of excitability were observed in rats following a single dose or 14 doses of 7 mg/kg (Moser et al. 1995); the excitability changes included increased arousal and reactivity to removal from the home cage and handling. The persistence of the effect was directly related to dose level. Decreases in motor activity and hunched posture in the home cage was observed 4 hours after a single dose of 129 mg/kg (Moser et al. 1995). Another study conducted by this group (Moser et al. 2003) found decreases in motor activity, increases in forelimb and hindlimb grip strength, handling reactivity and arousal, gait abnormalities, tremors, and piloerection in rats receiving gavage doses of 1–14 mg/kg/day for 10 days; a LOAEL cannot be identified from this study because the investigators did not provide data for individual end points or dose levels. Similar to the effects noted in the Moser studies, Akay and Alp (1981) reported difficulty in standing, walking, and righting in mice exposed to 19 mg/kg/day heptachlor in the diet for 10 weeks. In addition to these effects observed in mature animals, adverse neurological effects have been observed in the offspring of rats exposed to heptachlor during gestation, lactation, and postnatally; these studies are discussed in Section 3.2.2.6.

Statistically significant changes in electroencephalogram (EEG) patterns were reported in mature female rats administered heptachlor in the diet at levels of 1 and 5 mg/kg/day for three generations (Formanek et al. 1976). Interpretation of these findings is difficult because details of the dosing, the procedures used, and conditions of the rats were not described.

The highest NOAEL values and all reliable LOAEL values for neurological effects in each species following intermediate exposure are recorded in Table 3-1 and plotted in Figure 3-1.

3.2.2.5 Reproductive Effects

Significantly higher levels of heptachlor epoxide were detected in the sera of a group of women identified through hospital records with premature delivery than in the sera of a control group with normal delivery

(Wassermann et al. 1982). However, sera levels of 8 of the 10 organochlorine pesticides for which analytical data were obtained were all significantly higher in the premature delivery group. In addition, route, duration, and level of exposure information was not reported. Heptachlor epoxide has been detected in stillborn infant brain, adrenal, lung, heart, liver, kidneys, spleen, and adipose tissue, indicating transplacental transfer of heptachlor or heptachlor epoxide (Curley et al. 1969). These studies also reported the presence of polychlorinated biphenyls (PCBs), lindane, and dieldrin in the samples. It is difficult to assess the causal relationship between adverse reproductive outcome in humans and exposure to heptachlor and heptachlor epoxide due to lack of control for confounding factors such as smoking and concomitant exposure to other pesticides and lack of completeness of report data. No adverse effects on reproduction (no decrease in fertility, no increase in fetal or neonatal deaths) were reported by Le Marchand et al. (1986) among women of child-bearing age following ingestion of heptachlor-containing milk in excess of 0.1 ppm for 27–29 months.

A number of animal studies have demonstrated that exposure to heptachlor can result in decreased fertility and increased pregnancy losses. Impaired fertility was reported in female rats administered via gavage 0.65 mg/kg/day heptachlor in groundnut oil for 14 days prior to mating (Amita Rani and Krishnakumari 1995) and male and female rats fed 0.25 mg/kg/day heptachlor for 60 days (Green 1970); 100% infertility was observed in mice fed 9.3 mg/kg/day heptachlor for 10 weeks (Akay and Alp 1981). No effect on fertility was observed in male mice administered via gavage 10 mg/kg/day heptachlor or 8 mg/kg/day heptachlor epoxide for 5 days (Epstein et al. 1972) or a single dose of 15 mg/kg heptachlor:heptachlor epoxide (25%:75%) (Arnold et al. 1977).

Significant increases in resorptions were observed in male and female rats receiving gavage doses of 0.65 or 1.8 mg/kg/day, respectively, heptachlor for 70 or 14 days, respectively, prior to mating to control animals (Amita Rani and Krishnakumari 1995) and male and female rats fed 0.25 mg/kg/day heptachlor for 60 days prior to mating and throughout gestation (Green 1970). An increase in the incidence of stillbirths was observed in mink fed a diet containing 1.7 mg/kg/day heptachlor for 42 days prior to mating and throughout gestation (Crum et al. 1993). Similarly, a decrease in litter size was observed in rats exposed to 6 mg/kg/day heptachlor in the diet for an unspecified portion of an 18-month study (Mestitzova 1967). No alterations in preimplantation losses or early fetal deaths were observed in control females mated to males administered via gavage 10 mg/kg/day heptachlor or 8 mg/kg/day heptachlor epoxide for 5 days (Epstein et al. 1972) or a single dose of 15 mg/kg/day heptachlor:heptachlor epoxide mixture (Arnold et al. 1977).

Other reproductive alterations include a decrease in epididymal sperm count in rats administered gavage doses of 0.65 mg/kg/day heptachlor for 70 days (Amita Rani and Krishnakumari 1995) and decreases in estradiol-17β and progesterone levels in rats gavaged with 1.8 mg/kg/day for 14 days (Amita Rani and Krishnakumari 1995). Vaginal bleeding was reported in some rats exposed to 2.0 or 4.0 mg/kg/day technical-grade heptachlor for 80 weeks (NCI 1977); however, the incidence and statistical significance were not reported.

All reliable LOAEL values for reproductive effects in each species and duration category are recorded in Table 3-1 and plotted in Figure 3-1.

3.2.2.6 Developmental Effects

Several studies have examined potential developmental effects in the children of women exposed to elevated levels of heptachlor or heptachlor epoxide. Between 1980 and 1982, the commercial supply of cow's milk for the Hawaiian island of Oahu was contaminated with heptachlor epoxide; the source of exposure was treated pineapple plants that were used as cattle feed. Cow milk fat levels of heptachlor measured in Hawaii during this time ranged from 0.12 to 5.00 ppm (EPA's action level is 0.1 ppm); in a prior analysis (1978–1980), the levels were comparable to the rest of the United States. Using hospital records, Le Marchand et al. (1986) examined a possible association between heptachlor epoxide exposure and birth defects. No increase in fetal or neonatal deaths or incidence of low birth weight infants were found in this study cohort. Of the 23 categories of major congenital malformations evaluated, 22 were found to be decreased in the study population when compared with cohorts from the other Hawaiian islands and from the U.S. general population for the same time period. One type of malformation (anomalies of the abdominal wall) was found to be slightly increased in the study cohort during the period of known exposure compared with the control cohorts. However, the baseline data for this type of malformation were not available prior to study initiation, and birth defects may be underreported. It was, therefore, not possible to document the temporal change in the incidence of this type of malformation. Since women who might not have consumed the contaminated milk were included in the study group, positive findings may have been diluted as a result of misclassification bias. A subsequent study of high school students born on Oahu and likely prenatally exposed to heptachlor epoxide was conducted by Baker et al. (2004b; available as an abstract). As compared to high school students living on Oahu since first grade (but not born on Oahu), an association between gestational exposure to heptachlor epoxide and lower neurobehavioral performance was found. In particular, impaired performance was found on tests of abstract concept formation, visual perception, and motor planning; this group also had more reported

behavioral problems. No significant associations between school-based performance measures, such as grade point average, and gestational heptachlor epoxide exposure were found.

Two studies examined possible associations between maternal heptachlor epoxide levels and early childhood development (Hertz-Picciotto et al. 2004) or birth weight (Gladen et al. 2003). An association between maternal serum heptachlor epoxide levels and their children's performance on a test of nonverbal perceptual reasoning was found in San Francisco residents. However, inclusion of PCBs in the model severely reduced the magnitude of the association (Hertz-Picciotto et al. 2004; only available as an abstract). The investigators concluded that the cognitive deficits were probably due to co-exposure to other compounds. Gladen et al. (2003) found no significant association between breast milk levels of heptachlor epoxide (taken 4–5 days after birth) and birth weight among residents of two cities in Ukraine.

Several animal studies have examined the potential developmental toxicity of heptachlor. *In utero* exposure to heptachlor doses of 5.0 mg/kg and higher has resulted in pup mortality (Lawson and Luderer 2004; Narotsky et al. 1995; Purkerson-Parker et al. 2001b). These doses are also associated with significant maternal toxicity such as mortality, convulsions, and/or weight loss; however, at 5.0 mg/kg/day, increased pup mortality was also observed in dams without overt signs of toxicity (Lawson and Luderer 2004).

Gestational exposure to lower doses results in decreases in pup body weight. The threshold for this effect appears to be around 4–5 mg/kg/day (Lawson and Luderer 2004; Narotsky and Kavlock 1995; Narotsky et al. 1995). No alterations in pup body weight were observed at 3 mg/kg/day (Moser et al. 2001; Purkerson-Parker et al. 2001b; Smialowicz et al. 2001). Exposure to heptachlor does not appear to result increased frequency of anomalies or abnormalities (Narotsky et al. 1995; Smialowicz et al. 2001). Additionally, heptachlor does not appear to impair the development of the reproductive system. No delays in vaginal opening or prepuce separation (indices of female and male puberty respectively) were observed in the offspring of rats administered via gavage to 3 mg/kg/day heptachlor on gestational day 12 through postnatal day 21 (Smialowicz et al. 2001) or 5 mg/kg/day on gestational days 8–21 and lactational days 1–21 (Lawson and Luderer 2004). Additionally, continued exposure of the offspring until postnatal day 42 and subsequent mating with untreated animals did not result in adverse reproductive or developmental outcomes (Smialowicz et al. 2001).

Developmental studies have found neurological and immunological effects in offspring. Gestational, lactational, and offspring exposure until postnatal day 21 or 42 to 3 mg/kg/day heptachlor resulted in

significant delays in righting reflex, likely due to a delay in the ontogeny of righting rather than an inability to perform the task (Moser et al. 2001; Purkerson-Parker et al. 2001b). No alterations in motor activity ontogeny were observed (Moser et al. 2001; Purkerson-Parker et al. 2001b). Some alterations in functional observation battery tests were observed in rats exposed to heptachlor during gestation, lactation, and postnatally until day 21 or 42 (Moser et al. 2001). Many of these alterations were only significant at the lowest dose tested (0.03 mg/kg/day). In water maze tests, exposure to 0.03 mg/kg/day heptachlor and higher resulted in slowed acquisition of a spatial task and impaired recall (Moser et al. 2001). This was observed in rats exposed *in utero*, during lactation, and until postnatal day 42, but not in rats exposed until postnatal day 21.

Smialowicz et al. (2001) found a significant suppression of the immune response to sheep red blood cells in rats exposed to 0.03 mg/kg/day *in utero*, during lactation, and postnatally until day 42 of age. No significant alterations in the response to T-cell mitogens or in delayed-type and contact hypersensitivity were observed. A decrease in $OX12^{+}OX19^{-}$ splenic populations was observed at 3.0 mg/kg/day.

The reliable LOAEL values for developmental effects in rats following intermediate and chronic exposure are recorded in Table 3-1 and plotted in Figure 3-1.

