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 vinyl chloride. 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 adverse 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 produce 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 adverse 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 vinyl chloride are indicated in Tables 3-1 and 3-2 and Figures 3-1 and 3-2. Because cancer effects could occur at lower exposure levels, Figures 3-1 and 3-2 also shows a range for the upper bound of estimated excess risks, ranging from a risk of 1 in 10,000 to 1 in 10,000,000 (10^{-4} to 10^{-7}), as developed by EPA.

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

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

not protective in the case of altered health status caused by exposure to cigarette smoking or excessive alcohol consumption (i.e., altered lung and liver function).

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

A report by Danziger (1960) described the deaths of two vinyl chloride workers. In one case, a worker exposed to high concentrations of vinyl chloride emitted from an open valve was found dead. In another case, a worker responsible for cleaning a polymerization tank was found dead in the tank. Autopsies performed on these men showed congestion of the internal organs, particularly the lungs and kidneys, and failure of the blood to clot. Circumstances surrounding the deaths suggested that the deaths were due to breathing very high levels of vinyl chloride.

No increase in mortality was observed in 1,100 workers exposed to vinyl chloride compared to the same number of controls in a 7-year prospective cohort study of (Laplanche et al. 1992). At the time of the study interview, 36% of the 1,100 workers were currently being exposed to vinyl chloride, and 64% had been exposed in the past (Laplanche et al. 1987, 1992).

Brief exposures to concentrations of vinyl chloride ranging from 100,000 to 400,000 ppm have been shown to be fatal in experimental animals such as rats (Lester et al. 1963; Mastromatteo et al. 1960; Prodan et al. 1975), guinea pigs (Mastromatteo et al. 1960; Patty et al. 1930; Prodan et al. 1975), mice (Mastromatteo et al. 1960; Prodan et al. 1975), and rabbits (Prodan et al. 1975). At these concentrations, deaths occurred within 30–60 minutes. Male mice exposed to 30,000 ppm vinyl chloride 6 hours/day for 5 days, in a dominant lethal study showed an increased mortality rate (Anderson et al. 1976). An increased mortality rate was also observed at much lower concentrations in maternal mice in a developmental toxicity study (John et al. 1977, 1981). In this study, maternal mice had an increased incidence of deaths following exposure to 500 ppm for 10 days during gestation.

Decreased longevity was observed in intermediate-duration studies (Adkins et al. 1986; Drew et al. 1983; Feron et al. 1979a; Hong et al. 1981; Lee et al. 1978) and chronic-duration studies (Drew et al. 1983;

Feron et al. 1979a; Viola 1970). A treatment-related increase in the mortality rate was observed in mice exposed to 500 ppm of vinyl chloride for 6 hours/day, 5 days/week, for 6 months (Adkins et al. 1986). In mice and rats maintained for 12 months following a 6-month, 6 hour/day, 5 day/week exposure regime, decreased longevity was observed at concentrations as low as 50 ppm; however, statistical analyses of the data were not available to verify the significance of the decrease (Hong et al. 1981). Substantial increases in the mortality rate of mice and rats exposed to 250 ppm vinyl chloride for 12 months were observed by Lee et al. (1977a, 1978). In addition, small increases in mortality of mice and rats during the 12-month exposure period were observed at 50 ppm in these reports; however, the statistical significance of these increases was not reported.

The influence of the age on survival of female animals exposed to vinyl chloride was examined by Drew et al. (1983). In female hamsters exposed to 200 ppm, two strains of female mice exposed to 50 ppm, and female rats exposed to 100 ppm for 12 months, a higher death rate was observed when 2-month-old animals were exposed than when 8- or 14-month-old animals were exposed. Similar trends were observed when hamsters and mice were exposed to these concentrations for 6 months. The treatment-related deaths in this study may be due to the induction of vinyl chloride-induced carcinogenesis. These results demonstrate the importance of the latency period for cancer and associated mortality. Animals that were exposed at a younger age had a longer post-exposure period for the development of tumors. It is difficult to assess the sensitivity of younger animals to cancer mortality in this study because the same exposure concentrations were used for each age group. These results do not necessarily indicate that young people are more susceptible to the lethal effects of vinyl chloride, since animals that were exposed later in life may have died of age-related causes prior to the expression of the lethal effects. This study was limited in that only one dose of vinyl chloride was tested in each species.

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

3.2.1.2 Systemic Effects

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

Table 3-1 Levels of Significant Exposure to Vinyl Chloride - Inhalation

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (ppm)	LOAEL Less Serious (ppm)	LOAEL Serious (ppm)	Reference Chemical Form
ACUTE EXPOSURE							
Death							
1	Rat (NS)	30 min				300000 (5/5 died)	Mastromatteo et al. 1960
2	Rat (NS)	1x 2 hr				146625 (7/30 died)	Prodan et al. 1975
3	Mouse (CD-1)	5 d 6 hr/d				30000 M (11/20 died)	Anderson et al. 1976
4	Mouse (CF-1)	10 d 7 hr/d Gd6-15				500 F (5/29 died)	John et al. 1977, 1981
5	Mouse (NS)	30 min				200000 (1/5 died)	Mastromatteo et al. 1960
6	Mouse (NS)	1x 2 hr				107525 (15/61 died)	Prodan et al. 1975
7	Gn Pig (NS)	30 min				300000 (1/5 died)	Mastromatteo et al. 1960
8	Gn Pig (NS)	up to 8 hr				100000 (death)	Patty et al. 1930
9	Gn Pig (NS)	1x 2 hr				224825 (1/6 died)	Prodan et al. 1975

Table 3-1 Levels of Significant Exposure to Vinyl Chloride - Inhalation (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (ppm)	LOAEL Less Serious (ppm)	LOAEL Serious (ppm)	Reference Chemical Form
10	Rabbit (NS)	1x 2 hr				224825 (1/4 died)	Prodan et al. 1975
Systemic							
11	Rat (Holtzman)	1, 5 d 6 hr/d	Hepatic	50000 M	100000 M (hepatocellular vacuolization, increased AKT and SDH)		Jaeger et al. 1974
12	Rat (NS)	30 min	Resp		100000 (lung hyperemia)		Mastromatteo et al. 1960
			Hepatic	100000	200000 (fatty infiltration changes)		
			Renal	200000	300000 (renal congestion)		
13	Rat (Holtzman)	1, 5 d 6 hr/d	Hepatic	50000 M			Reynolds et al. 1975a
14	Rat (NS)	1 d 6 hr/d	Hepatic	50000 M			Reynolds et al. 1975b
15	Rat (Sprague-Dawley)	4 hr/d Gd 6-19	Bd Wt	1100 F			Thornton et al. 2002
16	Mouse (NS)	30 min	Resp		100000 (lung hyperemia)		Mastromatteo et al. 1960
			Hepatic	200000	300000 (liver congestion)		
			Renal		100000 (degenerative tubular epithelium)		

Table 3-1 Levels of Significant Exposure to Vinyl Chloride - Inhalation (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (ppm)	LOAEL Less Serious (ppm)		LOAEL Serious (ppm)		Reference Chemical Form
17	Gn Pig (NS)	30 min	Resp		100000	(slight pulmonary hyperemia)			Mastromatteo et al. 1960
			Cardio	400000					
			Hepatic	200000	300000	(fatty degeneration)			
			Endocr	400000					
			Ocular	400000					
Immuno/ Lymphoret									
18	Gn Pig (NS)	30 min		400000					Mastromatteo et al. 1960
Neurological									
19	Human	3 d 2 x/d 5 min		4000	8000	(dizziness)			Lester et al. 1963
20	Rat (Fischer- 344)	1 hr		50000					Hehir et al. 1981
21	Rat (Fischer- 344)	2 wk 5 d/wk 1 hr/d		500					Hehir et al. 1981
22	Rat (Holtzman)	1, 5 d 6 hr/d		50000 M			100000 M	(anesthesia)	Jaeger et al. 1974
23	Rat (Sherman)	2 hr			50000	(moderate intoxication)			Lester et al. 1963
24	Rat (NS)	30 min					100000	(narcosis)	Mastromatteo et al. 1960

Table 3-1 Levels of Significant Exposure to Vinyl Chloride - Inhalation (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (ppm)	LOAEL Less Serious (ppm)	LOAEL Serious (ppm)		Reference Chemical Form
25	Mouse (ICR)	1 hr		5000		50000	(ataxia)	Hehir et al. 1981
26	Mouse (NS)	30 min				100000	(narcosis)	Mastromatteo et al. 1960
27	Gn Pig (NS)	30 min				100000	(tremor, loss of consciousness)	Mastromatteo et al. 1960
28	Gn Pig (NS)	up to 8 hr		10000		25000	(narcosis)	Patty et al. 1930
Reproductive								
29	Rat (Sprague-Dawley)	16 wk (M) 19 wk (F) 2 gen 4 hr/d		1100				Thornton et al. 2002
30	Mouse (CD-1)	5 d 6 hr/d		30000 M				Anderson et al. 1976
Developmental								
31	Rat (Sprague-Dawley)	10 d 7 hr/d Gd6-15			2500 F	(ureter dilation)		John et al. 1977, 1981
32	Rat (Sprague-Dawley)	4 hr/d Gd 6-19		1100				Thornton et al. 2002

Table 3-1 Levels of Significant Exposure to Vinyl Chloride - Inhalation (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (ppm)	LOAEL Less Serious (ppm)	LOAEL Serious (ppm)	Reference Chemical Form
33	Mouse (CF-1)	10 d 7 hr/d Gd6-15		50[b] F	500 F (delayed ossification)		John et al. 1977, 1981
34	Rabbit (New Zealand)	13 d 7 hr/d Gd6-18			500 F (delayed ossification)		John et al. 1977, 1981
Cancer							
35	Mouse (ICR)	1 hr				5000 (CEL: bronchioalveolar adenoma)	Hehir et al. 1981
INTERMEDIATE EXPOSURE							
Death							
36	Rat (CD)	1-10 mo 5 d/wk 6 hr/d				50 (17/26 died)	Hong et al. 1981
37	Mouse (A/J)	6 mo 5 d/wk 6 hr/d				500 M (37/70 died) 500 F (23/70 died)	Adkins et al. 1986
38	Mouse (CD-1)	1-6 mo 5 d/wk 6 hr/d				50 (15/16 died)	Hong et al. 1981
Systemic							
39	Rat (Wistar)	3 mo 6 d/wk 6 hr/d	Cardio	10 M	100 M (increased relative heart weight)		Bi et al. 1985
			Renal	100 M	3000 M (increased relative kidney weight)		

Table 3-1 Levels of Significant Exposure to Vinyl Chloride - Inhalation (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (ppm)	LOAEL Less Serious (ppm)	LOAEL Serious (ppm)	Reference Chemical Form
40	Rat (Wistar)	6 mo 6 d/wk 6 hr/d	Cardio		10 M (increased relative heart weight)		Bi et al. 1985
			Hepatic		10 M (increased relative liver weight)		
41	Rat (Sherman)	19 d 8 hr/d	Hemato		50000 (decreased white blood cells)		Lester et al. 1963
			Hepatic		50000 (hepatocellular hypertrophy, large irregular vacuoles, compression of sinusoids, elevated relative liver weight)		
			Renal	50000			
			Dermal	50000 F	50000 M (thin coats, scaly tails)		
42	Rat (Sherman)	92 d 5 d/wk 8 hr/d	Hemato		20000 (decreased white blood cells)		Lester et al. 1963
			Hepatic		20000 (moderate hepatocellular hypertrophy, fine to medium vacuoles, compression of sinusoids)		
			Renal	20000			

Table 3-1 Levels of Significant Exposure to Vinyl Chloride - Inhalation (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (ppm)	LOAEL Less Serious (ppm)	LOAEL Serious (ppm)	Reference Chemical Form
43	Rat (Wistar)	10 mo 5 d/wk 5 hr/d	Musc/skel	20000 M			Sokal et al. 1980
			Hepatic		50 M (fatty changes)		
			Renal	50 M	500 M (increased kidney weight)		
			Bd Wt		50 M (10% decrease in body weight)		
44	Rat (Sprague-Dawley)	16 wk (M) 19 wk (F) 2 gen 4 hr/d	Hepatic		10[c] F (centrilobular hypertrophy in F1 female rats)		Thornton et al. 2002
			Bd Wt	1100			
45	Rat (NS)	6 mo 5 d/wk 0.5-7 hr/d	Hemato	200			Torkelson et al. 1961
			Hepatic		100 (increased relative liver weight)		
			Renal	200			
			Bd Wt	200			
46	Rat (Wistar)	10 mo 5 d/wk 5 hr/d	Hepatic		50 M (fatty changes)		Wisniewska- Knypl et al. 1980

Table 3-1 Levels of Significant Exposure to Vinyl Chloride - Inhalation (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (ppm)	LOAEL Less Serious (ppm)	LOAEL Serious (ppm)	Reference Chemical Form
47	Mouse (NS)	1-6 mo 5 d/wk 5 hr/d	Hepatic		2500 M (hyperplasia of hepatocytes and activated sinusoidal cells)		Schaffner 1978
48	Mouse (CD-1)	8 wk 5 d/wk 6 hr/d	Hemato	1000 M			Sharma and Gehring 1979
			Hepatic		1000 M (decreased liver weight)		
			Renal	1000 M			
			Bd Wt	1000 M			
49	Mouse (CD-1)	5-6 mo 5 d/wk 5 hr/d	Resp		2500 M (proliferation and hypertrophy of bronchial epithelium; hypersecretion of mucin; hyperplasia of alveolar epithelium)		Suzuki 1978, 1981
50	Rabbit (NS)	6 mo 5 d/wk 7 hr/d	Hepatic	100		200 (centrilobular degeneration and necrosis)	Torkelson et al. 1961
			Renal	200			
			Bd Wt	200			

Table 3-1 Levels of Significant Exposure to Vinyl Chloride - Inhalation (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (ppm)	LOAEL Less Serious (ppm)	LOAEL Serious (ppm)	Reference Chemical Form
Immuno/ Lymphoret							
51	Rat (Wistar)	3 mo 6 d/wk 6 hr/d		100 M	3000 M (increased spleen weight)		Bi et al. 1985
52	Rat (Wistar)	6 mo 6 d/wk 6 hr/d			10 M (increased spleen weight)		Bi et al. 1985
53	Rat (Wistar)	10 mo 5 d/wk 5 hr/d			50 M (increased spleen weight)		Sokal et al. 1980
54	Mouse (CD-1)	2-8 wk 5 d/wk 6 hr/d			10 M (increased spontaneous lymphocyte proliferation)		Sharma and Gehring 1979
Neurological							
55	Rat (Fischer- 344)	20 wk 5 d/wk 1 hr/d		50			Hehir et al. 1981
Reproductive							
56	Rat (Wistar)	3, 6 mo 6 d/wk 6 hr/d		10 M	100 M (decreased testes weight)		Bi et al. 1985
57	Rat (CD)	11 wk 5 d/wk 6 hr/d		50 M		250 M (reduced male fertility)	Short et al. 1977

Table 3-1 Levels of Significant Exposure to Vinyl Chloride - Inhalation (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (ppm)	LOAEL Less Serious (ppm)	LOAEL Serious (ppm)	Reference Chemical Form
58	Rat (Wistar)	10 mo 5 d/wk 5 hr/d		50 M		500 M (spermatogenic epithelial necrosis)	Sokal et al. 1980
Cancer							
59	Rat (Fischer- 344)	6 mo 5 d/wk 6 hr/d				100 F (CEL: hepatic hemangiosarcoma, hepatocellular carcinoma, neoplastic nodules; mammary fibroadenoma)	Drew et al. 1983
60	Rat (Sprague-Dawley)	33 d 6 d/wk 8 hr/d				500 M (CEL: hepatocellular carcinoma, angiosarcoma of the liver, benign cholangioma, nephroblastoma, angiomyoma, leukemia, Zymbal gland carcinoma, pituitary adenoma, mammary carcinoma and fibroma.	Froment et al. 1994
61	Rat (CD)	6 or 10 mo 5 d/wk 6 hr/d				250 (CEL: liver hemangiosarcoma, neoplastic nodules)	Hong et al. 1981