3.2.2.7 Cancer

There is limited information on the carcinogenicity of heptachlor or heptachlor epoxide in humans following oral exposure; several studies have examined the possible association between heptachlor epoxide tissue levels and cancer risk. Interpretation of the studies is limited by the lack of information on heptachlor exposure (including the route of exposure), variables that may affect organochlorine levels (including diet and body mass index), and possible concomitant exposure to other chemicals. No significant associations were found for endometrial cancer in women in the United States (Sturgeon et al. 1998) or breast cancer in Norwegian women (Ward et al. 2000). Another study found a significant association between heptachlor epoxide levels in breast tissue and the prevalence of breast cancer (Cassidy et al. 2005). Two case control studies examined the possible association between heptachlor epoxide levels and non-Hodgkin's lymphoma. One study found a significant association among individuals with the highest heptachlor epoxide adipose levels (odds ratio of 3.41, 95% confidence interval of 1.89–6.16) (Quintana et al. 2004), whereas the other study did not found a significant association between serum heptachlor epoxide levels and non-Hodgkin's lymphoma (Cantor et al. 2003).

Animal studies provide some evidence for the carcinogenicity of heptachlor. Significant increases in the incidence of hepatocellular carcinoma were observed in male and female mice exposed to time-weighted average (TWA) doses of 2.4 or 3.0 mg/kg/day, respectively, technical-grade heptachlor (72% heptachlor, 20% chlordane) in the diet for 80 weeks. Although increases in liver tumors were also observed at a lower dose (0.8 mg/kg/day for males and 1.2 mg/kg/day for females), the incidence was not significantly different than controls (NCI 1977). No significant increases in neoplastic tumor incidence were observed in male or female rats similarly exposed to technical-grade heptachlor TWA doses of 3.9 or 2.6 mg/kg/day, respectively (NCI 1977). Although a statistically significant increase in the incidence of follicular cell neoplasms (adenomas and carcinomas) was observed in the thyroid of female rats fed a TWA dose of 2.0 mg/kg/day technical-grade heptachlor for 80 weeks, the study investigators did not judge the alterations to be sufficient to clearly indicate a carcinogenic effect in the thyroid gland (NCI 1977). No other significant alterations were observed in the rats (NCI 1977). The Cancer Effect Level (CEL) in mice from chronic exposure to heptachlor is recorded in Table 3-1 and plotted in Figure 3-1.

The positive carcinogenicity findings of the mouse NCI (1977) study is supported by evidence that heptachlor is a tumor promoter. Dietary administration of ≥0.65 mg/kg/day heptachlor (97.6% purity) for 25 weeks promoted the development of hepatocellular foci and hepatocellular neoplasms in male mice previously initiated with 3.8 mg/kg/day diethylnitrosamine in drinking water for 14 weeks (Williams and Numoto 1984).

EPA has classified heptachlor and heptachlor epoxide in Group B2 (probable human carcinogen) (IRIS 2006). EPA has derived an oral slope factor of 4.5 per (mg/kg)/day for heptachlor and 9.1 per (mg/kg)/day for heptachlor epoxide. The doses corresponding to cancer risk levels ranging from 10^{-4} to 10^{-7} are $2.0x10^{-5}$–$2.0x10^{-8}$ mg/kg/day for heptachlor and $1.0x10^{-5}$–$1.0x10^{-8}$ mg/kg/day for heptachlor epoxide as indicated in Figure 3-1. The oral cancer potency factor is a plausible upper-bound estimate of the lifetime probability of an individual developing cancer as a result of oral exposure per unit intake of the chemical. The International Agency for Research on Cancer (IARC) has classified heptachlor and heptachlor epoxide as Group 2b chemicals (possibly carcinogenic to humans) (IARC 2001).

3.2.3 Dermal Exposure

There is very little information on dermal exposures in either humans or animals. Most occupational exposures to heptachlor and heptachlor epoxide are assumed to be some combination of inhalation and

dermal exposure, but there are no data to quantify the relative contribution of each route. The occupational studies on pesticide workers are discussed in Section 3.2.1.

3.2.3.1 Death

No studies were located regarding death in humans after dermal exposure to heptachlor or heptachlor epoxide.

For heptachlor dissolved in xylene and administered once, Gaines (1969) reported dermal LD_{50} values in Sherman rats of 195 mg/kg (males) and 250 mg/kg (females).

3.2.3.2 Systemic Effects

No studies were located regarding systemic effects in humans or animals after dermal exposure to heptachlor or heptachlor epoxide.

No studies were located regarding the following health effects in humans or animals after dermal exposure to heptachlor or heptachlor epoxide:

3.2.3.3 Immunological and Lymphoreticular Effects
3.2.3.4 Neurological Effects
3.2.3.5 Reproductive Effects
3.2.3.6 Developmental Effects
3.2.3.7 Cancer

3.3 GENOTOXICITY

There are limited mammalian *in vivo* data on the genotoxicity of heptachlor or heptachlor epoxide. Heptachlor, heptachlor epoxide, and a mixture of heptachlor and heptachlor epoxide (25:75) were found to be negative in *in vivo* dominant lethal studies in the germ-line cells of male Charles River or Swiss mice (Arnold et al. 1977; Epstein et al. 1972).

Several *in vitro* studies have examined the genotoxicity of heptachlor or heptachlor epoxide (Table 3-2). The available weight of evidence suggests that neither compound alters the frequency of gene mutations in prokaryotic organisms (Glatt et al. 1983; Marshall et al. 1976; NTP 1987; Probst et al. 1981; Zeiger et

Table 3-2. Genotoxicity of Heptachlor and Heptachlor Epoxide In Vitro

Species (test system)	End point	Results		Chemical form	Reference
		With activation	Without activation		
Prokaryotic organisms:					
Salmonella typhimurium (histidine reversion)	Gene mutation	−	−	Heptachlor	Zeiger et al. 1987
S. typhimurium (Ames assay)	Gene mutation	−	−	Heptachlor	Marshall et al. 1976; NTP 1987
S. typhimurium (Ames assay)	Gene mutation	−	−	Heptachlor epoxide	Marshall et al. 1976; NTP 1987
S. typhimurium (Ames assay)	Gene mutation	+	−	Heptachlor	Gentile et al. 1982
S. typhimurium (modified Ames assay)	Gene mutation	−	−	Heptachlor	Probst et al. 1981
S. typhimurium (modified Ames assay)	Gene mutation	−	−	Heptachlor epoxide	Glatt et al. 1983
Escherichia coli (modified Ames assay)	Gene mutation	−	−	Heptachlor	Probst et al. 1981
S. typhimurium (disc assay)	DNA damage	No data	−	Heptachlor	Rashid and Mumma 1986
E. coli (DNA repair assay)	DNA damage	No data	−	Heptachlor	Rashid and Mumma 1986
Eukaryotic organisms:					
Fungi:					
Saccharomyces cerevisiae (*ade*, *trp* loci assay)	Gene conversion	−	−	Heptachlor	Gentile et al. 1982
Aspergillus nidulans (strain 35/liquid medium)	Gene mutation	No data	−	Heptachlor epoxide	Crebelli et al. 1986
A. nidulans (strain P1/liquid medium)	Chromosome malsegregation	No data	−	Heptachlor epoxide	Crebelli et al. 1986
Mammaliam cells:					
Mouse (L5178Y tk^+/tk^- lymphoma cell forward mutation assay)	Gene mutation	No data	+	Heptachlor	McGregor et al. 1988
Rat (ARL-HGPRT assay)	Gene mutation	−	NA	Heptachlor	Telang et al. 1982
Chinese hamster (ovary cells)	Chromosomal aberrations	+	−	Heptachlor	NTP 1987
Chinese hamster (ovary cells)	Sister chromatid exchange	+	+	Heptachlor	NTP 1987

Table 3-2. Genotoxicity of Heptachlor and Heptachlor Epoxide In Vitro

Species (test system)	End point	Results		Chemical form	Reference
		With activation	Without activation		
Rat (primary hepatocytes)	Unscheduled DNA synthesis	−	NA	Heptachlor	Probst et al. 1981; Maslansky and Williams 1981
Mouse (primary hepatocyte)	Unscheduled DNA synthesis	−	NA	Heptachlor	Maslansky and Williams 1981
Syrian hamster (primary hepatocytes)	Unscheduled DNA synthesis	−	NA	Heptachlor	Maslansky and Williams 1981
Human (SV-40 transformed fibroblasts)	Unscheduled DNA synthesis	+	−	Heptachlor	Ahmed et al. 1977
Human (SV-40 transformed fibroblasts)	Unscheduled DNA synthesis	+	−	Heptachlor epoxide	Ahmed et al. 1977

− = negative result; + = positive result; *ade* = adenine; ARL = adult rat liver epithelial cell line; DNA = deoxyribonucleic acid; HGPRT = hypoxanthine-guanine phosphoribosyl transferase; NA = not applicable; tk = thymidine kinase locus; *trp* = tryptophan

al. 1987). Heptachlor without metabolic activation caused gene mutations in mouse lymphoma cells but not in adult rat liver cells (McGregor et al. 1988; Telang et al. 1982). No alterations in gene conversion were observed in *Saccharomyces cerevisiae* following heptachlor exposure with and without activation (Gentile et al. 1982); heptachlor epoxide was also negative for gene mutation in *Aspergillus nidulans* (Crebelli et al. 1986).

Heptachlor did not cause DNA damage in *Salmonella typhimurium* or *Escherichia coli* in the absence of metabolic activators (Rashid and Mumma 1986). Heptachlor was negative for unscheduled DNA synthesis (UDS) in rat, mouse, and hamster (Maslansky and Williams 1981; Probst et al. 1981). In contrast, an increase in UDS was observed in human SV-40 transformed fibroblasts after exposure to heptachlor and heptachlor epoxide in the presence of metabolic activators (Ahmed et al. 1977).

Heptachlor epoxide did not alter the occurrence of chromosome malsegregration in *A. nidulans* (Crebelli et al. 1986). Chromosomal alterations were observed in mammalian cells. Chromosomal aberrations were observed in Chinese hamster ovary cells following exposure to heptachlor with metabolic activation and sister chromatid exchange was observed both with and without metabolic activation (NTP 1987). Refer to Table 3-2 for a summary of the results of these *in vitro* studies.

Several studies were located involving heptachlor genotoxicity in plants. A positive response was noted for the waxy gene mutation in maize (*Zea mays*) following exposure to heptachlor *in situ* (Gentile et al. 1982). A micronucleus test in *Tradescantia* produced a significant positive dose-related response at 1.88 ppm heptachlor, suggesting that heptachlor has clastogenic potential in plants (Sandhu et al. 1989). Early separation during metaphase, condensation, stickiness, and chromatin bridges were observed after heptachlor treatment on mitotic chromosomes in *Lens culinaris*, *Lens esculenta*, *Pisum sativum*, and *Pisum arvense* (Jain and Sarbhoy 1987a). Chromosomal abnormalities such as stickiness, non-orientation during metaphase I, fragments, multivalents, and bridges were also observed in meiotic chromosomes after heptachlor treatment (Jain and Sarbhoy 1987b). These studies by Jain and Sarbhoy report no statistical comparisons with which to interpret the results; therefore, it is difficult to evaluate the significance of their research. Even though these plant studies suggest that heptachlor is potentially genotoxic, the applicability to mammalian genotoxicity remains questionable.

3.4 TOXICOKINETICS

3.4.1 Absorption

3.4.1.1 Inhalation Exposure

No studies were located regarding absorption in humans after inhalation exposure to heptachlor or heptachlor epoxide. One animal study provides suggestive evidence of absorption following inhalation exposure to heptachlor epoxide (Arthur et al. 1975). Elevated levels of heptachlor epoxide were found in the fat (0.039 versus 0.016 ppm in controls) of rabbits housed outdoors in an area of pesticide use; the air concentrations of DDT, dieldrin, and heptachlor epoxide were 649.6, 4.59, and 1.86 ng/m^3, respectively.