Table 3-1 Levels of Significant Exposure to Vinyl Chloride - Inhalation (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (ppm)	LOAEL Less Serious (ppm)	LOAEL Serious (ppm)	Reference Chemical Form
62	Rat (Sprague-Dawley)	16 wk (M) 19 wk (F) 2 gen 4 hr/d				1100 F (CEL: foci of hepatocellular alterations considered to be pre-neoplastic)	Thornton et al. 2002
63	Mouse (A/J)	6 mo 5 d/wk 6 hr/d				50 (CEL: pulmonary adenoma)	Adkins et al. 1986
64	Mouse (CD-1)	6 mo 5 d/wk 6 hr/d				50 F (CEL: hemangiosarcoma of skin, peritoneum; mammary gland carcinoma; lung carcinoma)	Drew et al. 1983
65	Mouse (B6C3F1)	6 mo 5 d/wk 6 hr/d				50 F (CEL: hemangiosarcoma of subcutis, peritoneum; mammary gland carcinoma)	Drew et al. 1983
66	Mouse (CD-1)	1, 3, 6 mo 5 d/wk 6 hr/d				50 F (CEL: mammary gland adenocarcinoma/carcinon	Hong et al. 1981
67	Mouse (Swiss)	30 wk 5 d/wk 4 hr/d				50 (CEL: liver angiosarcoma and angioma)	Maltoni et al. 1981

Table 3-1 Levels of Significant Exposure to Vinyl Chloride - Inhalation (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (ppm)	LOAEL Less Serious (ppm)	LOAEL Serious (ppm)	Reference Chemical Form
68	Mouse (CD-1)	4 wk 5 d/wk 6 hr/d				100 M (CEL: lung alveogenic tumors)	Suzuki 1983
69	Hamster (Golden Syrian)	6 mo 5 d/wk 6 hr/d				200 F (CEL: liver hemangiosarcoma; skin hemangiosarcoma, spleen hemangiosarcoma; mammary gland carcinoma)	Drew et al. 1983
70	Hamster (Golden Syrian)	30 wk 5 d/wk 4 hr/d				500 M (CEL: liver angiosarcoma)	Maltoni et al. 1981
CHRONIC EXPOSURE							
Systemic							
71	Rat (Wistar)	12 mo 6 d/wk 6 hr/d	Hepatic	100 M	3000 M (increased liver weight)		Bi et al. 1985
			Renal	10 M	100 M (increased kidney weight)		
			Bd Wt	10 M	100 M (14% decrease in body weight)		

Table 3-1 Levels of Significant Exposure to Vinyl Chloride - Inhalation (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (ppm)	LOAEL Less Serious (ppm)	LOAEL Serious (ppm)	Reference Chemical Form
Reproductive							
72	Rat (Wistar)	12 mo 6 d/wk 6 hr/d		10 M	100 M (degenerative seminiferous tubule changes)		Bi et al. 1985
Cancer							
73	Rat (Wistar)	12 mo 6 d/wk 6 hr/d				100 M (CEL: liver angiosarcoma; lung angiosarcoma)	Bi et al. 1985
74	Rat (Fischer- 344)	12, 18, 24 mo 5 d/wk 6 hr/d				100 F (CEL: hepatic hemangiosarcoma, hepatocellular carcinoma, neoplastic nodules; mammary gland fibroadenoma and adenocarcinoma)	Drew et al. 1983
75	Rat (albino)	6 hr/d 5 d/wk 26 or 52 wk				50 (CEL: lung, kidney, abdominal hemangiosarcoma)	Holmberg et al. 1976
76	Rat (CD)	1-12 mo 5 d/wk 6 hr/d				250 F (CEL: hepatic hemangiosarcoma)	Lee et al. 1978

Table 3-1 Levels of Significant Exposure to Vinyl Chloride - Inhalation (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (ppm)	LOAEL Less Serious (ppm)	LOAEL Serious (ppm)	Reference Chemical Form
77	Rat (Sprague-Dawley)	52 wk 5 d/wk 4 hr/d				5 F (CEL: mammary gland carcinoma)	Maltoni et al. 1981
78	Mouse (Swiss CD-1)	12, 18 mo 5 d/wk 6 hr/d				50 F (CEL: lung; hemangiosarcoma of peritoneum, subcutis; mammary gland carcinoma)	Drew et al. 1983
79	Mouse (B6C3F1)	12 mo 5 d/wk 6 hr/d				50 F (CEL: hemangiosarcoma of peritoneum, subcutis; mammary gland carcinoma)	Drew et al. 1983
80	Mouse (CD-1)	1-12 mo 5 d/wk 6 hr/d				50 F (CEL: mammary gland adenoma and adenocarcinoma)	Lee et al. 1977a, 1978
						50 (CEL: hepatic hemangiosarcoma; bronchiolo-alveolar adenoma; malignant lymphoma)	

Table 3-1 Levels of Significant Exposure to Vinyl Chloride - Inhalation (continued)

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (ppm)	LOAEL			Reference Chemical Form
					Less Serious (ppm)	Serious (ppm)		
81	Hamster (Golden Syrian)	12, 18, 24 mo 5 d/wk 6 hr/d				200 F	(CEL: liver hemangiosarcoma; skin carcinoma, hemangiosarcoma; spleen hemangiosarcoma; mammary gland carcinoma; stomach adenoma)	Drew et al. 1983

a Numbers correspond to entries in Figure 3-1.

b Used to derive an acute-duration inhalation Minimal Risk Level (MRL) of 0.5 ppm. A NOAEL was adjusted for intermittent exposure and converted to a Human Equivalent Concentration (HEC) before applying uncertainty factors. The MRL was obtained by dividing the NOAEL-HEC by an uncertainty factor of 30 (3 for extrapolation from animals to humans using a dosimetric adjustment, and 10 for human variability).

c Used to derive an intermediate-duration inhalation MRL of 0.03 ppm. LEC10 converted to an HEC and adjusted for intermittent exposure before applying uncertainty factors. The MRL was obtained by dividing the LEC10-HEC by an uncertainty factor of 30 (3 for extrapolation from animals to humans using a dosimetric adjustment, and 10 for human variability).

AKT = alpha-ketoglutarate transaminase; B - both; Bd Wt = body weight; Cardio = cardiovascular; CEL = cancer effect level; d = day(s); derm = dermal; Endocr = endocrine; F = Female; Gd = gestational day; Gn pig = guinea pig; hemato = hematological; hr = hour(s); Immuno = immunological; LOAEL = lowest-observed-adverse-effect level; M = male; min = minute(s); mo = month(s); Musc/skel = musculoskeletal; NOAEL = no-observed-adverse-effect level; NS = not specified; Resp = respiratory; SDH = sorbitol dehydrogenase; wk = week(s); x = time(s)

Figure 3-1 Levels of Significant Exposure to Vinyl Chloride - Inhalation

Acute (≤14 days)

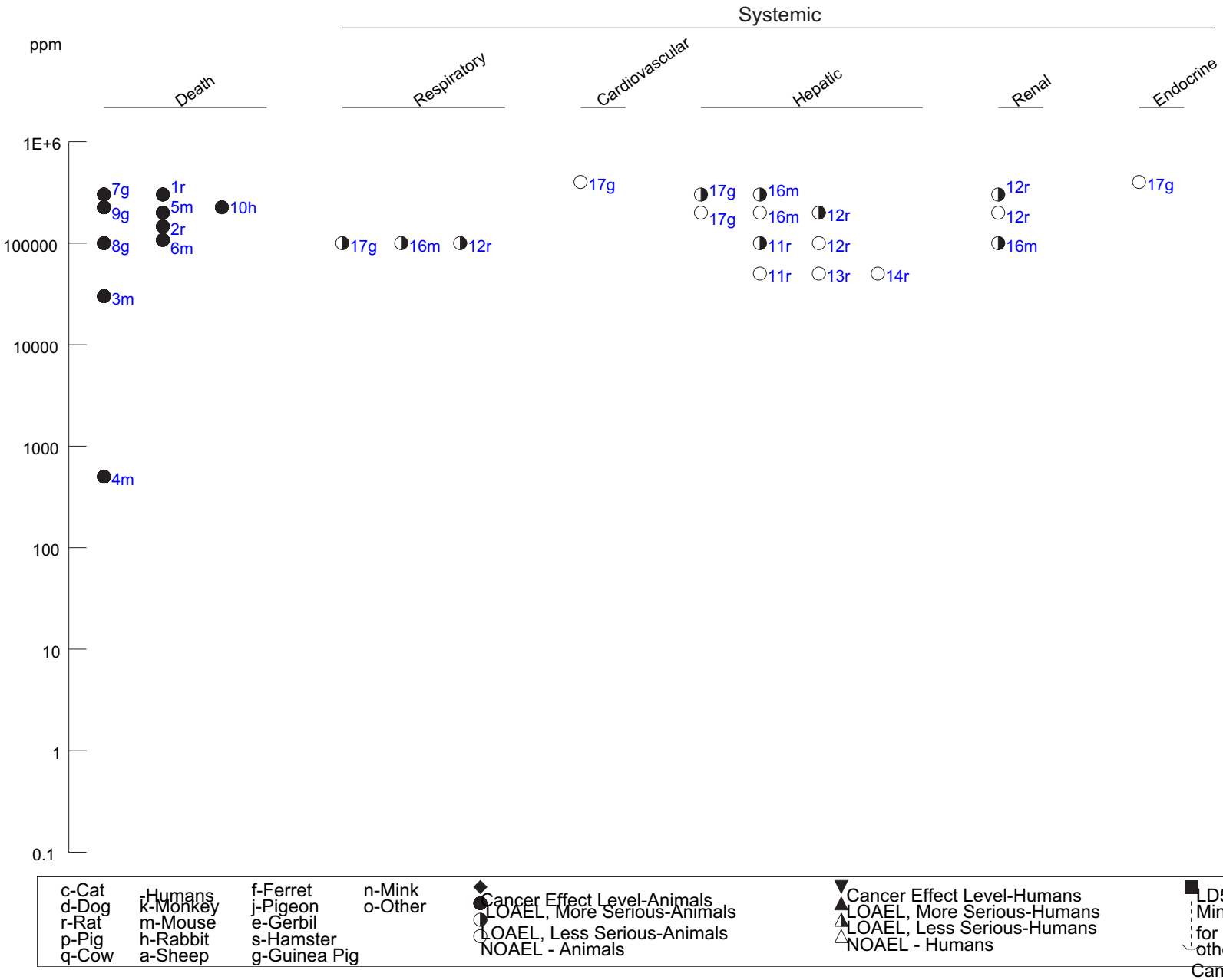


Figure 3-1 Levels of Significant Exposure to Vinyl Chloride - Inhalation *(continued)*

Acute (≤14 days)

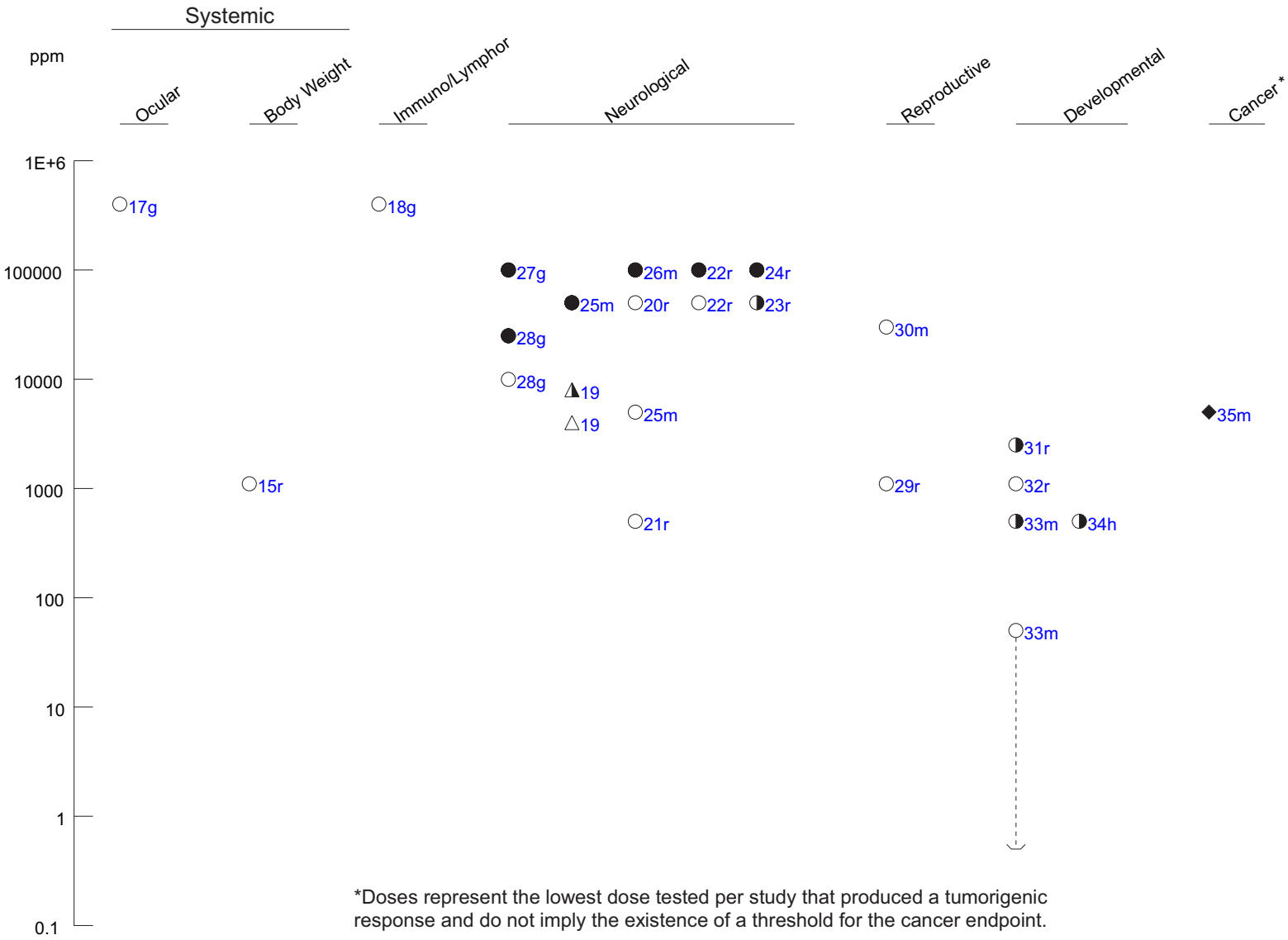


Figure 3-1 Levels of Significant Exposure to Vinyl Chloride - Inhalation *(continued)*

Intermediate (15-364 days)