3.4.1.2 Oral Exposure

No information on the extent of oral absorption of heptachlor or heptachlor epoxide in humans was identified. Qualitative evidence of absorption was found in a study of families consuming dairy products contaminated with heptachlor epoxide (Stehr-Green et al. 1988). Higher serum heptachlor epoxide levels were detected in the family members compared to an unexposed population (0.84 versus 0.50 ppb).

Heptachlor is absorbed from the gastrointestinal tract of rats (Radomski and Davidow 1953; Tashiro and Matsumura 1978) and cattle (Harradine and McDougall 1986) as indicated by the presence of heptachlor and/or its metabolites in serum, fat, liver, kidneys, and muscle (Radomski and Davidow 1953). Based on available toxicity data (Podowski et al. 1979), it is assumed that heptachlor epoxide is also absorbed via the gastrointestinal tract. One study provides suggestive evidence that at least 50% of an orally administered dose of heptachlor is absorbed by rats (Tashiro and Matsumura 1978). Ten days after administration of a single dose, 6% of the radioactivity from radiolabeled (^{14}C) heptachlor was found in the urine and 60% was detected in the feces (primarily present as heptachlor metabolites).

3.4.1.3 Dermal Exposure

No studies were located regarding absorption in humans after dermal exposure to heptachlor or heptachlor epoxide.

Heptachlor is absorbed through the skin following topical application as indicated by its dermal toxicity in rats (Gaines 1969), but quantitative data are not available. However, the data should be interpreted

cautiously because heptachlor epoxide ingestion was not prevented by restraining the animals or removing excess heptachlor from the skin.

3.4.2 Distribution

3.4.2.1 Inhalation Exposure

No studies were located regarding distribution of heptachlor or heptachlor epoxide in humans or animals following inhalation exposure.

3.4.2.2 Oral Exposure

No human studies were located regarding the distribution of heptachlor and its metabolites after oral exposure. A number of monitoring studies found elevated levels of heptachlor epoxide in fat, serum, liver, and brain. It is not known if the elevated levels of heptachlor epoxide were the result of heptachlor or heptachlor epoxide exposure or chlordane exposure (heptachlor epoxide is a minor metabolite of chlordane) (Adeshina and Todd 1990; Barquet et al. 1981; Burns 1974; Greer et al. 1980; Klemmer et al. 1977; Polishuk et al. 1977b; Radomski et al. 1968; Stehr-Green et al. 1988; Wassermann et al. 1974).

Heptachlor epoxide was measured in a strip of skin, fat, and subcutaneous tissue from 68 children who died in the perinatal period and ranged from not detected (nondetectable) to 0.563 ppm (mean, 0.173 ppm) (Zavon et al. 1969). In 10 other stillborn infants, heptachlor epoxide levels measured in various tissues were as follows: brain (nondetectable), lung (0.17±0.07 ppm), adipose (0.32±0.10 ppm), spleen (0.35±0.08 ppm), liver (0.68±0.50 ppm), kidneys (0.70±0.28 ppm), adrenals (0.73±0.27 ppm), and heart (0.80±0.30 ppm) (Curley et al. 1969). Selby et al. (1969) reported a placenta/maternal blood concentration ratio for heptachlor epoxide of 5.8. In another study, the following heptachlor epoxide levels were measured in extracted lipids from mothers and newborn infants: maternal adipose tissue (0.28±0.31 ppm), maternal blood (0.28±0.46 ppm), uterine muscle (0.49±0.51 ppm), newborn blood (1.00±0.95 ppm), placenta (0.50±0.40 ppm), and amniotic fluid (0.67±1.16 ppm) (Polishuk et al. 1977a). These data provide evidence of transplacental transfer to the fetus.

Animal studies regarding heptachlor and heptachlor epoxide distribution in body tissues are limited. Analysis of body fat from 20 adult female rats fed heptachlor in their diet at a level of 35 ppm for 3 months revealed a high concentration of heptachlor epoxide but not heptachlor (Radomski and Davidow 1953). Further analysis showed that accumulation of heptachlor epoxide was directly related to the dose

of heptachlor given. Examination of other tissues in addition to adipose tissue showed that fat had the highest concentrations of heptachlor epoxide; markedly lower amounts were found in liver, kidneys, and muscle, and none was found in the brain. In a parallel study, three dogs were also examined. Doses of 1 mg/kg/day for 12–18 months produced the same distribution pattern as in rats, but the livers of dogs contained more heptachlor epoxide than the kidneys and muscle tissue. Levels in all tissues were higher in female dogs than in males.

The rate of heptachlor epoxide accumulation in, and elimination from, fat was determined in rats fed diets containing 30 ppm heptachlor for 12 weeks, then fed untreated diets for 12 more weeks (Radomski and Davidow 1953). Interim sacrifices at various times during treatment showed that the residue in the fat of males reached a plateau at approximately 2–8 weeks. Thereafter, the levels decreased and were below the detection limit by the end of week 6 post-dosing. In females, the heptachlor epoxide level in fat was much higher than males by the second week and throughout the remainder of the study. By the end of the 8th week post-dosing, the heptachlor epoxide level was below the detection limit in females. No estimates of elimination half-lives from fat were provided.

Heptachlor and heptachlor epoxide residues were found in the fat (≥0.16 and ≥18.25 ppm, respectively), liver (≥0.08 and ≥2.11 ppm, respectively), and muscle (0 and ≥0.03 ppm, respectively) of pigs fed 2 mg/kg/day heptachlorine (purity unspecified) for 78 days (Halacka et al. 1974). When pigs were fed 5 mg/kg/day, the levels of heptachlor and heptachlor epoxide were higher: 0.37 and 25.82 ppm, respectively, in the fat; 0.23 and 4.94 ppm, respectively, in liver; and 0 and 0.7 ppm, respectively, in muscle.

3.4.2.3 Dermal Exposure

No studies were located regarding distribution in humans or animals after dermal exposure to heptachlor or heptachlor epoxide.

3.4.3 Metabolism

No studies were located regarding metabolism of heptachlor or heptachlor epoxide in humans exposed to these pesticides. However, information is available regarding *in vivo* metabolism in rats and *in vitro* metabolism by human and rat liver microsomes.

Ten days after male rats were administered a single oral dose of ^{14}C-heptachlor by gavage in corn oil, about 72% of the radioactivity was eliminated in the feces in the form of metabolites and 26% as parent compound. The major fecal metabolites were heptachlor epoxide (13.1% of total ^{14}C compounds), 1-exo-hydroxychlordene (19.5%), 1-exo-hydroxy-2,3-exo-epoxychlordene (17.5%), and 1,2-dihydroxydihydro-chlordene (3.5%), as well as two unidentified products (Tashiro and Matsumura 1978). The proposed metabolic scheme for heptachlor is presented in Figure 3-2.

Tashiro and Matsumura (1978) also conducted experiments to compare *in vitro* metabolism of ^{14}C-heptachlor in microsomal preparations from human livers and rat livers. The primary metabolites produced by in both preparations were heptachlor epoxide, 1-exo-hydroxychlordene, 1-exo-hydroxy-2,3-exo-epoxychlordene, and 1,2-dihydroxydihydrochlordane. However, the levels of heptachlor epoxide were 4 times higher in the rat microsomal preparations than in the humans; 85.8% of the radiolabel was in the form of heptachlor epoxide in rat microsomes compared to 20.4% for human microsomes. These *in vivo* and *in vitro* data suggest that the ratio of heptachlor to heptachlor epoxide stored in adipose tissue would be higher in humans than rats (Tashiro and Matsumura 1978).

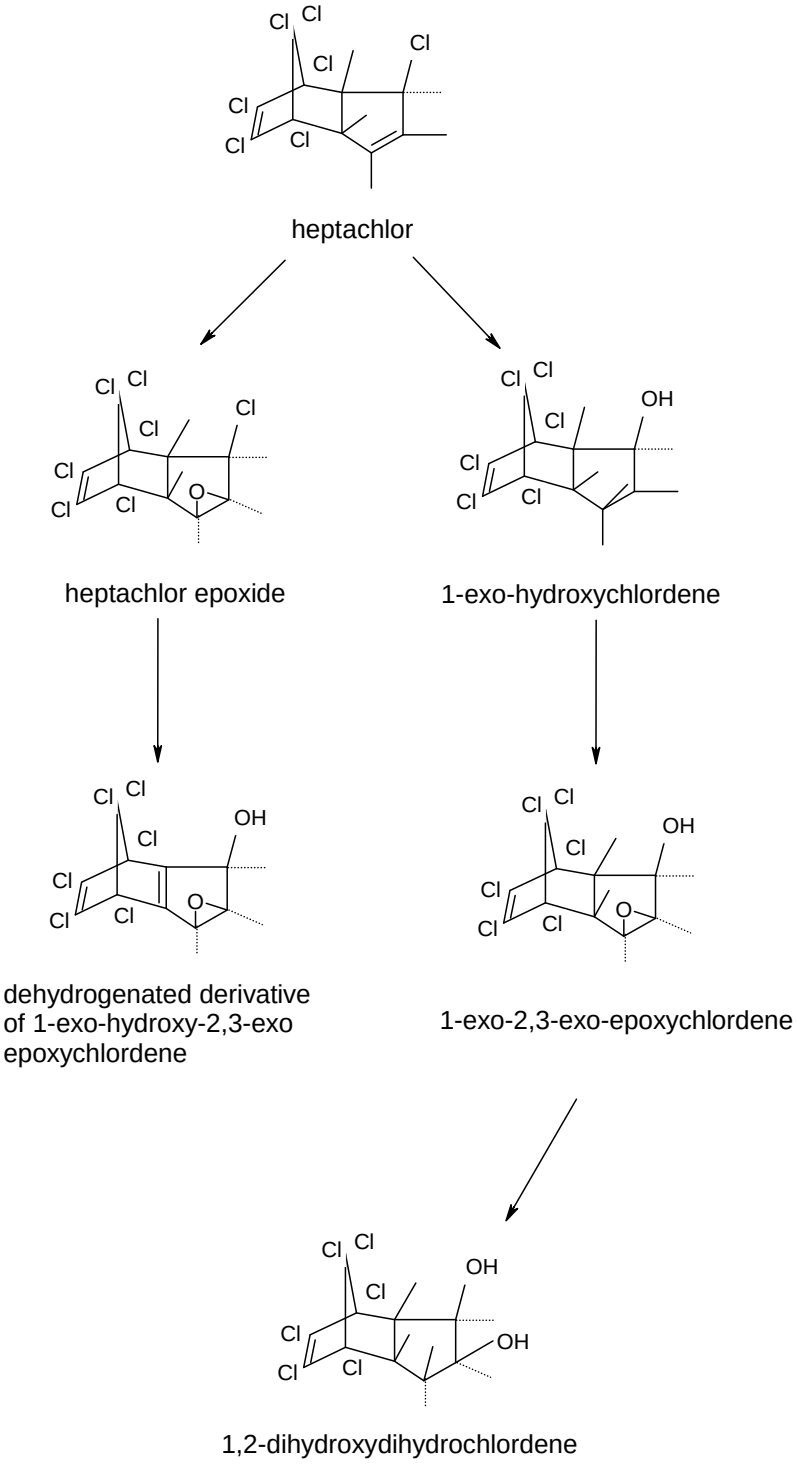
Heptachlor also is a metabolic product of chlordane. Both heptachlor and heptachlor epoxide are active inducers of microsomal epoxidation (Gillett and Chan 1968). In male Wistar rats fed a diet containing 5 ppm heptachlor or heptachlor epoxide for 10 days, the latter was much more effective in inducing epoxidation of aldrin than heptachlor (Gillett and Chan 1968). The minimally effective dietary concentration for inducing significant epoxidation was estimated to be between 1 and 5 ppm for both compounds (Gillett and Chan 1968). Heptachlor epoxide is considered more toxic than its parent compound and, like heptachlor, is primarily stored in adipose tissue (Barquet et al. 1981; Burns 1974; Greer et al. 1980; Harradine and McDougall 1986).

3.4.4 Elimination and Excretion

3.4.4.1 Inhalation Exposure

No studies were located regarding excretion in humans or animals after inhalation exposure to heptachlor or heptachlor epoxide. Based on the data from oral studies, heptachlor is expected to be excreted primarily in the form of metabolites and also as unchanged parent compound.

Figure 3-2. Metabolic Scheme for Heptachlor in Rats



Source: adapted from Tashiro and Matsumura (1978)

3.4.4.2 Oral Exposure

No studies were located regarding excretion in humans after oral exposure to heptachlor or heptachlor epoxide.

Due to their relatively high lipid solubility, heptachlor and heptachlor epoxide can accumulate in breast milk. For example, heptachlor and heptachlor epoxide were measured in 51 samples of human milk at average concentrations of 0.0027 and 0.019 ppm, respectively, from women with unknown exposure histories (Jonsson et al. 1977). Heptachlor epoxide was found in 24% of the samples, and heptachlor was found in 6%. Other investigators have reported the presence of heptachlor epoxide in human milk at concentrations ranging from not detected to 0.46 ppm (Kroger 1972; Larsen et al. 1971; Mussalo-Rauhamaa et al. 1988; Polishuk et al. 1977b; Ritcey et al. 1972; Savage et al. 1981; Takei et al. 1983). These findings suggest a potential for transfer to the nursing infant (see also Sections 3.5.1 and 6.5).

In a study in cows, the concentration of heptachlor epoxide in cow's milk reached a maximum within 3–7 days after the cows began grazing 18 hours/day on pastures immediately following treatment of the grasses with heptachlor and declined steadily thereafter. The level of heptachlor epoxide in the milk reached a concentration of 0.22 ppm (Gannon and Decker 1960).

The elimination of a single oral gavage dose of ^{14}C-heptachlor in male rats showed that most of the radioactivity was eliminated in the feces (Tashiro and Matsumura 1978). One day after dosing, 36% of the dose had been eliminated, and by day 10, approximately 62% had been eliminated in the feces. Elimination of the radioactive label in urine accounted for only 6% of the total dose in 10 days. Approximately 26.2% of the total radioactivity recovered from the feces was the parent compound and the remainder was in the form of metabolites.

3.4.4.3 Dermal Exposure

No studies were located regarding excretion in humans or animals after dermal exposure to heptachlor or heptachlor epoxide. Based on the data from oral studies, heptachlor is expected to be excreted primarily in the form of metabolites and also as unchanged parent compound.

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

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

PBPK/PD models refine our understanding of complex quantitative dose behaviors by helping to delineate and characterize the relationships between: (1) the external/exposure concentration and target tissue dose of the toxic moiety, and (2) the target tissue dose and observed responses (Andersen and Krishnan 1994; Andersen et al. 1987). These models are biologically and mechanistically based and can be used to extrapolate the pharmacokinetic behavior of chemical substances from high to low dose, from route to route, between species, and between subpopulations within a species. The biological basis of PBPK models results in more meaningful extrapolations than those generated with the more conventional use of uncertainty factors.

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

The structure and mathematical expressions used in PBPK models significantly simplify the true complexities of biological systems. If the uptake and disposition of the chemical substance(s) are adequately described, however, this simplification is desirable because data are often unavailable for

many biological processes. A simplified scheme reduces the magnitude of cumulative uncertainty. The adequacy of the model is, therefore, of great importance, and model validation is essential to the use of PBPK models in risk assessment.

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

If PBPK models for heptachlor and heptachlor epoxide 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 PBPK models were identified for heptachlor or heptachlor epoxide.

3.5 MECHANISMS OF ACTION

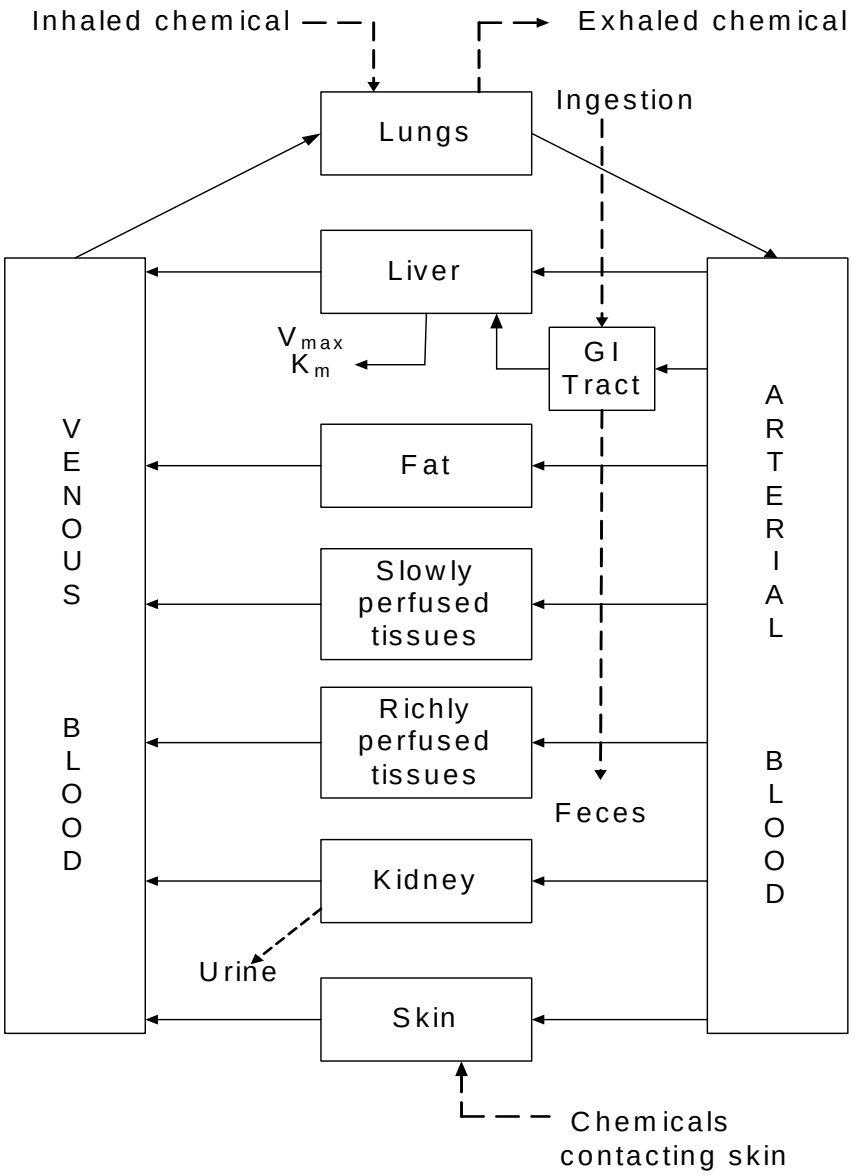
3.5.1 Pharmacokinetic Mechanisms

There is limited information on the toxicokinetics of heptachlor. No dermal toxicokinetic data were located. Heptachlor is absorbed via the lungs and digestive tract, although the site and mechanism of absorption are not known. Heptachlor is metabolized to heptachlor epoxide and is stored in the body as the parent compound and as this metabolite. Heptachlor and heptachlor epoxide are highly lipid soluble and are stored in adipose tissue; both compounds can also accumulate in breast milk. Heptachlor is primarily excreted in the feces as heptachlor epoxide.

3.5.2 Mechanisms of Toxicity

The available data suggest that the developing nervous system is the most sensitive target of heptachlor toxicity. Impaired spatial memory was observed in rats exposed to 0.03 mg/kg/day heptachlor during gestation and from postnatal day 7–42 (Moser et al. 2001). The cause of these alterions is not known. Moser et al. (2001) noted that heptachlor and other cyclodiene insecticides have a high affinity for $GABA_A$ (gamma-amino butyric acid) receptors and can alter the expression of the $GABA_A$ receptor

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



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

Source: adapted from Krishnan and Andersen 1994

during development. In the Moser et al. (2001) study, alterations in $GABA_A$ binding sites were observed in the brainstem of female rats, but not in the cortex. However, no alterations in the functional response of the GABA receptor binding were observed. This study does not address whether the heptachlor-induced alterations in $GABA_A$ receptors and the observed neurobehavioral alterations are related.

3.5.3 Animal-to-Human Extrapolations

There are limited available data with which to compare humans and other animal species. The absorption and distribution properties of heptachlor and heptachlor epoxide appear to be the same in both humans and animals. For the most part, the human toxicity data do not allow for quantitative or qualitative comparisons with the available animal data; an exception is the neurodevelopmental toxicity data. Impaired performance on tests of abstract concept formation, visual perception, and motor planning was observed in adolescents exposed during gestation to heptachlor and/or heptachlor epoxide (Baker et al. 2004b). In rats exposed during gestation and for the first 42 postnatal days, impaired spatial memory and learning were observed (Moser et al. 2001). These data provide some qualitative support for extrapolating the rat data for human risk assessment.

3.6 TOXICITIES MEDIATED THROUGH THE NEUROENDOCRINE AXIS

Recently, attention has focused on the potential hazardous effects of certain chemicals on the endocrine system because of the ability of these chemicals to mimic or block endogenous hormones. Chemicals with this type of activity are most commonly referred to as endocrine disruptors. However, appropriate terminology to describe such effects remains controversial. The terminology endocrine disruptors, initially used by Thomas and Colborn (1992), was also used in 1996 when Congress mandated the EPA to develop a screening program for "...certain substances [which] may have an effect produced by a naturally occurring estrogen, or other such endocrine effect[s]...". To meet this mandate, EPA convened a panel called the Endocrine Disruptors Screening and Testing Advisory Committee (EDSTAC), and in 1998, the EDSTAC completed its deliberations and made recommendations to EPA concerning endocrine disruptors. In 1999, the National Academy of Sciences released a report that referred to these same types of chemicals as hormonally active agents. The terminology endocrine modulators has also been used to convey the fact that effects caused by such chemicals may not necessarily be adverse. Many scientists agree that chemicals with the ability to disrupt or modulate the endocrine system are a potential threat to the health of humans, aquatic animals, and wildlife. However, others think that endocrine-active chemicals do not pose a significant health risk, particularly in view of the fact that hormone mimics exist in the natural environment. Examples of natural hormone mimics are the isoflavinoid phytoestrogens

(Adlercreutz 1995; Livingston 1978; Mayr et al. 1992). These chemicals are derived from plants and are similar in structure and action to endogenous estrogen. Although the public health significance and descriptive terminology of substances capable of affecting the endocrine system remains controversial, scientists agree that these chemicals may affect the synthesis, secretion, transport, binding, action, or elimination of natural hormones in the body responsible for maintaining homeostasis, reproduction, development, and/or behavior (EPA 1997). Stated differently, such compounds may cause toxicities that are mediated through the neuroendocrine axis. As a result, these chemicals may play a role in altering, for example, metabolic, sexual, immune, and neurobehavioral function. Such chemicals are also thought to be involved in inducing breast, testicular, and prostate cancers, as well as endometriosis (Berger 1994; Giwercman et al. 1993; Hoel et al. 1992).