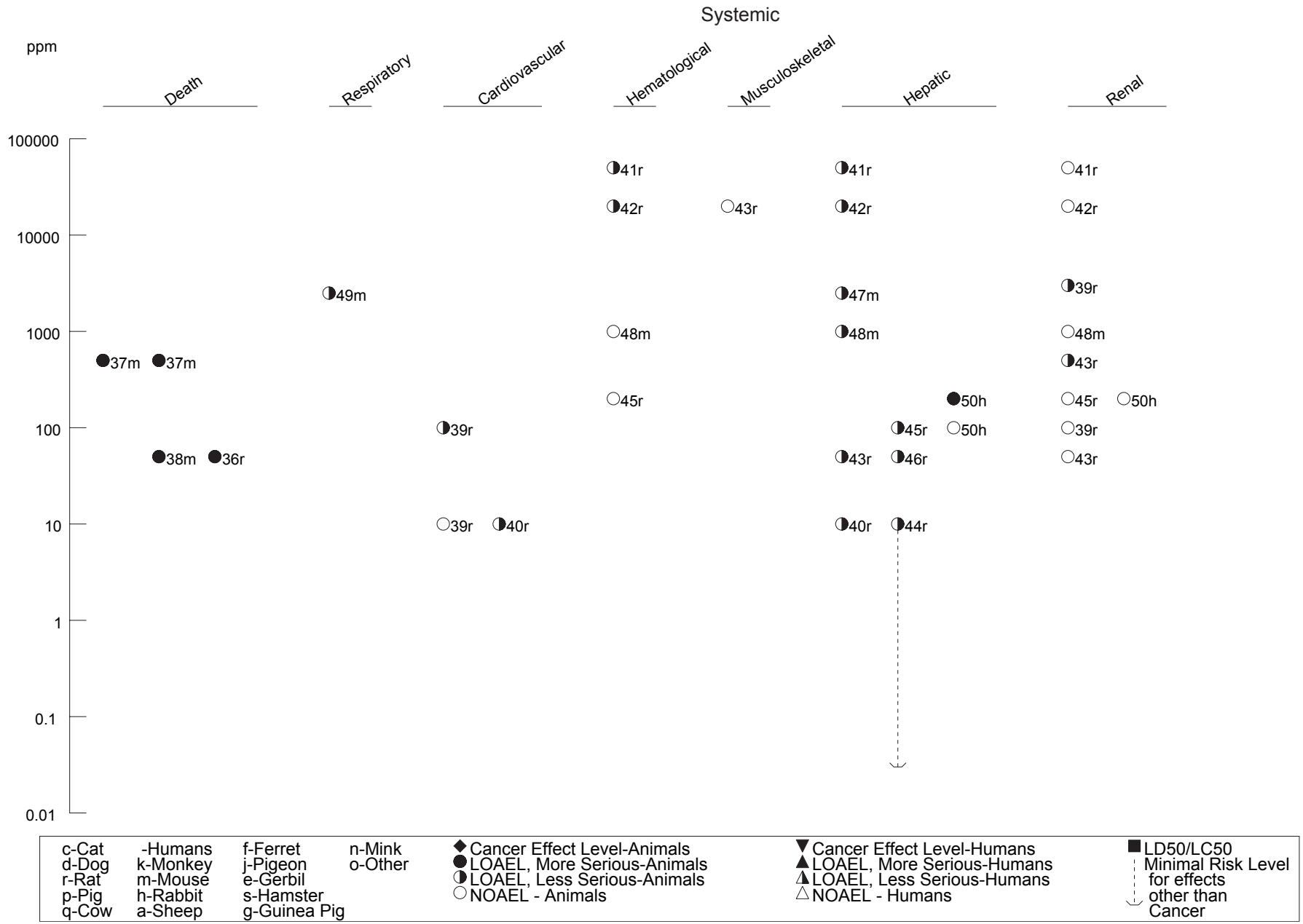
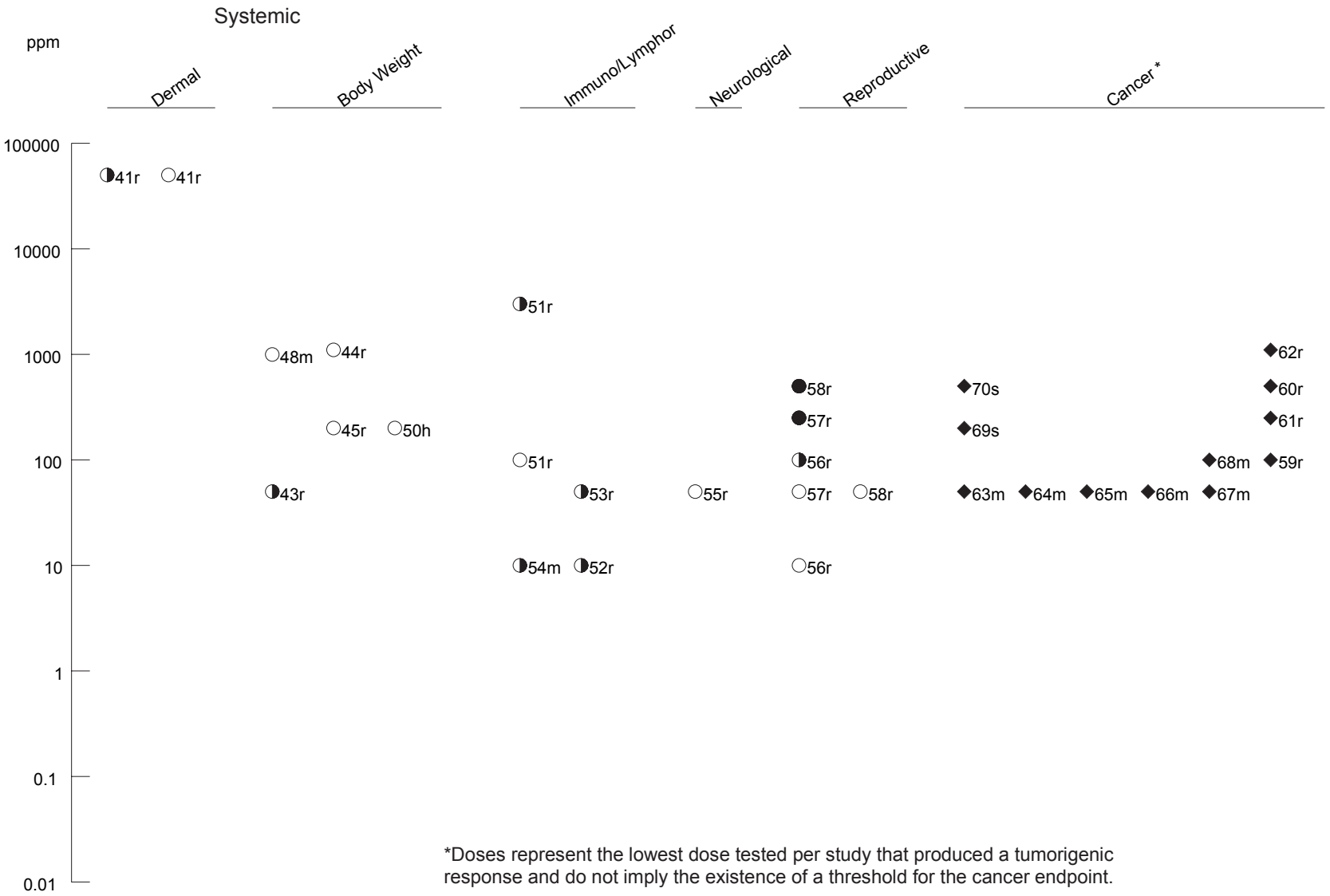


Figure 3-1 Levels of Significant Exposure to Vinyl Chloride - Inhalation *(continued)*

Intermediate (15-364 days)



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

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

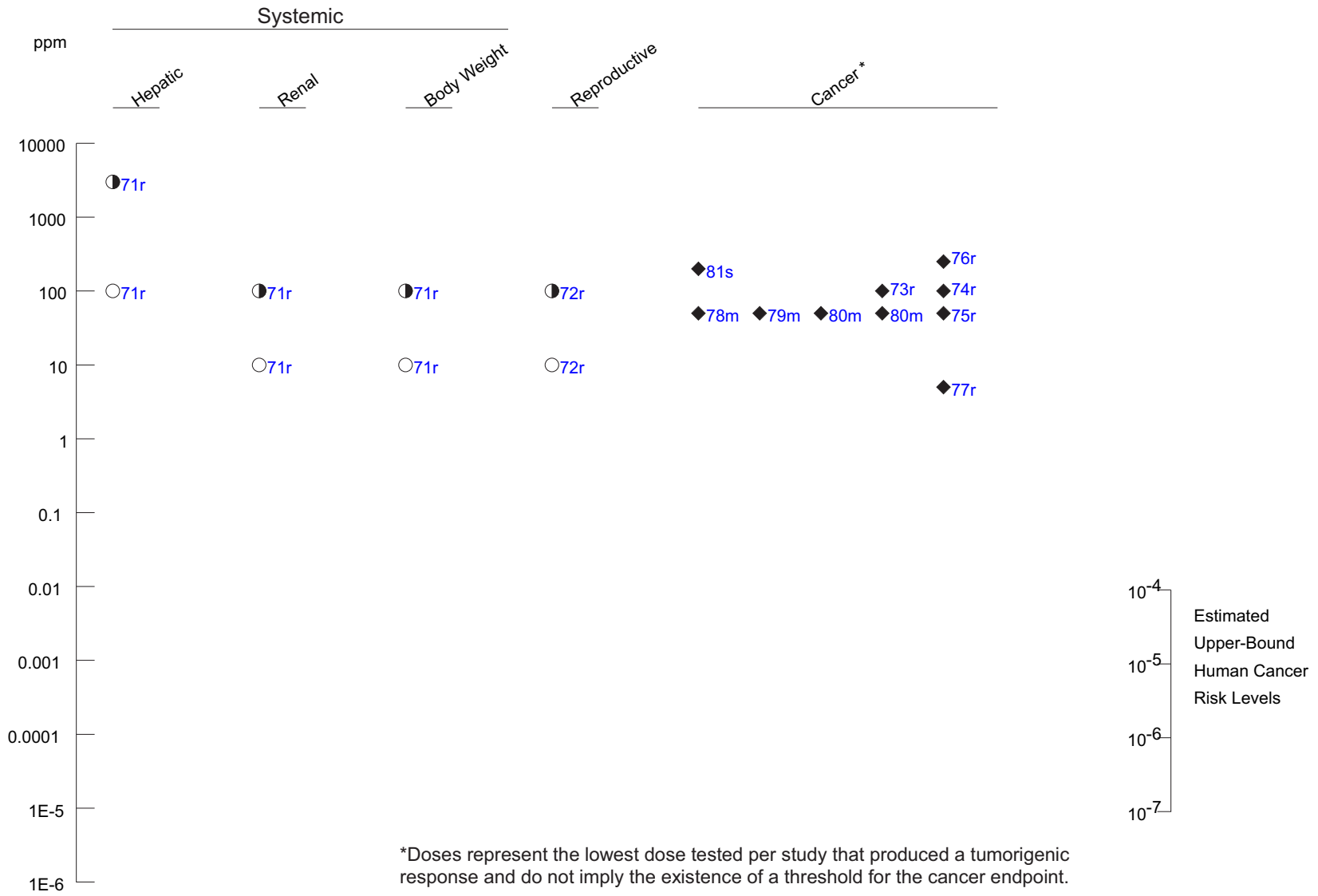
◆ Cancer Effect Level-Animals
● LOAEL, More Serious-Animals
◑ LOAEL, Less Serious-Animals
○ NOAEL - Animals

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

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

Figure 3-1 Levels of Significant Exposure to Vinyl Chloride - Inhalation *(continued)*

Chronic (≥365 days)



Respiratory Effects. Limited information is available on the acute adverse effects from inhalation of vinyl chloride by humans. Autopsy findings from a man who died after being overcome by vinyl chloride revealed the irritating nature of a high-level inhalation exposure. The lungs were found to be intensely hyperemic, and some desquamation of the alveolar epithelium had occurred (Danziger 1960). Reports regarding respiratory effects in workers who are occupationally exposed to vinyl chloride are contradictory. Several epidemiologic studies found no increased incidence of respiratory disease among vinyl chloride workers (Gamble et al. 1976; Laplanche et al. 1987, 1992; NIOSH 1977). However, adverse respiratory effects were reported in several epidemiologic surveys and case reports, with these effects including increased incidence of emphysema (Suciu et al. 1975; Wong et al. 1991), decreased respiratory volume and vital capacity, respiratory insufficiency (Suciu et al. 1975), decreased respiratory oxygen and carbon dioxide transfer (Lloyd et al. 1984), pulmonary fibrosis of the linear type (Suciu et al. 1975), abnormal chest x-rays (Lilis et al. 1975, 1976), and dyspnea (Walker 1976). Interpretation of many of these results is confounded by the inclusion of smokers among those exposed to vinyl chloride and the concurrent exposure of many vinyl chloride workers to PVC resin dust, which is known to produce respiratory lesions (Mastrangelo et al. 1979).

Brief inhalation of high concentrations of vinyl chloride produced respiratory inflammation in a variety of animals. A 30-minute exposure of guinea pigs, mice, and rats to 100,000 ppm of vinyl chloride produced hyperemia in all three species (Mastromatteo et al. 1960). Exposure to higher concentrations (200,000 ppm and 300,000 ppm) produced increased congestion, edema, and at the highest concentrations, pulmonary hemorrhages in all three species (Mastromatteo et al. 1960). Tracheal epithelium was also absent in one guinea pig exposed to 400,000 ppm for 30 minutes (Mastromatteo et al. 1960). Edema and congestion of the lungs of rats were also observed following a 2-hour exposure to 150,000 ppm (Lester et al. 1963).

Histopathologic examination of mice exposed to 2,500 ppm vinyl chloride 5 hours/day, 5 days/week for 5–6 months revealed proliferation and hypertrophy of the bronchiolar epithelium, hyperplasia of the alveolar epithelium, hypersecretion of mucin (Suzuki 1978, 1980, 1981), increased endoplasmic reticulum and free ribosomes in Clara cells, and mobilization of alveolar macrophages (Suzuki 1980). These changes were observed irrespective of the recovery period (2 or 37 days), indicating that they were not readily reversible. However, these studies were limited by the small number of animals tested and the absence of a statistical analysis.

Chronic exposure of rats to 5,000 ppm 7 hours/day, 5 days/week for 12 months produced hyperplasia of the olfactory epithelium, increased cellularity of the interalveolar septa of the lungs, and an increased incidence of pulmonary hemorrhage (Feron and Kroes 1979). Interstitial pneumonia and hemorrhagic lungs were observed in rats exposed to 30,000 ppm of vinyl chloride 4 hours/day, 5 days/week for 12 months (Viola et al. 1971). However, neither of these studies reported the statistical significance of these findings.

Cardiovascular Effects. Occupational exposure to vinyl chloride has been associated with the development of Raynaud's phenomenon, a condition in which the fingers blanch and become numb with discomfort upon exposure to the cold. It has also been reported in a worker exposed once to a vinyl chloride leak (Ostlere et al. 1992). Although only a small percentage of vinyl chloride workers develop Raynaud's phenomenon (Laplanche et al. 1987, 1992; Lilis et al. 1975; Marsteller et al. 1975; Suciu et al. 1963, 1975; Veltman et al. 1975; Walker 1976), the incidence is significantly higher than in unexposed workers (Laplanche et al. 1987, 1992). Investigation of the peripheral circulation of workers afflicted with Raynaud's phenomenon has revealed thickening of the walls of the digital arteries (Harris and Adams 1967), narrowing of the arterial lumen (Veltman et al. 1975), vascular occlusions (Walker 1976), arterial occlusions (Preston et al. 1976; Veltman et al. 1975), tortuosity (Preston et al. 1976), hypervascularity (Preston et al. 1976), inflammatory infiltration of the arterioles (Magnavita et al. 1986), deposition of immune products along the vascular endothelium (Ward 1976), vasomotor impairment (Suciu et al. 1963), and impaired capillary microcirculation (Magnavita et al. 1986; Maricq et al. 1976). Three reports indicate that upon removal from exposure, Raynaud's phenomenon gradually disappears (Freudiger et al. 1988; Suciu et al. 1963, 1975). For further discussion of Raynaud's phenomenon, see Immunological/Lymphoreticular Effects (Section 3.2.1.3).

Splenomegaly, with evidence of portal hypertension (dilated peritoneal veins and esophageal varices), has been reported by investigators studying the effects of vinyl chloride exposure (Marsteller et al. 1975). In addition, hypertension among vinyl chloride workers (NIOSH 1977; Suciu et al. 1975) and significantly increased mortality rate due to cardiovascular and cerebrovascular disease (Byren et al. 1976) have been reported. An association between vinyl chloride exposure and arterial hypertension was observed in an occupational worker study. Conclusive evidence was not provided for an association of vinyl chloride with coronary heart disease (Kotseva 1996).

Investigators studying the anesthetic properties of vinyl chloride in dogs have observed that doses producing anesthesia (100,000 ppm, Oster et al. 1947; 150,000–900,000 ppm, Carr et al. 1949) also

produced cardiac arrhythmias. Arrhythmias were characterized by intermittent tachycardia, extraventricular systoles, vagal beats, ventricular fibrillation, and atrioventricular block. However, the statistical significance of these effects was not reported. At high concentrations (>30,000 ppm), vinyl chloride was been shown to sensitize the heart to epinephrine, resulting in cardiac arrhythmias in dogs (Clark and Tinston 1973). No histopathological changes in the heart were noted in guinea pigs exposed to 400,000 ppm of vinyl chloride for 30 minutes (Mastromatteo et al. 1960).

A study by Bi et al. (1985) demonstrated an increase in the relative heart weight at concentrations of vinyl chloride as low as 10 ppm when administered to male rats 6 hours/day, 6 days/week for 6 months. Heart weight was also increased after 3 months in rats exposed to 100 ppm under this regimen (Bi et al. 1985). Chronic exposure of rats to 5,000 ppm vinyl chloride 7 hours/day, 5 days/week for 1 year resulted in increases in areas of myodegeneration in the heart and thickening of the walls of arteries (Feron and Kroes 1979). However, the statistical significance of this effect was not reported. Exposure of rats to 30,000 ppm of vinyl chloride 4 hours/day, 5 days/week for 1 year also produced thickening of the walls of small arterial vessels. The thickening was characterized by a proliferation of the endothelium. In some vessels, the thickening was severe enough to cause blockage of the lumen (Viola 1970).