No studies were located regarding endocrine disruption in humans or animals after exposure to heptachlor or heptachlor epoxide. An animal study examining the impact of in utero and lactational exposure to heptachlor on the development of the reproductive system (Smialowicz et al. 2001) did not find alterations in vaginal opening, prepuce separation, or adverse reproductive or developmental outcomes when exposed offspring were mated with controls. Additionally, no *in vitro* studies were located regarding endocrine disruption of heptachlor or heptachlor epoxide.

3.7 CHILDREN'S SUSCEPTIBILITY

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

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

Children sometimes differ from adults in their susceptibility to hazardous chemicals, but whether there is a difference depends on the chemical (Guzelian et al. 1992; NRC 1993). Children may be more or less susceptible than adults to health effects, and the relationship may change with developmental age (Guzelian et al. 1992; NRC 1993). Vulnerability often depends on developmental stage. There are

critical periods of structural and functional development during both prenatal and postnatal life, and a particular structure or function will be most sensitive to disruption during its critical period(s). Damage may not be evident until a later stage of development. There are often differences in pharmacokinetics and metabolism between children and adults. For example, absorption may be different in neonates because of the immaturity of their gastrointestinal tract and their larger skin surface area in proportion to body weight (Morselli et al. 1980; NRC 1993); the gastrointestinal absorption of lead is greatest in infants and young children (Ziegler et al. 1978). Distribution of xenobiotics may be different; for example, infants have a larger proportion of their bodies as extracellular water, and their brains and livers are proportionately larger (Altman and Dittmer 1974; Fomon 1966; Fomon et al. 1982; Owen and Brozek 1966; Widdowson and Dickerson 1964). The infant also has an immature blood-brain barrier (Adinolfi 1985; Johanson 1980) and probably an immature blood-testis barrier (Setchell and Waites 1975). Many xenobiotic metabolizing enzymes have distinctive developmental patterns. At various stages of growth and development, levels of particular enzymes may be higher or lower than those of adults, and sometimes unique enzymes may exist at particular developmental stages (Komori et al. 1990; Leeder and Kearns 1997; NRC 1993; Vieira et al. 1996). Whether differences in xenobiotic metabolism make the child more or less susceptible also depends on whether the relevant enzymes are involved in activation of the parent compound to its toxic form or in detoxification. There may also be differences in excretion, particularly in newborns who all have a low glomerular filtration rate and have not developed efficient tubular secretion and resorption capacities (Altman and Dittmer 1974; NRC 1993; West et al. 1948). Children and adults may differ in their capacity to repair damage from chemical insults. Children also have a longer remaining lifetime in which to express damage from chemicals; this potential is particularly relevant to cancer.

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

There are suggestive data indicating that children, particularly children exposed *in utero* and during infancy may be unusually susceptible to the toxicity of heptachlor. Although no studies comparing the toxicity of heptachlor in adults and children were identified, there is a possibility that very young children may exhibit particular susceptibility to hepatic effects because of the immaturity of the hepatic microsomal system. Heptachlor is bioactivated to produce heptachlor epoxide, which is more toxic than

heptachlor. Pre-adolescent children have a greater rate of glutathione turnover, and they are expected to be more susceptible to heptachlor epoxide-induced toxicity. Their susceptibility would probably depend upon their ability to detoxify heptachlor epoxide. However, Harbison (1975) observed that heptachlor was less toxic in newborn rats than in adult rats. Newborn rats pretreated with phenobarbital were more sensitive to the effects of heptachlor than those not pretreated. Thus, the ability to metabolize and bioactivate heptachlor correlates with its toxicity in the newborn.

Several developmental toxicity studies have identified the developing organisms as a sensitive subpopulation. Heptachlor exposure does not appear to increase the risk of malformations in humans (Le Marchand et al. 1986) or animals (Narotsky et al. 1995; Smialowicz et al. 2001), although increases in pup mortality (Narotsky et al. 1995; Purkerson-Parker et al. 2001b) and decreases in pup body weight (Narotsky and Kavlock 1995; Narotsky et al. 1995) have been observed in animal studies. There is some indication that the developing nervous system may be unusually susceptible to the toxicity of heptachlor. A study of high school students exposed to heptachlor epoxide *in utero* and during early childhood found impaired performance on tests of abstract concept formation, visual perception, and motor planning (Baker et al. 2004b). Delays in the righting reflex, slowed acquisition of a spatial task, and impaired recall were observed in rat offspring exposed during gestation, lactation, and postnatally (Moser et al. 2001; Purkerson-Parker et al. 2001b). The impaired learning and memory was the basis of the intermediate-duration oral MRL. In addition to the neurological effects, suppression of the immune response to sheep red blood cells was observed at the same dose level as the impaired learning and memory (Smialowicz et al. 2001).

3.8 BIOMARKERS OF EXPOSURE AND EFFECT

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

Due to a nascent understanding of the use and interpretation of biomarkers, implementation of biomarkers as tools of exposure in the general population is very limited. A biomarker of exposure is a xenobiotic substance or its metabolite(s) or the product of an interaction between a xenobiotic agent and some target molecule(s) or cell(s) that is measured within a compartment of an organism (NAS/NRC 1989). The preferred biomarkers of exposure are generally the substance itself, substance-specific metabolites in readily obtainable body fluid(s), or excreta. However, several factors can confound the use and

interpretation of biomarkers of exposure. The body burden of a substance may be the result of exposures from more than one source. The substance being measured may be a metabolite of another xenobiotic substance (e.g., high urinary levels of phenol can result from exposure to several different aromatic compounds). Depending on the properties of the substance (e.g., biologic half-life) and environmental conditions (e.g., duration and route of exposure), the substance and all of its metabolites may have left the body by the time samples can be taken. It may be difficult to identify individuals exposed to hazardous substances that are commonly found in body tissues and fluids (e.g., essential mineral nutrients such as copper, zinc, and selenium). Biomarkers of exposure to heptachlor and heptachlor epoxide are discussed in Section 3.8.1.

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

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

3.8.1 Biomarkers Used to Identify or Quantify Exposure to Heptachlor and Heptachlor Epoxide

Heptachlor and heptachlor epoxide can be measured in blood, adipose tissue, breast milk, and urine. The analytical methods available can be used to determine whether exposure has occurred, but the results cannot tell whether adverse health effects will occur. The presence of heptachlor epoxide may reflect an exposure to heptachlor or possibly chlordane since heptachlor epoxide is a metabolite of both these pesticides. However, in the absence of stable chlordane residues (e.g., nonachlor and oxychlordane), the heptachlor epoxide would most likely have been derived from heptachlor.

Detection of heptachlor or heptachlor epoxide in the body may indicate either recent or past exposure. Heptachlor epoxide has a long half-life, particularly in adipose tissue because it is very lipophilic and can remain dissolved in adipose tissue for months to years. An example of this can be found in a report in which analysis of human adipose tissue samples obtained during autopsy between 1987 and 1988 from residents of North Texas showed that tissue levels of heptachlor epoxide in subjects from the above geographical region had not significantly decreased since 1970 (Adeshina and Todd 1990). However, heptachlor epoxide is eventually mobilized into the blood and subsequently to the liver for further breakdown. Blood levels of heptachlor epoxide are often taken to indicate a more recent exposure.

As indicated in Section 3.4.4.2, due to their relatively high lipid solubility, heptachlor and heptachlor epoxide can accumulate in breast milk fat. Heptachlor and heptachlor epoxide were measured in 51 samples of human milk at average concentrations of 0.0027 and 0.019 ppm, respectively, from women with unknown exposure histories (Jonsson et al. 1977). Heptachlor epoxide was found in 24% of the samples, and heptachlor was found in 6%. Other investigators have reported the presence of heptachlor epoxide in human milk at concentrations ranging from not detected to 0.46 ppm (Kroger 1972; Polishuk et al. 1977b; Savage et al. 1981; Takei et al. 1983). These findings suggest a potential for transfer to the nursing infant (see also Sections 3.5.1 and 6.5). Other studies that have reported levels of heptachlor or heptachlor epoxide in humans' breast milk include Larsen et al. (1971), Ritcey et al. (1972), and Mussalo-Rauhamaa et al. (1988).

No studies were found correlating levels to which humans were exposed with actual body burdens.

3.8.2 Biomarkers Used to Characterize Effects Caused by Heptachlor and Heptachlor Epoxide

No clinical conditions due to specific exposure to heptachlor or heptachlor epoxide are known. The neurological and hepatic effects seen from exposure to heptachlor and heptachlor epoxide are typical of exposure to other chlorinated pesticides.

3.9 INTERACTIONS WITH OTHER CHEMICALS

Dietary administration of heptachlor (97.6% purity) at 0.65 or 1.3 mg/kg/day in diet for 25 weeks promoted the development of hepatocellular foci and hepatocellular neoplasms in male B6C3F_1 mice previously initiated with 3.8 mg/kg/day diethylnitrosamine given in the drinking water for 14 weeks (Williams and Numoto 1984).

Nutritional factors may influence the toxicity of pesticides. Research in this area has primarily focused on the role of dietary proteins, particularly sulfur-containing amino acids, trace minerals, and vitamins A, C, D, and E. Studies in rats show that inadequate dietary protein enhances the toxicity of most pesticides but decreases, or fails to affect, the toxicity of a few. The results of these studies have shown that at one-seventh or less normal dietary protein, the hepatic toxicity of heptachlor is diminished as evidenced by fewer enzyme changes (Boyd 1969; Shakman 1974). The lower-protein diets may decrease metabolism of heptachlor to heptachlor epoxide.

Male weanling rats were fed a 5, 20, or 40% casein diet for 10 days and then given heptachlor intraperitoneally. The animals receiving the 5% casein diet showed a 3-fold tolerance to heptachlor toxicity, but the toxicity of heptachlor epoxide was not affected (Weatherholtz et al. 1969). This was probably due to inability of weanling rats to metabolically convert heptachlor to the more toxic heptachlor epoxide. This fact is further supported by the observation that changes in protein percentage in diet did not affect the toxicity of heptachlor epoxide itself.

Walter Reed-Wistar and Charles River male adult rats were exposed to oral doses of turpentine or to turpentine vapors, which consisted of α- and β-pinene. These exposures were followed by oral administration of heptachlor epoxide or of one of three pesticides, paraoxon, heptachlor, or parathion, or by an intraperitoneal injection of hexobarbital. The studies revealed that pretreatment with turpentine reduced hexobarbital sleeping time, reduced the parathion LD_{50}, and increased the heptachlor LD_{50}. The paraoxon and heptachlor epoxide LD_{50} values were unchanged. α-Pinene and β-pinene vaporized from turpentine had no effect on either hexobarbital sleeping time or parathion, paraoxon, or heptachlor epoxide mortality but did increase the heptachlor LD_{50} (Sperling et al. 1972). The authors speculated that increases in hepatic microsomal enzyme activity are responsible for these differences.

3.10 POPULATIONS THAT ARE UNUSUALLY SUSCEPTIBLE

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

exposure to heptachlor and heptachlor epoxide are discussed in Section 6.7, Populations with Potentially High Exposures.

3.11 METHODS FOR REDUCING TOXIC EFFECTS

This section will describe clinical practice and research concerning methods for reducing toxic effects of exposure to heptachlor and heptachlor epoxide. 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 heptachlor and heptachlor epoxide. 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 heptachlor and heptachlor epoxide:

Ellenhorn MJ. 1997. Ellenhorn's medical toxicology. Diagnosis and treatment of human poisoning. 2nd ed. Baltimore, MD: Williams and Wilkins, 1614-1631.

EPA. 1999. Recognition and management of pesticide poisonings. 5th. Washington, DC: U.S. Environmental Protection Agency. EPA735R98003. PB99149551.

Goldfrank L, Flomenbaum N, Lewin N, et al. 2002. Goldfrank's toxicologic emergencies. 7th ed. New York, NY: McGraw-Hill, 1366-1378.

3.11.1 Reducing Peak Absorption Following Exposure

Human exposure to heptachlor or heptachlor epoxide can occur by inhalation, oral, or dermal contact. Treatment of exposure to these substances is primarily supportive. Following a significant inhalation exposure, the patient is removed from the source to fresh air. Treatment may include administering oxygen and, if needed, maintaining ventilation with artificial respiration (Bronstein and Currance 1988; HSDB 2007a). General recommendations for reducing absorption of heptachlor following acute dermal exposure have included removal of contaminated clothing followed by washing the skin and hair with soap and water, (HSDB 2007a; Morgan 1989). Since leather absorbs pesticides, it has been recommended that leather not be worn while using heptachlor or heptachlor epoxide, and that any leather contaminated with these substances be discarded (HSDB 2007a). Oils have not been recommended as dermal cleansing agents because they could increase absorption (Haddad and Winchester 1990). If the eyes have been exposed, they are flushed with water (Bronstein and Currance 1988; HSDB 2007a). Treatment for ingestion of this substance may require gastric emptying by gastric lavage (Haddad and Winchester 1990) and administration of activated charcoal and cathartic (Haddad and Winchester 1990; HSDB 2007a; Morgan 1989). Heptachlor may be present with a hydrocarbon vehicle, which could result

in aspiration pneumonitis following the induction of emesis. Therefore, emesis may not be indicated. Some sources do not recommend the use of emetics (Bronstein and Currance 1988), although others do under some circumstances (HSDB 2007a; Morgan 1989). Treatments such as emesis and lavage may be most appropriate following ingestion of large quantities; it is unlikely that the types of exposure likely to occur at hazardous waste sites would require such measures. Treatment with milk, cream, or other substances containing vegetable or animal fats, which enhance absorption of chlorinated hydrocarbons, has not been recommended (Haddad and Winchester 1990; Morgan 1989). If seizures occur, diazepam administration, followed if necessary by additional anticonvulsant medicines such as phenytoin, pentobarbital, thiopental, or succinylcholine, may be recommended (Bronstein and Currance 1988; HSDB 2007a; Morgan 1989). As adrenergic amines, such as epinephrine, may further increase myocardial irritability and produce refractory ventricular arrhythmias, their use has not been recommended (Bronstein and Currance 1988; Haddad and Winchester 1990; HSDB 2007a; Morgan 1989).

3.11.2 Reducing Body Burden

Heptachlor is rapidly metabolized by the body, mostly to heptachlor epoxide. Most of the metabolites are rapidly excreted in the feces, with the adipose tissue serving as the major storage depot for the remainder. From the fat, heptachlor epoxide can be slowly released into the bloodstream for further metabolism and excretion. Cholestyramine resin may accelerate the biliary-gastrointestinal excretion of the more slowly eliminated organochlorine compounds, and its use has been suggested (Morgan 1989). Because of the lipophilicity of heptachlor and heptachlor epoxide, dialysis and exchange transfusion are thought to be ineffective (HSDB 2007a).

Because heptachlor epoxide is lipophilic, it is likely that the loss of adipose tissue, as may occur during fasting, will mobilize the stored compound and increase the rate of its elimination. However, this mobilization is also likely to temporarily increase the blood levels of heptachlor epoxide. Hence, any possible benefits due to a reduced body burden accompanying fat reduction would need to be balanced against potential harmful results due to the expected temporary increase in blood levels.

3.11.3 Interfering with the Mechanism of Action for Toxic Effects

Since the metabolized form of heptachlor, heptachlor epoxide, is the most toxic, it may be possible to reduce the toxic effects of heptachlor by inhibiting the enzyme catalyzing this conversion. This is the same enzyme that catalyzes the epoxidation of aldrin to dieldrin (Gillett and Chan 1968). Further

research into the specificity of this enzyme, drugs that could inhibit the enzyme, and any side effects of these drugs could help to determine the feasibility of such a treatment strategy.

In the central nervous system, symptoms observed in animals following exposure include tremors, convulsions, ataxia, and changes in EEG patterns (Formanek et al. 1976). These central nervous system symptoms could be due either to (1) inhibition of the Na^+/K^+ ATPase or the Ca^+/Mg^+ ATPase activity, which can then interfere with nerve action or release of neurotransmitters (Yamaguchi et al. 1979) and/or (2) inhibition of the function of the receptor for GABA (Yamaguchi et al. 1980). In support of the latter possibility, another study showed that heptachlor epoxide inhibited the GABA-stimulated chloride uptake in the coxal muscle of the American cockroach and directly competed against [^{3}H]a-dihydropierotoxinin for binding in the rat brain synaptosomes. These results indicate that some of the nerve excitation symptoms that insecticides cause are probably due to their interaction with the picrotoxin binding site of the GABA receptor (Matsumura and Ghiasuddin 1983). A more detailed understanding of the mechanism of heptachlor/heptachlor epoxide action on the central nervous system may lead to new approaches for reducing the toxic effects.

3.12 ADEQUACY OF THE DATABASE

Section 104(I)(5) of CERCLA, as amended, directs the Administrator of ATSDR (in consultation with the Administrator of EPA and agencies and programs of the Public Health Service) to assess whether adequate information on the health effects of heptachlor and heptachlor epoxide 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 heptachlor and heptachlor epoxide.

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

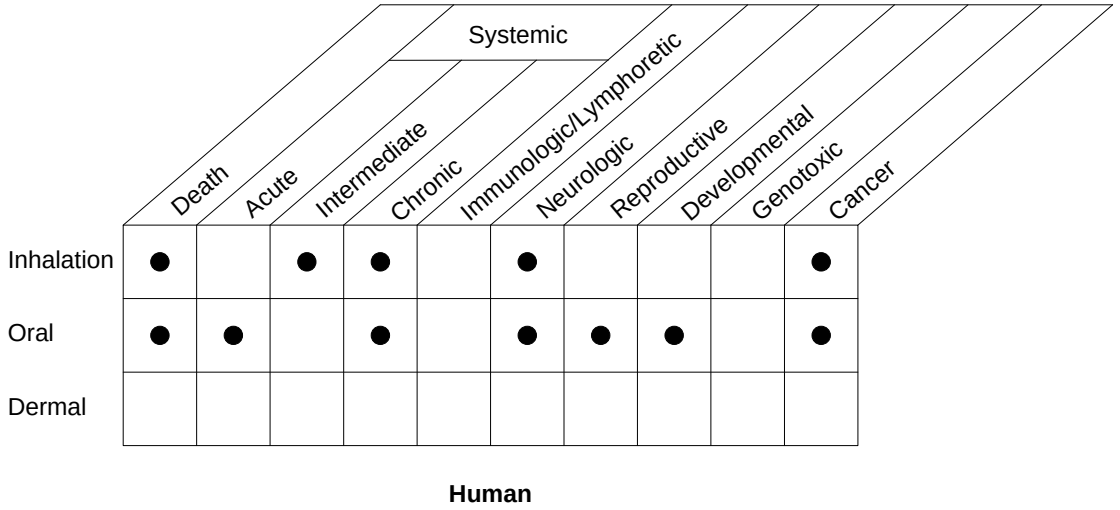
3.12.1 Existing Information on Health Effects of Heptachlor and Heptachlor Epoxide

The existing data on health effects of inhalation, oral, and dermal exposure of humans and animals to heptachlor and heptachlor epoxide are summarized in Figure 3-4. The purpose of this figure is to illustrate the existing information concerning the health effects of heptachlor and heptachlor epoxide. Each dot in the figure indicates that one or more studies provide information associated with that particular effect. The dot does not necessarily imply anything about the quality of the study or studies, nor should missing information in this figure be interpreted as a "data need". A data need, as defined in ATSDR's Decision Guide for Identifying Substance-Specific Data Needs Related to Toxicological Profiles (Agency for Toxic Substances and Disease Registry 1989), is substance-specific information necessary to conduct comprehensive public health assessments. Generally, ATSDR defines a data gap more broadly as any substance-specific information missing from the scientific literature.

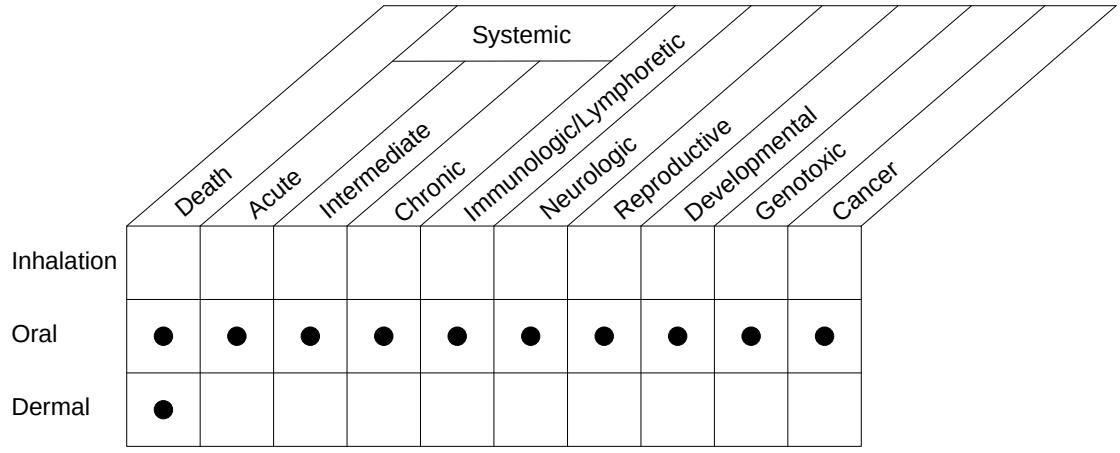
Most of the data located concerning the health effects of heptachlor and heptachlor epoxide in humans come from case reports and occupational epidemiology studies of workers engaged either in the manufacture or application of pesticides. There is some information on people who have consumed heptachlor-contaminated food or dairy products, but no adverse health effects have been related to these exposures. The occupational studies involve exposures that are predominantly inhalation with contributions from dermal exposure, whereas all the animal studies were conducted using oral or intraperitoneal exposures. The occupational and case reports provide no quantitation of dose or duration of exposure, which makes it impossible to determine with any precision the effect levels for humans. There are no data that indicate that heptachlor or heptachlor epoxide are carcinogenic to humans. However, human studies are limited by the long latency period of carcinogenesis and by ascertainment and follow-up biases.