Gastrointestinal Effects. Approximately 32% of the vinyl chloride workers examined by Lilis et al. (1975) reported a history of "gastritis, ulcers (gastric and duodenal), and upper gastrointestinal bleeding." Because these subjects were not compared to workers who had not been exposed to vinyl chloride, the significance of these findings is unknown. Other symptoms reported by vinyl chloride workers included nausea, abdominal distension, and heartburn. Loss of appetite and nausea have been reported in Singapore workers exposed to 1–21 ppm vinyl chloride (Ho et al. 1991). However, these workers were selected on the basis of liver dysfunction.

No studies were located regarding gastrointestinal effects in animals following inhalation exposure to vinyl chloride.

Hematological Effects. Blood tests performed at autopsy of two workers whose deaths were believed to be due to exposure to extremely high levels of vinyl chloride revealed that blood clotting did not occur (Danziger 1960). Slight-to-severe thrombocytopenia in workers occupationally exposed to vinyl chloride was reported in several studies (Marsteller et al. 1975; Micu et al. 1985; Veltman et al. 1975), but Lilis et al. (1975) found no increased incidence of thrombocytopenia in vinyl chloride workers. A prospective study of female workers exposed to vinyl chloride at levels ranging from 0.2 to 130.7 ppm

showed that the exposed workers had a significantly lower number of platelets than the nonexposed controls during the early part of their pregnancies (weeks 8–10) but that this effect abated by the end of the pregnancy (34–38 weeks) following a period free from exposure (Bao et al. 1988). Splenomegaly was reported in a number of studies (Ho et al. 1991; Marsteller et al. 1975; Popper and Thomas 1975; Suciu et al. 1963; Veltman et al. 1975). Thrombocytopenia was found in patients who both did and did not present with splenomegaly (Veltman et al. 1975). Increased levels of two plasma proteins (α_1- and α_2-globulin) were reported in studies examining the effects of occupational exposure to vinyl chloride (Harris and Adams 1967; Suciu et al. 1975).

A brief (30-minute) exposure of guinea pigs to 400,000 ppm vinyl chloride resulted in a failure of the blood to clot in the animals that died during the exposure (Mastromatteo et al. 1960). Mice that were exposed to 5,000 ppm (4 hours/day for 6 days) or 10,000 ppm (4 hours/day for 5 days) showed an increased emergence of basophilic stippled erythrocytes (Kudo et al. 1990). This effect was also noted in mice that were exposed for 10 weeks to 50 ppm intermittently (4 hours/day for 4–5 days/week,) or to 30–40 ppm continuously for 62 days (Kudo et al. 1990). Exposure of dogs and rats to 200 ppm for 7 hours/day, 5 days/week, for 6 months had no effect on hematologic values (Torkelson et al. 1961). Also, an 8-week exposure of mice to 1,000 ppm for 6 hours/day, 5 days/week had no effect on erythrocyte or leukocyte counts (Sharma and Gehring 1979). Exposure of rats to either 50,000 ppm for 8 hours/day for 19 consecutive days or 20,000 ppm for 8 hours/day, 5 days/week for 92 days resulted in a decrease in white blood cells (Lester et al. 1963). Exposure of rats to 5,000 ppm vinyl chloride for 7 hours/day, 5 days/week for 1 year produced increased hematopoiesis in the spleen (Feron and Kroes 1979). The statistical significance of these results was not reported. Blood clotting time was decreased in rats exposed to 5,000 ppm for7 hours/day for 1 year, but the statistical significance of these effects was not reported (Feron et al. 1979a).

Musculoskeletal Effects. Acroosteolysis, or resorption of the terminal phalanges of the finger, was observed in a small percentage of workers occupationally exposed to vinyl chloride (Dinman et al. 1971; Lilis et al. 1975; Marsteller et al. 1975; Sakabe 1975; Veltman et al. 1975; Wilson et al. 1967). Bone lesions were most often confined to the terminal phalanges of the fingers, but in a few cases the bones of the toes (Harris and Adams 1967), feet (Preston et al. 1976), sacroiliac joint (Harris and Adams 1967), and arms, legs, pelvis, and mandible (Preston et al. 1976) were also involved. Development of acroosteolysis was most often preceded by Raynaud's phenomenon (Dinman et al. 1971; Freudiger et al. 1988; Harris and Adams 1967; Magnavita et al. 1986; Markowitz et al. 1972; Preston et al. 1976; Sakabe 1975; Veltman et al. 1975; Wilson et al. 1967). In two reports, bone resorption was observed to progress

despite discontinuation of exposure (Markowitz et al. 1972; Preston et al. 1976). However, in two other reports, improvement was observed after exposure ceased (Veltman et al. 1975; Wilson et al. 1967). Joint pain was also reported by Lilis et al. (1975).

Although Sokal et al. (1980) found no alterations in the bones of male rats exposed to 20,000 ppm for 5 hours/day, 5 days/week for 10 months, Viola (1970) observed skeletal changes (i.e., osteochondroma) in the bones of rats exposed to 30,000 ppm for 4 hours/day, 5 days/week for 12 months. The statistical significance of these effects was not reported and only one exposure level was tested.

Hepatic Effects. Throughout the early years of the use of vinyl chloride, only a minimal degree of functional hepatic abnormalities were detected in workers. However, when it became apparent in the early 1970s that angiosarcoma of the liver was associated with long-term vinyl chloride exposure, an intensive effort was initiated by a number of investigators to characterize the hepatic effects of vinyl chloride. These studies revealed characteristic hepatic lesions produced by vinyl chloride exposure (Berk et al. 1975; Falk et al. 1974; Gedigke et al. 1975; Ho et al. 1991; Jones and Smith 1982; Lilis et al. 1975; Liss et al. 1985; Marsteller et al. 1975; NIOSH 1977; Popper and Thomas 1975; Suciu et al. 1975; Tamburro et al. 1984; Vihko et al. 1984). The incidence and severity of the effects correlated well with the duration of exposure (Gedigke et al. 1975; Lilis et al. 1975; NIOSH 1977).

Routine, noninvasive techniques revealed hepatomegaly in a limited number of workers (14–37%) (Ho et al. 1991; Lilis et al. 1975; Marsteller et al. 1975; NIOSH 1977; Suciu et al. 1963, 1975). However, when peritoneoscopy was performed or biopsies were obtained from exposed workers, Marsteller et al. (1975) found a much higher prevalence of hepatic abnormalities. Only 37% of the workers studied by Marsteller et al. (1975) were diagnosed with hepatomegaly, but peritoneoscopy revealed a 50% incidence of granular changes in the liver surface and an 86% incidence of capsular fibrosis with increased numbers of capsular vessels. Histopathological examination of the biopsied tissue from these workers revealed an 80% incidence of collagenization of the sinusoidal walls, a 90% incidence of proliferation of cells lining the sinusoids, a 30% incidence of septal fibrosis, and degeneration of hepatocytes (incidence not specified). A number of other investigators observed similar changes in liver tissues obtained from workers exposed to vinyl chloride (Falk et al. 1974; Gedigke et al. 1975; Popper and Thomas 1975; Tamburro et al. 1984). Based on these observations, a profile of vinyl chloride-induced liver damage was compiled and includes the following features: hypertrophy and hyperplasia of hepatocytes, activation and hyperplasia of sinusoidal lining cells, fibrosis of the portal tracts and the septa and intralobular perisinusoidal regions, sinusoidal dilation, and focal areas of hepatocellular degeneration. This pattern of changes was observed

to be highly unusual and was similar to the hepatic changes produced by arsenic (Gedigke et al. 1975). In addition, the degenerative changes in hepatocytes appeared to be less severe when biopsy material was obtained from workers who had not been exposed to vinyl chloride recently. However, sinusoidal changes were not influenced by the length of time since the last exposure (Gedigke et al. 1975).

One possible reason that the hepatotoxic effects of vinyl chloride went undetected for many years was the lack of sensitivity of standard biochemical liver function tests to detect the liver injury produced by vinyl chloride (Berk et al. 1975; Marsteller et al. 1975; Tamburro et al. 1984; Vihko et al. 1984). For example, the values obtained in several standard biochemical liver function tests (activities of serum alkaline phosphatase, aspartate aminotransferase, alanine aminotransferase, gamma-glutamyltransferase) from workers with biopsy evidence of vinyl chloride-associated liver damage were not significantly higher than those from unexposed controls (Liss et al. 1985). Gamma-glutamyltransferase levels were significantly higher in workers exposed to vinyl chloride at TWA exposure concentrations of >10 ppm compared to workers exposed to lower exposure concentrations (Du et al. 1995). Workers exposed to lower levels of vinyl chloride had gamma-glutamyltransferase levels that were within the normal range (Hensyl 1990). Abnormal liver function (i.e., increased alkaline phosphatase, alanine aminotransferase, or gamma-glutamyltransferase) was demonstrated in workers exposed to high concentrations of vinyl chloride (1–20 ppm) (Ho et al. 1991; Lilis et al.1975) and workers who experienced a combined exposure to vinyl chloride and ethylene dichloride (Cheng et al. 1999). In the mixed exposure situation, altered liver function may be related to the effect of each component or the interactive effect of the mixture. Serum bile acids (Berk et al. 1975; Liss et al. 1985) and/or indocyanine green clearance (Liss et al. 1985; Tamburro et al. 1984) correlated with liver injury. Furthermore, investigators have shown that levels of chenodeoxycholic acid (a serum bile acid) in asymptomatic vinyl chloride workers were elevated when compared to the 95% interval of values from a healthy reference population (Vihko et al. 1984). The serum hyaluronic acid concentration was demonstrated to be elevated in workers with angiosarcoma of the liver, while other liver function tests were normal (McClain et al. 2002).

A recent IARC update of a multi-center cohort study demonstrated an increase in mortality from liver cirrhosis in workers exposed to moderate to high concentrations of vinyl chloride (Ward et al. 2001). Morbidity associated with liver cirrhosis was also reported to be elevated among vinyl chloride workers (Du and Wang 1998). Alcohol intake was not evaluated as a critical confounding factor in these studies. Mastrangelo et al (2004) evaluated the possible interaction between alcohol consumption, hepatitis infection, and liver cirrhosis in a large cohort of vinyl chloride workers. Vinyl chloride was suggested to be an independent risk factor for liver cirrhosis with a synergistic interaction described for alcohol

consumption and an additive interaction observed for hepatitis infection. Liver ultrasonography illustrated an increase in the incidence of periportal fibrosis in vinyl chloride workers (Maroni et al. 2003). Portal fibrosis and portal hypertension were considered to contribute to mortality in several cases (Lee et al. 1996; Lelbach 1996).

Brief exposure of animals to extremely high concentrations of vinyl chloride has been shown to produce hepatic damage. For example, acute exposure (30 minutes) of guinea pigs and mice to 300,000 ppm of vinyl chloride produced liver congestion or severe fatty degeneration while 200,000 ppm caused fatty infiltration in rats (Mastromatteo et al. 1960). Exposure to 100,000 ppm for 6 hours produced centrilobular vacuolization and increased alanine serum α-ketoglutarate transaminase activity in rats (Jaeger et al. 1974). However, exposure of rats to 50,000 ppm for 6 hours produced no observable effects on the liver (Reynolds et al. 1975a, 1975b). In contrast, a single-concentration study in which pregnant rats were continuously exposed to 1,500 ppm for 7–9 days during either the first or second trimester of pregnancy resulted in an increase in the liver-to-body-weight ratio (Ungvary et al. 1978). Interestingly, a single 1-hour exposure of mice to 500, 5,000, or 50,000 ppm of vinyl chloride, followed by an 18-month observation period, resulted in an increased incidence of hepatocellular hypertrophy in these animals at terminal sacrifice (Hehir et al. 1981). The hypertrophy was not dose dependent; thus, the significance of this effect is uncertain.

In studies with longer durations of exposure, lower concentrations of vinyl chloride have produced hepatic toxicity. Symptoms of hepatotoxicity that have been observed in rats have included hepatocellular degeneration (Sokal et al. 1980; Torkelson et al. 1961; Wisniewska-Knypl et al. 1980), swelling of hepatocytes with compression of sinusoids (Lester et al. 1963), dilation of the rough endoplasmic reticulum (Du et al. 1979), proliferation (Sokal et al. 1980) or hypertrophy (Thornton et al. 2002; Wisniewska-Knypl et al. 1980) of smooth endoplasmic reticulum, changes in metabolic enzyme activities (Du et al. 1979; Wisniewska-Knypl et al. 1980), proliferation of reticulocytes (Sokal et al. 1980), and an increased liver-to-body-weight ratio (Bi et al. 1985; Lester et al. 1963; Sokal et al. 1980; Thornton et al. 2002; Torkelson et al. 1961). For example, exposure of rats to 500 ppm for 7 hours/day, 5 days/week for 4.5 months resulted in an increase in liver-to-body-weight ratio and granular degeneration (Torkelson et al. 1961). An increased liver-to-body-weight ratio was also found in rats exposed to 100 ppm vinyl chloride for 7 hours/day, 5 days/week for 6 months (Torkelson et al. 1961). Relative liver weight was decreased in mice exposed to 1,000 ppm vinyl chloride for 6 hours/day, 5 days/week for 8 weeks (Sharma and Gehring 1979). The liver-to-body-weight ratio was shown to be increased in male rats exposed to 3,000 ppm, but not 100 ppm, vinyl chloride for 6 hours/day,

5 days/week for 12 months (Bi et al. 1985). Significantly increased liver-to-body-weight ratio was also observed in rats exposed to concentrations of vinyl chloride as low as 10 ppm for 6 hours/day, 6 days/week for 6 months (Bi et al. 1985). Exposure of rats to 500 ppm for 5 hours/day, 5 days/week for 10 months produced swelling of hepatocytes and proliferation of reticuloendothelial cells, increased liver weight, and cellular degeneration; at 50 ppm, small lipid droplets and proliferation of smooth endoplasmic reticulum were noted (Sokal et al. 1980). Histopathological examination of rats exposed to either 50,000 ppm vinyl chloride for 8 hours/day for 19 consecutive days or 20,000 ppm vinyl chloride for 8 hours/day, 5 days/week, for 92 days showed hepatocellular hypertrophy, vacuolization, and sinusoidal compression (Lester et al. 1963). Mice exposed to 2,500 ppm vinyl chloride 5 hours/day, 5 days/week for up to 6 months showed histopathological changes in the liver that included hyperplasia of hepatocytes and activated sinusoidal cells (Schaffner 1978). Centrilobular necrosis and degeneration were noted in rabbits exposed to 200 ppm vinyl chloride 7 hours/day, 5 days/week for 6 months but not at 100 ppm vinyl chloride in this regimen (Torkelson et al. 1961). Also, exposure of rats to 50 ppm for 5 hours/day, 5 days/week for 10 months produced fatty degeneration and proliferation of the smooth endoplasmic reticulum (Wisniewska-Knypl et al. 1980). Liver effects were observed in a 2-generation reproductive toxicity study where rats were exposed to 0, 10, 100, or 1,100 ppm vinyl chloride (6 hours/day for a 10-week premating period and a 3-week mating period) (Thornton et al. 2002). Absolute and relative mean liver weights were significantly increased at all exposure levels in F_0 males and in 100- and 1,100-ppm F_1 males. Centrilobular hypertrophy, considered to be a minimal adverse effect, was noted in the livers of all 1,100-ppm male and female F_0 and F_1 rats, most 100-ppm male and female F_0 and F_1 rats, and in 2/30 and 6/30 of the 10-ppm F_0 male and F_1 female rats, respectively. Centrilobular hypertrophy was not noted in the 30 female rats of the control group. Histopathological alterations occurring at 100 and 1,100 ppm included centrilobular hypertrophy and acidophilic, basophilic, and clear cell foci. Based on this study, an intermediate-duration MRL of 0.03 ppm was derived from a benchmark dose of 5 ppm as described in the footnote in Table 3-1.

The NOAELs for liver effects in a number of species following a 6-month exposure to vinyl chloride indicated that mice and rats were the most sensitive (NOAEL=50 ppm), rabbits were the next most sensitive (NOAEL=100 ppm), and dogs and guinea pigs were the least sensitive (NOAEL>200 ppm) (Torkelson et al. 1961).

Popper et al. (1981) compared histopathological findings from sections of liver from mice and rats exposed by Maltoni and LeFemine (1975) with the liver biopsy material obtained from vinyl chloride workers. Hyperplasia and hypertrophy of hepatocytes and/or sinusoidal cells, with areas of sinusoidal

dilation, were observed in both humans and rodents. The major difference between the species was the greater degree of fibrosis, seen as reticulin deposition and collagen formation, in the livers of humans. Also, inflammatory cells were present in the livers of humans but not rodents.

Renal Effects. No studies were located regarding renal effects in humans after inhalation exposure to vinyl chloride.

Acute exposure of mice and rats to 300,000 ppm of vinyl chloride for 30 minutes resulted in kidney congestion (Mastromatteo et al. 1960). Also, the kidneys of one mouse out of five exposed to either 100,000 or 200,000 ppm of vinyl chloride for 30 minutes showed degenerative changes (Mastromatteo et al. 1960). Exposure of rats to 50,000 ppm for 8 hours/day for 19 consecutive days or 20,000 ppm for 8 hours/day, 5 days/week for 92 days produced no adverse effects on the kidneys (Lester et al. 1963). However, exposures of male rats to 3,000 ppm for 6 hours/day, 6 days/week, for 3 months produced an increase in the kidneys-to-body-weight ratio (Bi et al. 1985). After a 6-month observation period, there was also an increased kidneys-to-body-weight ratio noted in the male rats exposed to 100 ppm vinyl chloride for 6 hours/day, 6 days/week for 12 months; no effect was noted at 10 ppm (Bi et al. 1985). Relative kidney weights were increased in male rats exposed to 500 ppm vinyl chloride for 5 hours/day, 5 days/week, for 10 months, although no histopathological changes in the kidney were noted (Sokal et al. 1980). No changes in kidney weights were reported when mice were exposed to 1,000 ppm vinyl chloride 6 hours/day, 5 days/week for 8 weeks (Sharma and Gehring 1979). Urinalysis values were within normal limits in rats and rabbits exposed to 200 ppm vinyl chloride for up to 7 hours/day, 5 days/week, for 6 months (Torkelson et al. 1961). One year of exposure to 5,000 ppm vinyl chloride for 7 hours/day, 5 days/week produced an increase in the kidneys-to-body-weight ratio (Feron et al. 1979a) and tubular nephrosis in rats (Feron and Kroes 1979). However, the statistical significance of these findings was not reported in the study.

Endocrine Effects. A study of workers exposed to vinyl chloride in PVC manufacturing plants reported that most workers who presented with scleroderma were shown to have thyroid insufficiency (Suciu et al. 1963).

No histopathological effects on the adrenals were reported in guinea pigs exposed to 400,000 ppm for 30 minutes (Mastromatteo et al. 1960). Rats exposed to 30,000 ppm vinyl chloride 4 hours/day, 5 days/week for 12 months were found to have colloid goiter and markedly increased numbers of perifollicular cells (Viola 1970).

Dermal Effects. Occupational exposure to vinyl chloride was observed to produce scleroderma-like skin changes on the hands of a small percentage of exposed workers (Freudiger et al. 1988; Lilis et al. 1975; Marsteller et al. 1975; Suciu et al. 1963, 1975; Veltman et al. 1975; Walker 1976). The skin changes were characterized by a thickening of the skin (Lilis et al. 1975; Markowitz et al. 1972; Ostlere et al. 1992; Preston et al. 1976; Veltman et al. 1975; Walker 1976), decreased elasticity (Lilis et al. 1975), and edema (Lilis et al. 1975; Suciu et al. 1975) and were almost exclusively observed in exposed individuals who also suffered from Raynaud's phenomenon. Skin biopsies revealed increased collagen bundles in the subepidermal layer of the skin (Harris and Adams 1967; Markowitz et al. 1972; Ostlere et al. 1992; Veltman et al. 1975). Biochemical analyses by Jayson et al. (1976) demonstrated that a high rate of collagen synthesis was taking place in the affected skin. Most often the skin changes were confined to the hands and wrists, but Jayson et al. (1976) reported scleroderma-like skin changes on the hands, arms, chest, and face of one afflicted worker.

Skin changes were observed in rats exposed to 30,000 ppm for 12 months (Viola 1970). The skin of the paws of the exposed rats showed areas of hyperkeratosis, thickening of the epidermis, edema, collagen dissociation, and fragmentation of the elastic reticulum. Interpretation of these results is limited by the absence of a statistical analysis and insufficient information on the treatment of control animals. Lester et al. (1963) reported that male rats exposed to 50,000 ppm vinyl chloride 8 hours/day for 19 days had thin coats and scaly tails, while females exposed to the same concentration showed no effects. For further information regarding scleroderma-like responses to vinyl chloride exposure, see Immunological and Lymphoreticular Effects (Section 3.2.1.3).

Ocular Effects. Ocular effects that have been reported after inhalation exposure are believed to have resulted from direct contact of the vinyl chloride gas with the eyes and are discussed under Dermal Exposure (Section 3.2.3.2). No studies were located regarding ocular effects in humans that were related solely to the inhalation of vinyl chloride. No histopathological changes were noted in the eyes of guinea pigs exposed to 400,000 ppm vinyl chloride for 30 minutes (Mastromatteo et al. 1960).

Body Weight Effects. Several studies have reported that workers intoxicated by vinyl chloride experienced anorexia (Suciu et al. 1963, 1975).

No effects on body weight were noted in ICR mice exposed to either 10,000 ppm vinyl chloride 4 hours/day for 5 days or to 5,000 ppm vinyl chloride 4 hours/day for 6 days (Kudo et al. 1990). No

consistent or dose-related differences in body weight were noted between control rats and rats exposed to up to 50,000 ppm for 1 hour or rats exposed to 500 ppm 5 days/week, for 2 weeks (Hehir et al. 1981). However, statistical analysis was not performed. No changes in body weight gain were noted in rats or rabbits exposed to 200 ppm vinyl chloride 7 hours/day, 5 days/week for 6 months (Torkelson et al. 1961) or in mice exposed to 1,000 ppm vinyl chloride 6 hours/day, 5 days/week for 8 weeks (Sharma and Gehring 1979). Significant decreases were found in the body weight of rats exposed to 100 ppm vinyl chloride 6 hours/day, 6 days/week for 12 months; these changes were not observed at 10 ppm (Bi et al. 1985). Significant decreases were also noted in mean body weights of rats exposed to 5,000 ppm vinyl chloride 7 hours/day, 5 days/week for 4–52 weeks but these data were not quantified (Feron et al. 1979a). This study was limited since only one concentration was tested. Body weight was decreased 10% in male rats exposed to 50 ppm vinyl chloride 5 hours/day, 5 days/week for 10 months (Sokal et al. 1980). Maternal body weight gain was significantly decreased in mice exposed to 500 ppm for 7 hours/day during gestation days (Gd) 6–15 (John et al. 1977).

3.2.1.3 Immunological and Lymphoreticular Effects

A number of studies have examined the immunologic profiles of workers occupationally exposed to vinyl chloride. Male workers exposed to vinyl chloride for an average of 8 years, with concentrations ranging from 1 to 300 ppm during sampling periods, were found to have significantly increased percentages of lymphocytes compared to controls (Fucic et al. 1995, 1997, 1998). Additionally, 75 out of these 100 workers showed disturbances of mitotic activity in these cells. A statistically significant increase in circulating immune complexes in workers exposed to vinyl chloride was observed when compared to levels in unexposed workers (Bogdanikowa and Zawilska 1984). The increase in circulating immune complexes was greatest in women and in those with duties involving exposure to relatively higher levels of vinyl chloride. Compared to controls, immunoglobulin G (IgG) levels were significantly increased in women exposed to the high levels of vinyl chloride in the same study.

Studies of workers who have developed "vinyl chloride disease," a syndrome consisting of Raynaud's phenomenon, acroosteolysis, joint and muscle pain, enhanced collagen deposition, stiffness of the hands, and scleroderma-like skin changes, indicate that this disease may have an immunologic basis. Sera obtained from patients with varying degrees of severity of symptoms of vinyl chloride disease demonstrate a close correlation between the disease severity and the extent of the immunologic abnormality (Grainger et al. 1980; Langauer-Lewowicka et al. 1976; Ward 1976), although these

symptoms have been reported without immunological findings (Black et al. 1986; Ostlere et al. 1992). The most frequent immunologic finding in workers with vinyl chloride disease is an increase in circulating immune complexes or cryoglobulinemia. In workers with the most severe clinical signs, there also are an increased incidence of B-cell proliferation, hyperimmunoglobulinemia (Ward 1976), cryoglobulinemia (Grainger et al. 1980), and complement activation (Grainger et al. 1980; Ward 1976). Evidence of a structurally altered IgG has been obtained, and it has been proposed that vinyl chloride (or a metabolite) binds to IgG (Grainger et al. 1980).

Based on the similarity of vinyl chloride disease and systemic sclerosis, which may be a genetically linked autoimmune disease, Black et al. (1983, 1986) examined the human lymphocyte antigen (HLA) phenotypes of patients with vinyl chloride disease. Many autoimmune diseases show statistically significant associations with certain HLA alleles. These authors found that when compared to unexposed controls or asymptomatic controls, workers with vinyl chloride disease had a significantly greater incidence of possessing the HLA-DR5 allele. Furthermore, among those with the disease, the severity of the symptoms was significantly related to the possession of the HLA-DR3 and B8 alleles. These authors concluded that susceptibility was increased in the presence of HLA-DR5 or a gene in linkage disequilibrium with it, and progression was favored by HLA-DR3 and B8 phenotypes. Immune system dysfunction has also been linked to a case of polymyositis (i.e., muscle fiber necrosis and atrophy) in an exposed worker, with involvement of antibodies to histidyl-t-RNA synthetase (Jo-1) (Serratrice et al. 2001).

Splenomegaly was reported in a number of studies (Ho et al. 1991; Marsteller et al. 1975; Popper and Thomas 1975; Suciu et al. 1963; Veltman et al. 1975). No histopathological changes were noted in the spleen or lymph nodes of guinea pigs exposed to 400,000 ppm vinyl chloride for 30 minutes (Mastromatteo et al. 1960). An increase in the relative spleen weight was observed in rats exposed to 50 ppm for 5 hours/day, 5 days/week for 10 months (Sokal et al. 1980). Although no dose response was evident, increased relative spleen weight was also reported by Bi et al. (1985) when rats were exposed to either 10 ppm for 6 hours/day, 6 days/week for 6 months or 3,000 ppm for 6 hours/day, 6 days/week for 3 months. This effect was not observed at 100 ppm in the 3-month study (Bi et al. 1985).

The immunologic effects of vinyl chloride have been examined in mice (Sharma and Gehring 1979). Lymphocytes isolated from the spleens of mice exposed to concentrations as low as 10 ppm vinyl chloride 6 hours/day, 5 days/week for 4 weeks had increased spontaneous and lectin-stimulated

transformation. This increase was not observed when lymphocytes from unexposed mice were cultured in the presence of vinyl chloride.

The highest NOAEL value and all reliable LOAEL values for immunological effects in guinea pigs, mice, and rats exposed in acute- and intermediate-duration studies are recorded in Table 3-1 and plotted in Figure 3-1. For further information on Raynaud's phenomenon and scleroderma-like responses to vinyl chloride, see Cardiovascular and Dermal Effects (Section 3.2.1.2).

3.2.1.4 Neurological Effects

Vinyl chloride was once considered for use as an inhalation anesthetic (ACGIH 2003). Investigators studying the effects of vinyl chloride exposure frequently report central nervous system symptoms that are consistent with the anesthetic properties of vinyl chloride. The most commonly reported central nervous system effects are ataxia or dizziness (Ho et al. 1991; Langauer-Lewowicka et al. 1983; Lilis et al. 1975; Marsteller et al. 1975; Spirtas et al. 1975; Suciu et al. 1963, 1975; Veltman et al. 1975), drowsiness or fatigue (Langauer-Lewowicka et al. 1983; Spirtas et al. 1975; Suciu et al. 1963, 1975; Walker 1976), loss of consciousness (NIOSH 1977), and/or headache (Langauer-Lewowicka et al. 1983; Lilis et al. 1975; Marsteller et al. 1975; NIOSH 1977; Spirtas et al. 1975; Suciu et al. 1963, 1975; Veltman et al. 1975). Other central nervous system effects that have been reported by vinyl chloride workers include euphoria and irritability (Suciu et al. 1963, 1975), visual and/or hearing disturbances (Marsteller et al. 1975), nausea (Marsteller et al. 1975; Spirtas et al. 1975), memory loss (Langauer-Lewowicka et al. 1983; Suciu et al. 1963, 1975), and nervousness and sleep disturbances (Langauer-Lewowicka et al. 1983; Suciu et al. 1963). Central nervous system tests revealed pyramidal signs and cerebellar disturbances in some exposed subjects (Langauer-Lewowicka et al. 1983); however, reliable estimates of exposure levels producing these effects were not available.

Exposure of volunteers to known levels of vinyl chloride has provided some indication of the levels of vinyl chloride associated with the effects noted above. Volunteers exposed to 25,000 ppm vinyl chloride for 3 minutes, in a single-exposure study, reported experiencing dizziness, disorientation, and burning sensations in the feet during exposure (Patty et al. 1930). Recovery from these effects was rapid upon termination of exposure, but the subjects developed headaches. Exposure of volunteers to concentrations of vinyl chloride ranging from 4,000 to 20,000 ppm for 5 minutes twice a day in periods separated by 6 hours on 3 consecutive days was studied by Lester et al. (1963). No effects were noted at 4,000 ppm.

However, at 8,000 ppm one of six subjects reported feeling dizzy. The incidence of dizziness increased at higher concentrations. Nausea was experienced at higher concentrations, and recovery from all effects was rapid upon termination of exposure. Headaches developed following exposure to 20,000 ppm.

Indications of an exposure-related peripheral neuropathy have been observed in a number of the occupational studies. A peripheral neuropathy, most severe in hands and feet, was diagnosed in 70% of the vinyl chloride workers examined in a study by Perticoni et al. (1986). The peripheral neuropathy was manifested as denervation-related fasciculations and fibrillations and increased duration and amplitude of motor unit potentials (indicating collateral sprouting). Similar effects were observed by Magnavita et al. (1986) in a case study of a vinyl chloride worker. Other peripheral nervous system symptoms have been reported by a number of investigators studying the effects of occupational exposure to vinyl chloride. The symptom most frequently reported was tingling (paresthesia) in the extremities (Lilis et al. 1975; Sakabe 1975; Spirtas et al. 1975; Suciu et al. 1963, 1975; Veltman et al. 1975; Walker 1976). Additional peripheral nervous system symptoms included numbness in the fingers (Lilis et al. 1975; Sakabe 1975), weakness (Langauer-Lewowicka et al. 1983; Suciu et al. 1963, 1975), depressed reflexes (NIOSH 1977), warmth in the extremities (Suciu et al. 1963, 1975), and pain in the fingers (Sakabe 1975). It is unclear whether some of these symptoms are associated with tissue anoxia due to vascular insufficiency, or whether they represent the direct toxic effects of vinyl chloride on peripheral nerves.

Acute exposure of a number of species to high levels of vinyl chloride has provided additional information on the characteristics of the central nervous system effects that are produced. Exposure of guinea pigs to 10,000 ppm for 8 hours (Patty et al. 1930) was observed to be without effects. Exposure to 25,000 ppm resulted in ataxia, which developed into unconsciousness during the 8-hour exposure. As the concentration was increased, the development of unconsciousness was more rapid. At 100,000 ppm, Mastromatteo et al. (1960) observed the development of unconsciousness within 30 minutes. Mice experienced similar signs at approximately equivalent exposure levels. At 5,000 ppm, vinyl chloride was without effect during a 1-hour exposure. Exposure to 50,000 ppm produced ataxia and twitching (Hehir et al. 1981), and at 100,000 ppm for 30 minutes, unconsciousness was produced, proceeded by increased motor activity, incoordination, twitching, and tremors (Mastromatteo et al. 1960). Similar effects in rats were observed by Lester et al. (1963), Jaeger et al. (1974), and Mastromatteo et al. (1960). In contrast, in two reports using rats, exposure to 50,000 ppm for either 1 or 6 hours was without effect (Hehir et al. 1981; Jaeger et al. 1974). No effects were noted in rats exposed to 500 ppm vinyl chloride for 2 weeks (1 hour/day, 5 days/week) or in rats exposed to 50 ppm for 20 weeks (1 hour/day, 5 days/week) (Hehir et

al. 1981). In addition, tolerance developed to the intoxicating effects of exposure to 50,000 ppm vinyl chloride after five or six 8-hour exposures (Lester et al. 1963).