The animal studies for oral exposure to heptachlor and heptachlor epoxide are almost all limited to some extent by the number of doses used, the lack of appropriate statistics, or the small number or lack of controls. No information was located regarding the health effects of inhalation or dermal exposure, with the exception of a dermal LD_{50} in rats. Exposure of the general population via the inhalation and dermal routes may result from contaminated soil or vapors from treated houses. Some exposures from contaminated soil or water may occur in populations located near hazardous waste sites in which these chemicals have been stored or from food grown in contaminated soil.

Figure 3-4. Existing Information on Health Effects of Heptachlor and Heptachlor Epoxide



Human



Animal

● Existing Studies

3.12.2 Identification of Data Needs

Acute-Duration Exposure. There are no studies that have evaluated the acute toxicity of heptachlor or heptachlor epoxide following inhalation exposure; thus, acute-duration inhalation MRLs were not derived. A number of studies have evaluated the toxicity of heptachlor following acute oral exposure. The results of these studies suggest several sensitive targets of toxicity including the liver, nervous system, reproductive system, and the developing organism (Amita Rani and Krishnakumari 1995; Berman et al. 1995; Krampl 1971; Narotsky and Kavlock 1995; Narotsky et al. 1995; Purkerson-Parker et al. 2001b). The available data suggest that the most sensitive effect is impaired fertility observed in female rats administered heptachlor for 14 days prior to mating (Amita Rani and Krishnakumari 1995); this end point was used to derive an acute-duration oral MRL. Additional studies that examined a variety of systemic, neurological, reproductive, and developmental end points are needed to support the identification of critical effect and to establish dose-response relationships. Although there are limited toxicokinetic and mechanistic data for heptachlor, it is likely that its toxicity is not route-specific. The identified target organs would likely be the same for oral, inhalation, and dermal exposure; however, it is not possible to predict threshold concentrations. Toxicokinetic studies, which would allow for route-to-route extrapolation, and inhalation and dermal toxicity studies would be useful for confirming whether the toxicity of heptachlor is independent of route of exposure.

The available acute-duration studies for heptachlor epoxide are limited to oral lethality and dominant lethal studies (Epstein et al. 1972; Podowski et al. 1979). The toxicity of heptachlor is likely due to heptachlor epoxide; thus, the targets of toxicity are likely to be the same as those observed following heptachlor exposure. However, the toxic thresholds are likely to be different. Studies are needed to establish dose-response relationships and to confirm whether the targets of toxicity are the same as those identified for heptachlor.

Intermediate-Duration Exposure. The targets of toxicity of heptachlor following intermediate-duration oral exposure appear to be the same as those identified following acute-duration oral exposure and include the liver, nervous system, reproductive system, and the developing organism (Akay and Alp 1981; Amita Rani and Krishnakumari 1995; Aulerich et al. 1990; Crum et al. 1993; Izushi and Ogata 1990; Moser et al. 2001; NCI 1977; Pelikan 1971; Purkerson-Parker et al. 2001b; Smialowicz et al. 2001). Of these targets, the developing organism appears to be the most sensitive (Moser et al. 2001; Smialowicz et al. 2001). Impaired development of the nervous and immune systems have been observed

in rats exposed to heptachlor *in utero*, during lactation, and from postnatal day 7 through 42; a NOAEL has not been identified for these end points. The intermediate-duration oral MRL for heptachlor was based on these developmental effects. The potential systemic toxicity of heptachlor has not been adequately assessed; although several studies have evaluated systemic end points (Akay and Alp 1981; Akay et al. 1982; Akhtar et al. 1996; Izushi and Ogata 1990; Pelikan 1971), many of these studies were poorly reported or examined a limited number of end points. Studies examining a variety of systemic end points would be useful for identifying target tissues and establishing dose-response relationships. No intermediate-duration inhalation or dermal studies were identified. As discussed in the Acute-Duration Exposure section, it is likely that the targets of toxicity would be the same for inhalation, oral, and dermal exposure; however, toxicokinetic data are not available to confirm this conjecture. Inhalation and dermal exposure studies are needed.

No publicly available studies on the intermediate-duration toxicity of heptachlor epoxide were identified. A 60-day dog study, which was submitted to EPA under the Federal Insecticide, Fungicide, and Rodenticide Act (FIFRA) identifies the liver as a critical target of toxicity (IRIS 2006). It is likely that the targets of heptachlor epoxide toxicity are the same as those for heptachlor. Additional studies, particularly those examining the development of the nervous and immune systems are needed to identify targets of toxicity and establish dose-response relationships.

Chronic-Duration Exposure and Cancer. There are no data on chronic oral exposures in humans. There are occupational studies of workers engaged in the manufacture of heptachlor in which the exposures are presumed to be predominantly inhalation with contributions from the dermal route. No adverse health effects have been identified in these cohorts that could be positively associated with heptachlor exposure (Infante et al. 1978; MacMahon et al. 1988; Stehr-Green et al. 1988). There is a limited publicly available database on the chronic oral toxicity of heptachlor. The database is limited to a multigeneration study (Mestitzova 1967), which reported increased postnatal mortality at the lowest dose tested, and a study examining a limited number of noncancer end points (NCI 1977); thus, the database was not considered adequate for derivation of a chronic duration oral MRL. A 2-year study submitted to EPA under FIFRA identified the liver as a critical target of toxicity (IRIS 2006). This finding is consistent with the available intermediate-duration studies. However, intermediate-duration studies have also identified the nervous system, reproductive system, and the developing organism as targets of toxicity. Additional studies are needed to identify the most sensitive target following chronic duration exposure and to establish dose-response relationships; these studies would be useful for deriving an oral

MRL. Inhalation and dermal studies are also needed; these studies in animals would be useful for determining whether the target organ is the same across routes of exposure.

As with the other durations of exposure, limited publicly available data were located for heptachlor epoxide. A study submitted to EPA under FIFRA identified the liver as the most sensitive target of toxicity (IRIS 2006). Additional studies are needed that could be used to derive inhalation and oral MRLs for heptachlor epoxide and to establish the targets of toxicity for dermal exposure.

There are occupational mortality studies that have collected data appropriate for determining whether those engaged in the manufacture or application of heptachlor are at increased risk for dying of cancer. These studies have not shown an increased risk of cancer mortality (Infante et al. 1978; MacMahon et al. 1988). Occupational studies that collected cancer incidence data, rather than just mortality data, would be useful for further exploration of this issue. Carcinogenicity studies have been identified for rats and mice (NCI 1977). These data show increases in tumorigenesis following exposure to heptachlor. Chronic studies of inhalation exposure in relation to oncogenesis in animals might be useful for determining mechanism of action and the consistency of effect across routes of exposure. There are no toxicokinetic data that indicate that there will be route-specific differences.

Genotoxicity. Information on the *in vivo* genotoxic effects of heptachlor or heptachlor epoxide is limited to dominant lethality assays with negative results (Arnold et al. 1977; Epstein et al. 1972). More case reports and epidemiology studies are needed to properly evaluate genotoxic effects in humans exposed to heptachlor or heptachlor epoxide. The results of *in vitro* studies suggest that neither compound alters the frequency of gene mutations (Crebelli et al. 1986; Gentile et al. 1982; Glatt et al. 1983; Marshall et al. 1976; NTP 1987; Probst et al. 1981; Zeiger et al. 1987), and that heptachlor does not induce DNA damage in bacteria (Rashid and Mumma 1986) or rodents (Maslansky and Williams 1981; Probst et al. 1981). Alterations were observed in assays of unscheduled DNA synthesis in human fibroblasts (Ahmed et al. 1977) and chromosomal alterations in Chinese hamster ovary cells (NTP 1987). *In vivo* animal research into the effects of heptachlor and heptachlor epoxide on sister chromatid exchange, chromosomal aberrations and anomalies, DNA adduct formation, gene mutation, and other genotoxic parameters would be helpful in assessing the genotoxic potential of heptachlor and heptachlor epoxide.

Reproductive Toxicity. Although a couple of studies have attempted to establish an association between heptachlor epoxide blood levels and premature delivery or stillbirth among women presumably

exposed via ingestion (Curley et al. 1969; Wassermann et al. 1982), elevated levels of other compounds (particularly PCBs, lindane, and dieldrin) limit the interpretation of the results. Animal studies have found impaired fertility and pregnancy losses following oral exposure to heptachlor (Akay and Alp 1981; Amita Rani and Krishnakumari 1995; Green 1970; Mestitzova 1967). The mechanism of the reproductive toxicity has not been elucidated; the available data suggest that both males and females may be affected. Additional studies are needed to identify NOAELs for reproductive effects, confirm the observed results, and identify the critical targets within the reproductive system.

Developmental Toxicity. Several studies have examined the potential developmental toxicity of heptachlor or heptachlor epoxide. These studies examined potential effects in the children of women exposed to heptachlor and heptachlor epoxide in contaminated cow's milk (Baker et al. 2004b; Le Marchand et al. 1986) or examined the possible association between maternal heptachlor epoxide levels and developmental effects (Gladen et al. 2003; Hertz-Picciotto et al. 2004). Several animal studies have also examined developmental toxicity (Lawson and Luderer 2004; Moser et al. 2001; Narotsky and Kavlock 1995; Narotsky et al. 1995; Purkerson-Parker et al. 2001b; Smialowicz et al. 2001). The finding of impaired development of the nervous and immune systems was used as the basis of the intermediate-duration oral MRL for heptachlor. This study (Moser et al. 2001; Smialowicz et al. 2001) did not identify a NOAEL for these effects, additional studies would be useful for more clearly defining the threshold of toxicity. Impaired spatial memory was observed at the lowest dose tested among the offspring exposed until postnatal day 42, but not in rats exposed until postnatal day 21 (Moser et al. 2001); studies that would address the cause of the conflicting results would also be useful.

Immunotoxicity. No studies were located that specifically addressed immune function parameters following heptachlor or heptachlor epoxide exposure, although several studies have reported alterations in the lymphoreticular system (e.g., fibrosis in spleen, and increased size of spleen; decreased relative spleen weight) (Akay and Alp 1981; Aulerich et al. 1990; Pelikan 1971). A developmental toxicity study found suppression of the immune response in rats orally exposed *in utero*, during lactation, and postnatally until day 42 (Smialowicz et al. 2001). It is not known if these effects would also occur in mature animals. A study involving a battery of immune function tests would be useful for establishing whether heptachlor or heptachlor epoxide is toxic to the immune system.

Neurotoxicity. No human data on the neurotoxicity of heptachlor or heptachlor epoxide were identified. No data exist describing neurologic effects in animals following inhalation or dermal exposure of any duration. Several studies have demonstrated that exposure to heptachlor can result in neurological

effects, in particular, excitability and decreased motor activity (Akay and Alp 1981; Aulerich et al. 1990; Crum et al. 1993; Moser et al. 1995, 2003). As discussed in the Developmental Toxicity section, exposure to heptachlor (and presumably heptachlor epoxide) can result in impaired learning and memory; studies are needed to evaluate whether exposure of mature animals to heptachlor or heptachlor epoxide can also result in impaired learning and memory.