Chronic exposure of rats to high levels of vinyl chloride has produced damage to nervous tissue. Rats exposed to 30,000 ppm for 4 hours/day, 5 days/week for 12 months in a single-concentration study were soporific during exposures (Viola 1970; Viola et al. 1971). Following 10 months of exposure, the rats had decreased responses to external stimuli and disturbed equilibrium. Histopathological examination revealed diffuse degeneration of gray and white matter. Cerebellar degeneration in the Purkinje cell layer was pronounced. Also, peripheral nerve endings were surrounded and infiltrated with fibrous tissue (Viola 1970; Viola et al. 1971). Nonneoplastic lesions in the brain were not noted in rats exposed to 5,000 ppm for 7 hours/day, 5 days/week for 12 months in a single-concentration study by Feron and Kroes (1979).

The highest NOAEL values and all reliable LOAEL values for neurological effects in each species from acute- or intermediate-duration studies are recorded in Table 3-1 and plotted in Figure 3-1.

3.2.1.5 Reproductive Effects

A number of case reports of workers occupationally exposed to vinyl chloride suggest that sexual performance may be affected by vinyl chloride. However, these studies are limited by the lack of quantification of exposure levels and possible concomitant exposures to other chemicals. Sexual impotence was reported by 24% of the workers examined by Suciu et al. (1975). Approximately 20% of the workers examined by Veltman et al. (1975) complained of potency troubles. A loss of libido in 35% and impotence and decreased androgen secretion in 8% of workers exposed at least once to very high levels of vinyl chloride were also reported by Walker (1976).

In retrospective and prospective studies by Bao et al. (1988), increased incidence and severity of elevated blood pressure and edema during pregnancy (preeclampsia) were found in female workers exposed to vinyl chloride when compared to unexposed workers. Company records indicated that exposure levels ranged from 3.9 to 89.3 ppm during the retrospective study and from 0.2 to 130.7 ppm during the prospective study. More detailed information regarding the exposure levels was not presented.

A 2-generation reproductive toxicity study was conducted in rats exposed to vinyl chloride via inhalation (Thornton et al. 2002). Male and female Sprague-Dawley rats were exposed to 0, 10, 100, or 1,100 ppm vinyl chloride 6 hours/day for a 10-week premating period and a 3-week mating period. No adverse effects were noted in reproductive capability over the two generations at any dose. No effects were seen in body weight, feed consumption, ability to reproduce, gestation index or length, or pre- and postweaning developmental landmarks. Sperm counts, motility, and morphology were also unaffected by vinyl chloride exposure. Changes in liver weights and/or histopathological alterations were seen in F_0 and F_1 generation male and female rats. For further information regarding the liver toxicity of vinyl chloride, see Section 3.2.1.2.

Two dominant lethal studies examined the reproductive performance of exposed male rats. A brief exposure (5 days, 6 hours/day) of mice to concentrations of vinyl chloride as high as 30,000 ppm had no effect on male fertility or pre- or postimplantation loss (Anderson et al. 1976). In contrast, exposure of male rats to concentrations as low as 250 ppm for 6 hours/day, 5 days/week for 11 weeks produced a decrease in the ratio of pregnant to mated females, indicating a decrease in male fertility; this effect was not observed at 50 ppm (Short et al. 1977). These results are supported by two studies using rats in which adverse effects of vinyl chloride on the testes were observed (Bi et al. 1985; Sokal et al. 1980). Exposure of rats to 100 ppm for 6 hours/day, 6 days/week for 12 months produced a significant increase in the incidence of damage to the seminiferous tubules and depletion of spermatocytes (Bi et al. 1985). At the 6-month interim sacrifice, a significant decrease in testicular weight was also observed at 100 ppm. No effect on male reproductive organs was observed in this study at 10 ppm. Several methodological limitations have been identified for this study. Temperature and humidity conditions in the inhalation chambers were not maintained within the normal range. Inhalation chamber volume and air flow were also not held constant across dose groups. A significant increase in damage to the spermatogenic epithelium and disorders of spermatogenesis were found with exposure to 500 ppm vinyl chloride for 5 hours/day, 5 days/week for 10 months, but was not observed after exposure to 50 ppm vinyl chloride (Sokal et al. 1980). Temperature and relative humidity values were not reported for this study. No significant change in testicular weight was found in rats exposed to 500 ppm for 7 hours/day, 5 days/week for 4.5 months or in dogs, rabbits, or guinea pigs exposed to 200 ppm for 7 hours/day, 5 days/week for 6 months (Torkelson et al. 1961). However, the quality of this study is limited because of the small number of animals tested. Exposures involved up to 10 rats or guinea pigs of each gender, three rabbits of each sex, and one dog of each sex. No histopathological data on the testes of these animals were presented.

The highest NOAEL values and 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.

3.2.1.6 Developmental Effects

Although evidence has been presented indicating that members of communities with nearby vinyl chloride polymerization facilities have significantly greater incidences of some forms of developmental toxicity, these studies failed to demonstrate a statistically significant correlation between the developmental toxicity and either parental occupation or proximity to the facility (Edmonds et al. 1978; Infante 1976; Rosenman et al. 1989; Theriault et al. 1983).

The pregnancy outcome of wives of workers employed at a vinyl chloride polymerization facility was compared to the pregnancy outcome of wives of a control group made up of unexposed rubber workers and PVC fabricators believed to be exposed to "very low" levels of vinyl chloride (Infante et al. 1976a, 1976b). Pregnancy outcomes were determined based on the responses given by fathers on a questionnaire. Infante et al. (1976a, 1976b) and NIOSH (1977) reported a significant excess of fetal loss in the group whose husbands had been exposed to vinyl chloride. The greatest difference occurred in wives of men under 30 years of age, where fetal loss was 5.3% for controls and 20.0% for exposed workers. However, this study has been severely criticized based on the conduct of the study and method of statistical analysis used (Hatch et al. 1981; Stallones 1987). Furthermore, Hatch et al. (1981) and Stallones (1987) concluded that the study failed to demonstrate an association of parental exposure to vinyl chloride with increased fetal loss.

Additional work by Infante (1976) and Infante et al. (1976b) examined the occurrence of congenital malformations among populations exposed to emissions from PVC polymerization facilities. A statistically significant increase in birth defects was observed in three cities in which polymerization facilities were located when compared to statewide and countywide averages. The greatest increases were noted in malformations of the central nervous system, upper alimentary tract, and genital organs and in the incidence of club foot. However, this study has also been criticized based on the conduct and analyses used (Hatch et al. 1981; Stallones 1987). These authors concluded that the study failed to demonstrate an association between exposure to emissions and the prevalence of birth defects. Furthermore, another study that examined the incidence of malformations in one of the cities studied by Infante (1976) concluded that, although the city had statistically increased incidences of congenital

malformations, no correlation existed with parental proximity to the polymerization plant or with parental employment at the plant (Edmonds et al. 1975). In fact, more parents of control infants worked at the plant or lived closer to the plant than parents of infants with central nervous system malformations.

Additional studies have also examined the prevalence of congenital malformations in populations exposed to emissions from polymerization facilities (Edmonds et al. 1978; Rosenman et al. 1989; Theriault et al. 1983). The incidence of central nervous system defects in a West Virginia county with a polymerization plant was compared to incidences in other regions in the United States with no known exposure to vinyl chloride (Edmonds et al. 1978). Although the rate of central nervous system defects in the West Virginia county exceeded that in control areas, no correlation was noted between the increased central nervous system defects and parental occupation or potential exposure based on proximity to the plant or prevailing wind patterns.

A significantly greater prevalence of birth defects was found in residents of a town with a polymerization facility than in three matched towns without potential for exposure to vinyl chloride (Theriault et al. 1983). The most commonly reported defects included those of the musculoskeletal, alimentary, urogenital, and central nervous systems. The incidences were observed to fluctuate with seasonal changes in emissions. However, no correlations were found between the presence of defects and proximity of the residence to the plant or parental occupation. Also, other industrial emissions could not be eliminated as potential sources of the increased incidence of congenital malformations observed and additional confounding factors such as nutritional status, smoking, and alcohol and other drug use were not eliminated.

No significant increases in birth defects were observed in a community with two polymerization facilities, but odds ratios for central nervous system defects were found to correlate with the amount of emissions from the individual facilities and with the distance of the residences of affected parents from the facilities (Rosenman et al. 1989). However, this study was limited by the small sample size.

Pregnancy outcomes of mothers occupationally exposed to vinyl chloride for >1 year were compared to those of pregnant workers not exposed to vinyl chloride in retrospective and prospective studies (Bao et al. 1988). Company records indicated that exposure levels ranged from 3.9 to 89.3 ppm during the retrospective study and from 0.2 to 130.7 ppm during the prospective study. More detailed information regarding the exposure levels was not presented. The study authors concluded that exposure to vinyl

chloride did not correlate with changes in sex ratio, birth weight or height, perinatal mortality, or the incidence of congenital abnormalities.

A number of inhalation studies have examined the effects of vinyl chloride exposure on pregnancy outcome in animals. Results of these studies indicate that vinyl chloride produces adverse developmental effects at concentrations that are also toxic to maternal animals. John et al. (1977, 1981) exposed rats and rabbits to 0, 500, or 2,500 ppm and mice to 0, 50, or 500 ppm throughout the period of organogenesis. Separate control groups were used for each of the mice exposure concentrations. Mice were most sensitive to the effects of vinyl chloride. In mice exposed to 500 ppm, maternal toxicity was evidenced by decreased food consumption, decreased body weight gain, and increased mortality rate (John et al. 1977, 1981). Delayed ossification was noted in fetuses at 500 ppm. The only significant fetal effect observed at 50 ppm was an increase in crown-rump length. The biological significance of this effect is unknown. Based on this NOAEL of 50 ppm, an acute-duration MRL of 0.5 ppm was calculated as described in the footnote in Table 3-1. In rats, 500 ppm produced decreased maternal weight gain and fetal weight, increased crown-rump length, and vertebral lumbar spurs. Increasing the exposure level to 2,500 ppm was not associated with a dose-dependent increase in these effects. The only effects observed at 2,500 ppm were decreased maternal food consumption and, in fetuses, an increased incidence of dilated ureters. In rabbits exposed to 500 ppm, maternal animals had decreased food consumption, and fetal animals had delayed ossification. These effects were not observed in rabbits at 2,500 ppm. However, the number of animals that were tested at 2,500 ppm was much lower than at 500 ppm (5 versus 20); thus, no conclusions may be drawn as to the dose response of these effects.

An embryo-fetal developmental toxicity study was conducted in rats exposed to vinyl chloride via inhalation (Thornton et al. 2002). Female Sprague-Dawley rats were exposed to 0, 10, 100, or 1,100 ppm vinyl chloride 6 hours/day on Gd 6–19. No adverse effects were noted in embryo-fetal developmental parameters including uterine implantation, fetal gender distribution, fetal body weight, and fetal malformations and variations. Vinyl chloride produced a decrease in maternal body weight gain at all exposure levels; however, no changes were observed in feed consumption, clinical signs, or postmortem gross findings. Maternal liver and kidney weights were increased relative to total body weight.

Exposure of rats to either 0 or 1,500 ppm of vinyl chloride during the first, second, or third trimester of pregnancy was examined (Ungvary et al. 1978). In maternal animals, an increased liver-to-body weight ratio was observed in those exposed during the first and second trimesters, but no histopathologic alterations were found. A significant increase in resorptions was observed in animals exposed during the

first trimester of pregnancy. Two central nervous system malformations (microphthalmia and anophthalmia) were observed in exposed fetuses but not in controls, but the incidence of these malformations did not reach statistical significance. This study is limited in that only a single concentration of vinyl chloride was tested, precluding conclusions as to the dose-response relationship of the effects observed.

The effects of exposure of rats to vinyl chloride throughout gestation were examined by Mirkova et al. (1978) and Sal'nikova and Kotsovskaya (1980). An unspecified number of pregnant rats were exposed to 0, 1.9, or 13.9 ppm for 4 hours/day for the 21 days of gestation. Fetuses were examined for abnormalities just prior to the end of gestation, and offspring were examined at 6 months postparturition (Sal'nikova and Kotsovskaya 1980). At 13.9 ppm, a decrease in maternal erythrocyte count was observed. At 1.9 and 13.9 ppm, fetuses had an increased incidence of hemorrhages, and at 13.9 ppm, increased edema. However, the affected organs were not specified. Rats examined at 6 months, following *in utero* exposure to 1.9 ppm, were found to have decreased hemoglobin and leukocytes and decreased organ weights (males: liver, kidneys, spleen; females: lung, liver). In addition to these effects, exposure to 13.9 ppm *in utero* resulted in an increased hexanol sleep time and a decreased ability of the rats to orient themselves.

Continuous exposure of an unspecified number of rats throughout gestation to 2.4 ppm of vinyl chloride resulted in decreased fetal weight and increased early postimplantation loss, hematomas, and hydrocephaly with intracerebral hematoma. Weanling rats had hepatotoxic effects including decreased bile enzyme activity, decreased bile secretion, and decreased cholic acid content,. No histological data on the livers of pups, or information regarding maternal health, or statistical analyses of the data were presented (Mirkova et al. 1978). Also, both this study and the report by Sal'nikova and Kotsovskaya (1980) failed to provide information on the number of animals in each test group.

The developmental toxicity of vinyl chloride was examined using a whole embryo culture system (Zhao et al. 1996). Vinyl chloride induced embryo growth retardation, but was not shown to be teratogenic in the rat *in vitro* whole embryo culture system.

The highest NOAEL value and all reliable LOAEL values for developmental effects in mice, rats, and/or rabbits in acute-duration studies are recorded in Table 3-1 and plotted in Figure 3-1.

3.2.1.7 Cancer

A recent review pooled the analyses of worker cohorts from 56 vinyl chloride plants in North America and Europe (Bosetti et al. 2003). This analysis includes over 22,000 workers and represents the most comprehensive data on occupational risks of vinyl chloride exposure. An elevated risk of liver cancer mortality was observed. While differences between the two cohorts were observed for excess soft tissue sarcoma and brain cancer, no significant excess for these effects were seen in the pooled data. Deaths from lung and laryngeal cancer were lower than expected, and no excess cancer risk was observed for lymphoid and hematopoietic system cancers. The most compelling evidence for the carcinogenic potential of vinyl chloride in humans comes from the cluster of reports of greater than expected incidences of angiosarcoma of the liver in workers occupationally exposed to vinyl chloride (Byren et al. 1976; Creech and Johnson 1974; Forman et al. 1985; Fox and Collier 1977; Infante et al. 1976b; Jones et al. 1988; Laplanche et al. 1992; Lee et al. 1996; Monson et al. 1975; Pirastu et al. 1990; Rinsky et al. 1988; Simonato et al. 1991; Teta et al. 1990; Theriault and Allard 1981; Waxweiler et al. 1976; Weber et al. 1981; Wong et al. 1991; Wu et al. 1989). Angiosarcoma of the liver is considered to be a very rare type of cancer (25–30 cases/year in the United States) (Heath et al. 1975). However, approximately 30 years after the introduction of vinyl chloride for use in the industrial production of PVC, it became apparent that workers exposed to high levels of vinyl chloride had an unusually high incidence of this type of tumor. Investigators identified an increased likelihood of developing hepatic angiosarcoma among those exposed to the highest levels of vinyl chloride and those exposed to vinyl chloride for the longest duration (Fortwengler et al. 1999; Fox and Collier 1977; Infante et al. 1976b; Jones et al. 1988; Rinsky et al. 1988; Weber et al. 1981; Wong et al. 1991; Wu et al. 1989). Angiosarcoma of the liver was not found in residents living in the vicinity of vinyl chloride sites, unless they were also exposed to high concentrations of vinyl chloride in the workplace (Elliott and Kleinschmidt 1997). Based on this information, vinyl chloride is considered to be a carcinogen in humans (EPA 1994c; IARC 1987).

Histopathological examination of liver tissue from humans with hepatic angiosarcoma has led to the hypothesis that angiosarcoma develops as a result of hyperplastic changes in sinusoidal cells. Areas of transition to angiosarcoma contained greatly increased numbers of sinusoidal cells with greatly expanded sinusoidal spaces. Also, hepatic cells were replaced by fibrous tissue forming trabeculae. These areas also showed infiltration of angiosarcoma cells. In fully developed angiosarcoma, multiple areas with nodules of angiosarcoma cells were noted, the centers of which exhibited hemorrhagic necrosis (Popper et al. 1981). A recent case report suggests that vinyl chloride can also produce malignant hemangio-pericytoma in the liver, which is a vascular tumor similar to angiosarcoma (Hozo et al. 1997, 2000).

Other liver tumors, including hepatocellular carcinoma and cholangiocellular carcinoma, have also been associated with occupational exposure to vinyl chloride (Cheng et al. 1999; Du and Wang 1998; Lelbach 1996; Saurin et al. 1997; Ward et al. 2001; Weihrauch et al. 2000; Wong et al. 2002a, 2003a). A meta-analysis of eight independent studies confirms an increased risk of hepatocellular carcinoma for occupational workers exposed to vinyl chloride (Boffetta et al. 2003). The risk of developing liver cancer appears elevated in those with a history of Hepatitis B viral infection (Du and Wang 1998; Wong et al. 2003a). Mastrangelo et al (2004) evaluated the possible interaction between alcohol consumption, hepatitis infection and hepatocellular carcinoma in a large cohort of vinyl chloride workers. Vinyl chloride was suggested to be an independent risk factor for hepatocellular carcinoma with a synergistic interaction described for alcohol consumption and an additive interaction observed for hepatitis infection. Lewis (2003) reports the continuing occurrence of angiosarcoma of the liver in retirees from a PVC production plant in Louisville, Kentucky. This ongoing incidence is reported primarily for those workers employed prior to 1960, suggesting that those exposed to the highest concentrations of vinyl chloride remain at risk for developing cancer for the remainder of their lives. The reported latency period for workers diagnosed prior to 1975 was 12–28 years, while those diagnosed after 1975 showed a latency of 27–47 years.

Other cancers that have shown a statistically significant increase in mortality rate among vinyl chloride workers, in at least some studies, include cancer of the brain and central nervous system, the lung and respiratory tract, connective and other soft tissues, and the lymphatic/hematopoietic system. With regard to cancer of the brain and central nervous system, Cooper (1981), Waxweiler et al. (1976), and Wong et al. (1991) reported statistically significant increases; Monson et al. (1975) reported an increase in central nervous system cancer mortality in a proportional mortality study; Byren et al. (1976), Simonato et al. (1991), and Tabershaw and Gaffey (1974) reported increases that were not statistically significant; and Fox and Collier (1977), Jones et al. (1988), Thomas et al. (1987), and Wu et al. (1989) found no increase in cancer of the central nervous system among workers occupationally exposed to vinyl chloride. It should be noted that the Cooper (1981), Tabershaw and Gaffey (1974), and Wong et al. (1991) studies were all based on the same cohort from a Chemical Manufacturers Association (CMA) study (Wong and Whorton 1993). Workers in the studies by Waxweiler et al. (1976) and Wu et al. (1989) were also employed at the same plants used for the CMA study (Wong and Whorton 1993). At least one analysis of epidemiological studies exposed certain weaknesses in the data that support a causal link between vinyl chloride and brain cancer (Doll 1988).

Follow up mortality studies at polymer production plants indicate that liver cancer mortality remained elevated while brain cancer mortality was markedly reduced (as compared to earlier studies). It should be noted that increased brain cancer incidence was not associated with vinyl chloride exposure in these later studies (Lewis 2001; Lewis and Rempala 2003; Lewis et al. 2003; Mundt et al. 2000). In a meta-analysis of eight independent studies, no statistically significant increase in brain cancer mortality was observed (Boffetta et al. 2003). An IARC update of a European multi-center cohort study was also negative for brain cancer (Ward et al. 2001).

An association between respiratory tract cancer and vinyl chloride exposure has not been consistently observed. A significant increase in cancer of the respiratory tract was reported by Belli et al. (1987), Infante et al. (1976b), and Waxweiler et al. (1976), and also by Monson et al. (1975) in a proportional mortality study. Although smoking history was not considered in these studies, Waxweiler et al. (1976) noted that the types of respiratory tract cancer most frequently recorded were large-cell undifferentiated carcinoma or adenocarcinoma, which are two lung cancer types not usually associated with smoking, but may be due to concomitant exposure. Increased risk of lung cancer was also associated with exposure to high concentrations of polyvinyl chloride dust particles (Mastrangelo et al. 2002). Respiratory tract cancer was not reported as elevated in studies by Buffler et al. (1979), Cheng et al. (1999), Cooper (1981), Fox and Collier (1977), Jones et al. (1988), Mundt et al. (2000), Scelo et al. (2004), Simonato et al. (1991), Wong et al. (1991, 2002a), and Wu et al. (1989). Similarly, a meta-analysis of eight independent studies (Boffetta et al. 2003) and an IARC update of a multi-center cohort study did not demonstrate excess mortality from lung cancer (Ward et al. 2001).

A significant increase in cancers of connective and other soft tissues was observed in a recent follow up mortality study (Mundt et al. 2000) and in a meta-analysis of eight independent studies (Boffetta et al. 2003). Rhomberg (1998) also suggests that vinyl chloride can induce soft tissue sarcoma outside of the liver; however, an IARC update of a multi-center cohort study was negative for soft tissue sarcoma (Ward et al. 2001). A meta-analysis of five occupational exposure studies additionally suggests a weak association between vinyl chloride exposure and pancreatic cancer (Ojajarvi et al. 2001).

A statistically significant increase in cancers of the lymphatic/hematopoietic system was reported by Rinsky et al. (1988), Smulevich et al. (1988), Weber et al. (1981), and Wong et al. (2002a). Monson et al. (1975) also reported an increase in their proportional mortality study. However, no statistically significant increase in these types of cancer was reported by Infante et al. (1976b), Jones et al. (1988), Mundt et al. (2000), or Wong et al. (1991). In a meta-analysis of eight independent studies, the mortality

data for cancers of the lymphatic/hematopoietic system were highly variable. A strong association was not observed between vinyl chloride exposure and lymphatic/hematopoietic system cancers; however, a negative conclusion was considered premature (Boffetta et al. 2003).

An increased incidence of malignant melanoma among vinyl chloride workers has been reported (Heldaas et al. 1984, 1987), but the significance of this finding has been disputed (ten Berge 1987). A follow up to the original Heldaas (1984, 1987) studies reported only one additional case of melanoma between 1985 and 1993, weakening the proposed association between vinyl chloride exposure and the development of malignant melanoma (Langard et al. 2000).

Few studies directly address the incidence of cancer in women occupationally exposed to vinyl chloride. However, one study found that women employed in the production of vinyl chloride and PVC had a significantly greater chance of developing leukemia or lymphomas (Smulevich et al. 1988). Furthermore, the subgroup of women who were exposed to the highest levels of vinyl chloride had increased incidences of stomach cancer and the highest incidences of leukemia and lymphoma. No significant increase in any type of cancer was observed in exposed males in this report, irrespective of the level of exposure.

The human epidemiology data demonstrate a clear association between vinyl chloride exposure and liver cancer (i.e., angiosarcoma and hepatocellular carcinoma). Although other cancers have been previously reported for vinyl chloride workers (i.e., respiratory tract cancer, brain cancer), recent follow-up studies do not demonstrate a consistent association between vinyl chloride exposure and tumor formation in these organ systems (Boffetta et al. 2003; Lewis 2001; Lewis and Rempala 2003; Lewis et al. 2003; Mundt et al. 2000; Ward et al. 2001).

Studies in several animal species support the conclusion that vinyl chloride is carcinogenic. A large series of experiments was performed by Maltoni et al. (1981) using rats (Sprague-Dawley and Wistar), mice, and hamsters. All animals were chamber exposed; controls were chamber exposed to air only. The test material was >99.9% pure. A complete gross and histopathological examination of every animal was performed. However, extremely limited histopathological data were presented and cancer incidences were presented only in summary tables. Also, survival of control animals was poor in some of the experiments. Furthermore, statistical analyses, where present, appear to be based on a compilation of data from several individual studies. In one group of studies, Maltoni et al. (1981) exposed Sprague-Dawley rats to vinyl chloride for 52 weeks at concentrations ranging from 1 to 30,000 ppm. Animals were examined at the time of their spontaneous death. Statistically significant increases were noted in the

incidence of mammary gland carcinomas, Zymbal gland carcinomas, nephroblastoma, and liver angiosarcoma. Exposure of Swiss mice to 50 ppm vinyl chloride for 4 hours/day, 5 days/week for 30 weeks also appeared to increase the incidence of liver angiosarcoma and angioma (Maltoni et al. 1981). Maltoni et al. (1981) also reported that decreasing the duration of exposure decreased the incidence of vinyl chloride-related tumors (nephroblastomas, liver angiosarcomas, Zymbal gland carcinomas, and to some extent, neuroblastomas), but statistics were not presented to support these conclusions.

Some variation in the target organs that developed tumors was observed when different species were exposed to vinyl chloride (Maltoni et al. 1981). Whereas angiosarcomas of the liver were reported to occur in rats, mice, and hamsters, mammary gland carcinomas were found only in rats and mice; Zymbal gland carcinomas, neuroblastomas, and nephroblastomas were found only in rats; lung tumors were found only in mice; and melanomas, acoustical duct epithelial tumors, and leukemias were found only in hamsters.

Other inhalation experiments support the carcinogenicity of vinyl chloride. Rats and mice exposed to 0, 50, 250, or 1,000 ppm for 6 hours/day, 5 days/week for 6 months (Hong et al. 1981) or up to 12 months (Lee et al. 1977a, 1978) had a significantly increased incidence of hemangiosarcoma of the liver at ≥250 ppm. Increases in bronchio-alveolar adenoma of the lung and mammary gland tumors (adenocarcinomas, squamous and anaplastic cell carcinomas) were also observed in mice at ≥50 ppm, although it is unclear whether the increases in these tumor types are statistically significant (Lee et al. 1977a, 1978). Mice exposed to 50 or 500 ppm vinyl chloride for 6 hours/day, 5 days/week for 6 months or 1 year had an increased incidence of lung adenoma, as well as hemangiosarcoma of fat tissue in various organs (Holmberg et al. 1976). Only one liver hemangiosarcoma was noted. Male rats exposed to concentrations as low as 100 ppm for 6 hours/day, 6 days/week, for 12 months had significantly increased incidence of cancer, including angiosarcoma of the liver and lung, when sacrificed at 18 months (Bi et al. 1985). Rats exposed to 30,000 ppm vinyl chloride 4 hours/day, 5 days/week, for 12 months had an increased incidence of epidermoid carcinoma of the skin, adenocarcinoma of the lungs, and osteochondroma in the bones (Viola et al. 1971), and rats exposed to 5,000 ppm for 52 weeks had primary tumors in the brain, lung, Zymbal gland, and nasal cavity (Feron and Kroes 1979). However, these studies (Feron and Kroes 1979; Viola et al. 1971) are limited by the absence of statistical analysis of the data. A concentration-dependent increase in tumor formation (alveologenic adenomas of the lung, angiosarcomas of the liver, and adenosquamous carcinoma of the mammary gland) was observed in mice exposed to 0, 50, 200, or 2,500 ppm vinyl chloride in a study performed for the Manufacturing Chemists

Association (Keplinger et al. 1975). However, no statistics were presented to support these conclusions. Furthermore, an audit of data performed for the Manufacturing Chemists Association (CMA 1979) indicated that mishandling of the tissues precluded making statements regarding the relationship of tumors other than angiosarcoma of the liver to vinyl chloride exposure. Female mice exposed to 50 ppm vinyl chloride showed increased incidence of hemangiosarcoma of the subcutis and peritoneum as well as tumors of the lung and mammary gland (Drew et al. 1983), i.e., hemangiosarcoma of the skin, spleen, or liver and mammary gland carcinomas.

In a preliminary study with a limited number of animals, alveogenic lung tumors developed in 26 of 27 mice exposed to 2,500 or 6,000 ppm for 5–6 months (Suzuki 1978). A concentration-related increase in the incidence of alveogenic tumors was observed in a study in which a greater number of mice were exposed to 0–600 ppm for 4 weeks and then observed for up to 40 weeks postexposure (Suzuki 1983). The lowest concentration at which multiple foci tumors were observed was 100 ppm (Suzuki 1983.) A significant increase in the incidence of pulmonary adenomas was reported in mice exposed to 50 ppm, 6 hours/day, 5 days/week for 6 months (Adkins et al. 1986). An increase in bronchio-alveolar adenoma was observed in a lifespan study in mice that were exposed to 50 ppm for 100 1-hour exposures, 500 ppm for 10 1-hour exposures, or 5,000 ppm for a single 1-hour exposure (Hehir et al. 1981). The statistical significance of these observations was not reported.

Some data suggest that exposure of animals early in their lives may increase the likelihood of developing tumors due to the latency period for vinyl chloride-induced cancer (Drew et al. 1983). Early life exposure may also affect the type of tumor that develops (Maltoni et al. 1981). When hamsters, mice, and rats were exposed to vinyl chloride for periods of 6–24 months starting at various times after weaning, the incidence of tumors such as hemangiosarcoma of the liver, skin, and spleen, and angiosarcoma of the stomach was greater when animals were exposed for 12 months immediately after weaning than if animals were held for 12 months and then exposed for the next 12 months (Drew et al. 1983). Mammary gland carcinoma was also significantly increased when 2- or 8-month-old hamsters, but not 14- or 20-month-old hamsters, were exposed to 200 ppm vinyl chloride for 6 months (Drew et al. 1983). Fibroadenoma of the mammary gland was also increased in female rats exposed to 100 ppm of vinyl chloride for 6 hours/day, 5 days/week, over 6–24 months (Drew et al. 1983). Also, when pregnant rats were exposed to 6,000 ppm vinyl chloride from gestation day 12 through 18, the incidence of mammary gland carcinomas, Zymbal gland carcinomas, and forestomach epithelial tumors was reported to be greater in transplacentally exposed animals than in maternal animals (Maltoni et al. 1981). At 10,000 ppm in this study, nephroblastomas were increased in transplacentally exposed animals compared

to maternal animals (Maltoni et al. 1981). No control group was used, however, and no statistics were presented to support the conclusions. Maltoni and Cotti (1988) also exposed pregnant rats to 2,500 ppm vinyl chloride starting on Gd 12 and continued to expose both maternal animals and offspring for a total of 76 weeks. Hepatocarcinoma, hepatic angiosarcoma, and neuroblastoma were increased in treated animals compared to controls. The incidence of hepatocarcinoma was reported to be much higher in offspring than in maternal animals. In contrast, the incidence and latency period of neuroblastomas and hepatic angiosarcomas was similar between offspring and parents. However, no statistics were presented to support these conclusions.

Many of the tumors that were observed in the Drew et al. (1983) and Maltoni et al. (1981) studies were also observed in a study performed by Froment et al. (1994). In this study, Sprague-Dawley pups were exposed to 500 ppm vinyl chloride 8 hours/day, 6 days/week, on postpartum days 3–28. After weaning, 22 animals/gender were exposed for an additional 2 weeks, for a total exposure duration of 33 days. Rats were observed daily until death or development of tumors, and the surviving rats were sacrificed at 19 months. All livers from exposed animals that appeared normal at gross examination were found to contain multiple nodular hyperplastic foci of hepatocytes. Liver tumors that were found in exposed animals included angiosarcomas, hepatocellar carcinomas, and benign cholangiomas. Other tumors found included pulmonary angiosarcoma (probably metastatic), nephroblastoma, abdominal angiomyoma, leukemia, Zymbal gland carcinoma, pituitary adenoma, mammary carcinoma, and mammary fibroma. Tumor incidence was not reported in control animals. Only one concentration (500 ppm) of vinyl chloride was used because the purpose of the study was to examine the genotoxicity of vinyl chloride in liver tumors produced by exposure.