Epidemiological and Human Dosimetry Studies. The existing epidemiological studies are primarily of occupational cohorts (Blair et al. 1983; MacMahon et al. 1988; Shindell and Associates 1981; Wang and MacMahon 1979a), case reports of health effects seen in groups exposed to contaminated milk (Baker et al. 2004b; Chadduck et al. 1987; Le Marchand et al. 1986; Stehr-Green et al. 1986, 1988), or studies examining the possible association between elevated heptachlor/heptachlor epoxide blood levels and adverse health effects (Cantor et al. 2003; Cassidy et al. 2005; Curley et al. 1969; Gladen et al. 2003; Hertz-Picciotto et al. 2004; Pines et al. 1986; Quintana et al. 2004; Sturgeon et al. 1998; Wang and Grufferman 1981; Ward et al. 2000; Wassermann et al. 1982). These studies have generally not included good quantitation of the exposure to heptachlor or heptachlor epoxide. In many cases, it is not possible to determine the exact identity of the contaminants involved. Although use of this compound has been discontinued, exposure could nevertheless occur through food grown in contaminated soil, through contact with pesticides applied to homes and other structures, or from hazardous waste sites. Analytical methods are available to determine exposure to heptachlor or heptachlor epoxide (Curley et al. 1969; Klemmer et al. 1977; Radomski et al. 1968). However, no information is available that correlates levels of heptachlor epoxide in tissue with either level or duration of exposure. Occupational exposure levels are likely to be high enough to enable distinction from background levels. However, many epidemiological studies examining outcomes of exposure are limited by the accuracy of determining the exposure status of those individuals who show adverse health effects and those who show none. The precision and reliability of categorizing exposed individuals and non-exposed individuals contribute significantly to the statistical power of a study and greatly assist in accurate estimation of an increased risk. If data on exposure parameters are sparse or show very wide variation, it is difficult to determine what constitutes an exposure. More data on the correlation of tissue levels to exposure parameters would be useful for increasing the power of epidemiological studies to measure statistically significant associations between heptachlor exposure and health effects in cohorts from both occupational and contaminated community environments. Additionally epidemiology studies should focus on critical end points identified in animal studies including developmental toxicity (including neurological and immunological end points), liver effects, and cancer.

Biomarkers of Exposure and Effect.

Exposure. Exposure to heptachlor and heptachlor epoxide is currently measured by determining the level of these chemicals in the blood or adipose tissue in living organisms (Curley et al. 1969; Klemmer et al. 1977; Radomski et al. 1968). This measure is specific for both heptachlor and heptachlor epoxide. Heptachlor epoxide is also a metabolite of chlordane, and thus its presence is not specific for exposure to heptachlor alone. However, in the absence of stable chlordane residues (e.g., nonachlor and oxychlordane), the heptachlor epoxide would most likely have been derived from heptachlor. Because heptachlor is believed to be converted rapidly in the body to heptachlor epoxide, it is impossible to determine whether the exposure was to one or the other of these two compounds. Heptachlor and heptachlor epoxide accumulate in adipose tissue and are released slowly over long periods of time. Therefore, it is not possible to accurately identify whether the exposure was recent or what the duration of exposure was. However, the ratio of heptachlor epoxide to heptachlor increases over time and therefore may be used as a biomarker of possible exposure to heptachlor. The sensitivity of the methods for identifying these compounds in human tissue appears to be only sufficient to measure background levels of heptachlor epoxide in the population. Additional biomarkers of exposure to heptachlor would be helpful at this time.

Effect. There is no clinical disease state unique to heptachlor. A major problem in developing a biomarker of effect for heptachlor or heptachlor epoxide is that human exposures to these compounds have occurred concomitantly with exposures to other chemicals, and it is difficult to attribute the health effects to heptachlor or heptachlor epoxide alone. More data that quantify the biological effects as well as data that distinguish heptachlor and heptachlor epoxide exposures from those of other chemicals would be useful for developing biomarkers of effect for population monitoring. Biomarkers that could indicate the length of time since exposure would also be useful.

Absorption, Distribution, Metabolism, and Excretion. There are very few data available to assess the relative rates of pharmacokinetic parameters with respect to route of exposure for either heptachlor or heptachlor epoxide. There are no human or animal inhalation or dermal studies on absorption, distribution, metabolism, or excretion. The only human data on metabolism come from *in vitro* studies using liver microsomes that indicate that, qualitatively, human microsomes metabolize heptachlor to the same end products as do rat microsomes (Tashiro and Matsumura 1978). Oral exposure in members of farm families led to elevated serum levels of heptachlor metabolites (Stehr-Green et al. 1986), indicating that the compound is absorbed through the gastrointestinal tract. Animal studies also

suggest that uptake occurs through the gastrointestinal tract following oral dosing; excretion of these doses occurs primarily through the bile duct into the feces (Tashiro and Matsumura 1978). Lethality data suggest that heptachlor can be absorbed through the skin (Gaines 1969), but there are no data on absorption, distribution, metabolism, or excretion of dermally absorbed doses. Heptachlor epoxide is more toxic than heptachlor and has a longer half-life. Additional absorption, distribution, metabolism, and excretion data would be useful in order to gain a thorough understanding of the pharmacokinetic parameters of heptachlor and heptachlor epoxide.

Comparative Toxicokinetics. There are limited available data with which to compare humans and other animal species. Although human toxicity data are available (Baker et al. 2004b; Blair et al. 1983; Cantor et al. 2003; Cassidy et al. 2005; Chadduck et al. 1987; Curley et al. 1969; Gladen et al. 2003; Hertz-Picciotto et al. 2004; Le Marchand et al. 1986; MacMahon et al. 1988; Pines et al. 1986; Quintana et al. 2004; Shindell and Associates 1981; Stehr-Green et al. 1986, 1988; Sturgeon et al. 1998; Wang and Grufferman 1981; Wang and MacMahon 1979a; Ward et al. 2000; Wassermann et al. 1982), the results of these studies are difficult to compare with the animal studies (Akay and Alp 1981; Amita Rani and Krishnakumari 1995; Aulerich et al. 1990; Berman et al. 1995; Crum et al. 1993; Izushi and Ogata 1990; Krampl 1971; Moser et al. 2001; Narotsky and Kavlock 1995; Narotsky et al. 1995; NCI 1977; Pelikan 1971; Purkerson-Parker et al. 2001b; Smialowicz et al. 2001) because the exposure was not well characterized in the human studies and often involved exposure to multiple chemicals. As discussed in the previous section, there are limited data on the toxicokinetics of heptachlor and heptachlor epoxide. With the exception of human monitoring studies examining the levels of heptachlor/heptachlor epoxide in various tissues (Adeshina and Todd 1990; Barquet et al. 1981; Burns 1974; Greer et al. 1980; Klemmer et al. 1977; Polishuk et al. 1977b; Radomski et al. 1968; Stehr-Green et al. 1988; Wassermann et al. 1974), the available toxicokinetic data are in animals. Thus, direct comparisons between humans and animals can not be made. An *in vitro* comparative study found that the metabolites produced in humans and rats are identical, but the amounts differ (Tashiro and Matsumura 1978). Moreover, the rate of metabolism is not similar in both species. Thus, the rat may not be an appropriate metabolic model for humans. Additional studies, particularly *in vivo* studies, are needed to support these findings and identify the most appropriate animal model. There is a lack of information regarding kinetic changes after prolonged exposure. This kind of information would be useful because most exposures in the general population (e.g., from contaminated food or improperly applied pesticides) are likely to be long-term and low-dose.

Methods for Reducing Toxic Effects. The mechanism by which heptachlor and heptachlor epoxide are absorbed from the gastrointestinal tract is unknown. Current methods for reducing absorption

from the gastrointestinal tract involve removing these chemicals from the site of absorption (Haddad and Winchester 1990; HSDB 2007a; Morgan 1989). Additional studies examining the method of absorption would provide valuable information for developing methods that can interfere with gastrointestinal absorption. Numerous studies have examined the distribution of heptachlor and heptachlor epoxide (Barquet et al. 1981; Burns 1974; Curley et al. 1969; Greer et al. 1980; Jonsson et al. 1977; Polishuk et al. 1977b; Radomski et al. 1968). Additional studies on distribution are not necessary at this time. No established methods exist for reducing body burden of heptachlor and heptachlor epoxide. However, available information suggests that removal of these compounds via biliary-gastrointestinal excretion can be accelerated (Morgan 1989). Reducing enterohepatic recirculation before these chemicals partition to tissues may be effective (Haddad and Winchester 1990; HSDB 2007a). Thus, studies examining the effectiveness of repeated doses of activated charcoal or cholestyramine in reducing body burden would be useful. Adipose tissue serves as a major storage repository for both heptachlor and heptachlor epoxide (Barquet et al. 1981; Burns 1974; Greer et al. 1980; Harradine and McDougall 1986). Losing fat can mobilize the stored compound and increase the rate of its elimination. However, it may temporarily increase the blood levels of heptachlor epoxide. Studies that would examine the benefits of reducing body burden with accompanying fat reduction while balancing against harmful effects from temporary increase in blood level would be useful. Since heptachlor undergoes epoxidation to produce heptachlor epoxide which is more toxic than the parent compound, studies examining drugs that would inhibit the enzyme catalyzing this conversion would be helpful. Neurotoxicity of heptachlor epoxide is believed to result, at least in part, from interference with GABA receptor function (Yamaguchi et al. 1980). The available data suggest that benzodiazepenes and barbiturates may be useful in mitigating some of the neurological symptoms of heptachlor epoxide (Bronstein and Currance 1988; HSDB 2007b; Morgan 1989). However, additional studies examining the effectiveness of GABAergic function in mitigating heptachlor epoxide's neurologic effects would be useful. The liver also appears to be a major target organ for the toxic effects of heptachlor and heptachlor epoxide in animals (Akay and Alp 1981; Krampl 1971; Pelikan 1971). An understanding of the mechanism of action in the liver may identify new approaches for reducing the toxic effects.

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

There are suggestive data indicating that children, particularly children exposed *in utero* and during infancy, may be unusually susceptible to the toxicity of heptachlor. A study in adolescents exposed to

heptachlor and heptachlor epoxide during gestation found significant alterations in neurobehavioral function (Baker et al. 2004b). Similarly, alterations in neurobehavioral function were observed in rats exposed during gestation through postnatal day 42 (Moser et al. 2001). Because neurobehavioral performance has not been investigated in adults, it is difficult to determine whether children are more susceptible to the neurotoxicity of heptachlor than adults. Additionally, no studies have investigated whether there are age-specific differences in the toxicokinetic properties of heptachlor or heptachlor epoxide. Additional studies are needed to evaluate potential age-related differences in the toxicity of heptachlor and heptachlor epoxide.

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

3.12.3 Ongoing Studies

Patrick Wong at the University of California at Davis is investigating ligand-independent endocrine disruption by several pesticides include heptachlor epoxide. Ongoing research by D.E. Wooley, also at the University of California at Davis, is investigating the neurotoxic effects and mechanisms of action of environmental toxicants including heptachlor following acute and chronic exposure.