In general, the available evidence from inhalation studies in animals supports the finding in humans; that vinyl chloride is a carcinogen by this route of exposure. Based on these and other findings, the National Toxicology Program of the Department of Health and Human Services has determined vinyl chloride to be a known human carcinogen (DHHS 2002). In addition, IARC has concluded that sufficient evidence for carcinogenicity in humans and animals exists and has placed vinyl chloride in carcinogenicity category 1 (i.e., carcinogenic to humans) (IARC 1987). EPA also has concluded that sufficient evidence of carcinogenicity exists in humans and animals and has classified vinyl chloride according to its 1986 classification scheme as a Group A or known human carcinogen (EPA 1994c). EPA's current weight-of-evidence characterization for vinyl chloride concludes that vinyl chloride is a *known human carcinogen by the inhalation route of exposure*, based on human epidemiological data. By analogy, vinyl chloride is *carcinogenic by the oral route* because of positive animal bioassay data as well as pharmacokinetic data

allowing dose extrapolation across routes. Vinyl chloride is also considered highly *likely to be carcinogenic by the dermal route* because it is well absorbed and acts systemically (EPA 2000); however, animal data suggest that dermal absorption of vinyl chloride gas is not likely to be significant (Hefner et al. 1975a). Because the epidemiological evidence does not provide sufficient exposure and incidence data to quantify risk based solely on human data, EPA cancer potency factors for inhalation and oral exposure have been calculated based on animal studies. An inhalation unit risk of $8.8x10^{-6}$ per ug/m^3 for continuous lifetime exposure from birth was estimated by EPA (2000) based on the incidence of liver tumors observed in rats in the inhalation study by Maltoni et al. (1981). An inhalation unit risk of $4.4x10^{-6}$ per ug/m^3 for continuous lifetime exposure during adulthood was also estimated by EPA (2000). Air concentrations associated with excess cancer risks of 10^{-4}, 10^{-5}, 10^{-6}, and 10^{-7} are $9.0x10^{-3}$, $9.0x10^{-4}$, $9.0x10^{-5}$, and $9.0x10^{-6}$ ppm, respectively, and are plotted in Figure 3-1. These risks were calculated using physiologically based pharmacokinetic (PBPK) modeling, which is discussed in further detail in Section 3.4. The lowest concentrations tested that produced a tumorigenic response CEL for each species and duration category are recorded in Table 3-1 and plotted in Figure 3-1.

3.2.2 Oral Exposure

All dosages of vinyl chloride administered in the diet are reported as mg/kg (body weight)/day unless otherwise specified.

3.2.2.1 Death

No studies were located regarding lethal effects in humans following oral exposure to vinyl chloride.

No studies were located regarding acute or intermediate lethal effects of vinyl chloride in animals. However, decreased longevity has been observed in rats as a result of chronic ingestion of vinyl chloride. Significant increases in mortality were observed by Feron et al. (1981) when Wistar rats were allowed to consume vinyl chloride doses as low as 5.6 mg/kg/day in the diet for 4 hours/day over a 2-year period. Also, the effects of consumption of vinyl chloride during a lifespan study in Wistar rats lasting almost 3 years (149 weeks) were examined by Til et al. (1983, 1991). These authors found a decreased survival rate at a vinyl chloride dosage of 1.7 mg/kg/day. In both of these studies, vinyl chloride was administered by incorporating PVC resin that was high in vinyl chloride content into the diet. In the Til et al. (1991) study, the diets of the control animals contained 1% PVC powder that did not contain residual vinyl

chloride. Vaporization of vinyl chloride from the diets was limited by presenting feed containing the vinyl chloride to the rats for only a 4-hour period.

All reliable LOAEL values for death in rats following chronic exposure are recorded in Table 3-2 and plotted in Figure 3-2.

3.2.2.2 Systemic Effects

The highest NOAEL values and all reliable LOAEL values for hematological, hepatic, dermal, and body weight effects in rats following chronic oral exposure are recorded in Table 3-2 and plotted in Figure 3-2.

No studies were located regarding adverse respiratory, cardiovascular, gastrointestinal, musculoskeletal, renal, endocrine, or ocular effects in humans or animals following oral exposure to vinyl chloride.

Hematological Effects. No studies were located regarding adverse hematological effects in humans after oral exposure to vinyl chloride.

Rats fed 17 mg/kg/day for 2 years showed decreased clotting time of the blood, which was not observed at 5.6 mg/kg/day (Feron et al. 1981). No changes in thrombocyte count or prothrombin times were noted in Wistar rats fed diets containing low concentrations of vinyl chloride in PVC resin (1.7 mg/kg/day) for 149 weeks (Til et al. 1983, 1991).

Hepatic Effects. No studies were located regarding adverse hepatic effects in humans after oral exposure to vinyl chloride.

Chronic exposure of rats to vinyl chloride in their feed for 149 weeks produced an increase in the incidence of several types of microscopic liver lesions in male and female rats. Neoplastic and preneoplastic lesions in the liver included several types of foci of cellular alteration (i.e., clear-cell, basophilic, eosinophilic, or mixed), neoplastic nodules, hepatocellular carcinoma, and angiosarcoma. Other liver lesions associated with vinyl chloride exposure included liver-cell polymorphism and hepatic cysts. The high-dose group in male and female rats (1.7 mg/kg/day) represents a LOAEL for noncancer liver effects in this study (i.e, liver cell polymorphism, hepatic cysts) (Til et al. 1983, 1991). The human equivalent dose derived from a NOAEL of 0.17 mg/kg/day identified in this study was used as the basis

Table 3-2 Levels of Significant Exposure to Vinyl Chloride - 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
CHRONIC EXPOSURE							
Death							
1	Rat (Wistar)	2 yr 5 d/wk 4 hr/d (F)				5.6 (100% died)	Feron et al. 1981
2	Rat (Wistar)	149 wk 4 hr/d (F)				1.7 (increased mortality)	Til et al. 1983, 1991
Systemic							
3	Rat (Wistar)	2 yr 5 d/wk 4 hr/d (F)	Hemato	5.6	17 (decreased clotting time)		Feron et al. 1981
			Hepatic		1.8 (cellular alteration)	17 M extensive necrosis 5.6[b] F (extensive necrosis)	
4	Rat (Wistar)	2 yr 1 x/d (GO)	Dermal		30 (increased skin thickness, collagen)		Knight and Gibbons 1987
5	Rat (Wistar)	149 wk 4 hr/d (F)	Hemato	1.7			Til et al. 1983, 1991
			Hepatic	0.17[c] F	1.7 F (liver cell polymorphism)		
			Bd Wt	1.7			

Table 3-2 Levels of Significant Exposure to Vinyl Chloride - 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
Cancer							
6	Rat (Wistar)	2 yr 5 d/wk 4 hr/d (F), (GO)				5.6 M (CEL: angiosarcoma of lung; neoplastic nodules of liver, hepatic angiosarcoma)	Feron et al. 1981
						1.8[b] F (CEL: neoplastic nodules of liver)	
7	Rat (Sprague-Dawley)	52 wk 5 x/wk (GO)				50 M (CEL: liver angiosarcoma)	Maltoni et al. 1981
						16.65[b] F (CEL: liver angiosarcoma)	
8	Rat (Sprague-Dawley)	52 wk 5 x/wk (GO)				0.3 (CEL: liver angiosarcoma, hepatoma)	Maltoni et al. 1981

Table 3-2 Levels of Significant Exposure to Vinyl Chloride - 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
9	Rat (Wistar)	149 wk 4 hr/d (F)				1.7 M (CEL: hepatocellular carcinoma)	Til et al. 1983, 1991
						1.7 F (CEL: neoplastic nodules of liver)	
						0.018[b] F (CEL: basophilic foci considered to be pre-neoplastic lesions)	
						1.7 (CEL: hepatic angiosarcoma)	

a Numbers correspond to entries in Figure 3-2.

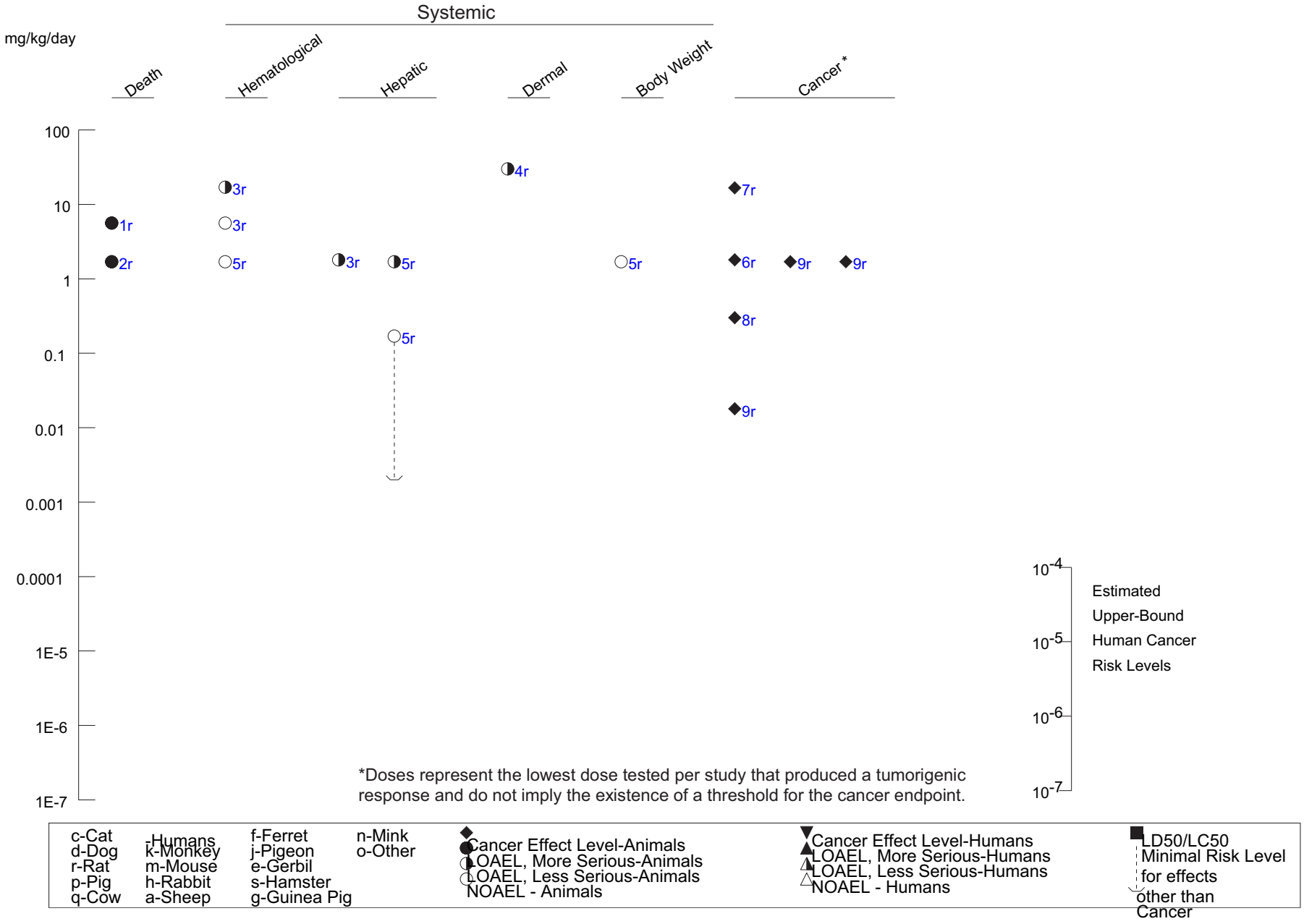
b Differences in levels of health effects and cancer effects between male and females are not indicated in Figure 3-2. Where such differences exist, only the levels of effect for the most sensitive gender are presented.

c Used to derive an chronic-duration Minimal Risk Level (MRL) of 0.002 mg/kg/day; dose divided by an uncertainty factor of 100 (10 for extrapolation from animals to humans and 10 for human variability).

B - both; Bd Wt = body weight; CEL = cancer effect level; d = day(s); derm = dermal; (F) = feed; F = Female; (GO) = gavage in oil; hemato = hematological; hr = hour(s); LOAEL = lowest-observed-adverse-effect level; M = male; NOAEL = no-observed-adverse-effect level; wk = week(s); x = time(s); yr = year(s)

Figure 3-2 Levels of Significant Exposure to Vinyl Chloride - Oral

Chronic (≥365 days)



for a chronic oral MRL of 0.003 mg/kg/day. Chronic oral exposure of rats fed vinyl chloride daily during a 4-hour period for 2 years also resulted in areas of hepatocellular alteration at concentrations as low as 1.8 mg/kg/day (Feron et al. 1981). In this study, areas of necrosis were observed in the liver of female rats fed 5.6 mg/kg/day and male rats fed 17 mg/kg/day (Feron et al. 1981). Increased incidence of hepatic cysts were found in female rats fed 1.7 mg/kg/day and clear or basophilic areas of cellular alteration were found in male rats fed 1.7 mg/kg/day in the Til et al. (1983, 1991) studies.

Dermal Effects. No studies were located regarding adverse dermal effects in humans after oral exposure to vinyl chloride.

Daily administration of 30 mg/kg of vinyl chloride to rats by gavage for 2 years produced increased thickness, moisture content, and collagen content of the skin. Newly synthesized intermolecular and intramolecular collagen crosslinks were also significantly increased (Knight and Gibbons 1987).

Body Weight Effects. No studies were located regarding adverse body weight effects in humans after oral exposure to vinyl chloride.

No changes in body weight were noted in Wistar rats fed 1.7 mg/kg/day vinyl chloride mixed with PVC powder in the diet for 149 weeks (Til et al. 1983, 1991).

No studies were located regarding the following health effects in humans or animals after oral exposure to vinyl chloride:

3.2.2.3 Immunological and Lymphoreticular Effects

3.2.2.4 Neurological Effects

3.2.2.5 Reproductive Effects

3.2.2.6 Developmental Effects

3.2.2.7 Cancer

No studies were located regarding cancer in humans following oral exposure to vinyl chloride.

Four studies were located that examined the carcinogenic potential of vinyl chloride in animals when administered by the oral route. In two of these studies in Wistar rats, conducted for 149 weeks, vinyl chloride was added to the diet by incorporating PVC powder containing a high level of the monomer (Feron et al. 1981; Til et al. 1983, 1991). To limit volatilization of vinyl chloride from the diet, the rats were allowed access to the diet for only 4 hours/day. The actual intake of vinyl chloride in these reports was calculated by taking into consideration both the food consumption and the rate of vinyl chloride evaporation. Statistically significant increases in hepatic angiosarcoma of the liver were observed in the 2-year study by Feron et al. (1981) at 5.6 mg/kg/day in males and 17 mg/kg/day in females. In the same study, statistically significant increases in neoplastic nodules of the liver were also observed at a concentration of 5.6 mg/kg/day in males but as low as 1.8 mg/kg/day in females (Feron et al. 1981). Also, in the 149-week study by Til et al. (1983, 1991), statistically significant increases in hepatocellular carcinoma were observed in males at 1.7 mg/kg/day and hepatic neoplastic nodules in females at 1.7 mg/kg/day. A few animals exposed to 1.7 mg/kg/day in this study developed hepatic angiosarcoma. An increased incidence of Zymbal gland tumors was also observed in the study by Feron et al. (1981). Although the increase was not statistically significant, the tumors were considered to be treatment related based on the historical rarity of this type of tumor.

Two studies were located in which vinyl chloride was administered to Sprague-Dawley rats by gavage for 52 weeks. In one of these studies, a statistically significant increase in the incidence of hepatic angiosarcomas was observed at doses as low as 16.65 mg/kg/day in females and 50 mg/kg/day in males. Zymbal gland tumors at 16.65 and 50 mg/kg/day, even though not statistically significant, were considered to be treatment related because of the rarity of this type of tumor (Maltoni et al. 1981). Lower doses of vinyl chloride were also tested in a similar study in which hepatic angiosarcomas were observed at doses as low as 0.3 mg/kg/day and Zymbal gland tumors at 1 mg/kg/day. Although neither of these findings reached statistical significance, the tumors were considered to be treatment related because of the historically rare observation of these tumor types in the colony (Maltoni et al. 1981).

Based on the evidence of carcinogenicity in animals after oral exposure, it would be prudent to consider the potential for carcinogenic effects in humans by this route as well. The National Toxicology Program of the Department of Health and Human Services has determined vinyl chloride to be a known human carcinogen (DHHS 2002). In addition, IARC has concluded that sufficient evidence for carcinogenicity in humans and animals exists and has placed vinyl chloride in carcinogenicity category 1 (i.e., carcinogenic to humans) (IARC 1987). EPA also has concluded that sufficient evidence of carcinogenicity exists in humans and animals and has classified vinyl chloride according to its 1986

classification scheme as a Group A or known human carcinogen (EPA 1994c). EPA's current weight-of-evidence characterization for vinyl chloride concludes that vinyl chloride is a *known human carcinogen by the inhalation route of exposure*, based on human epidemiological data. By analogy, vinyl chloride is considered *carcinogenic by the oral route* because of positive animal bioassay data as well as pharmacokinetic data allowing dose extrapolation across routes. By inference, vinyl chloride is also considered highly *likely to be carcinogenic by the dermal route* because it acts systemically (EPA 2000). Because the epidemiological evidence does not provide sufficient exposure and incidence data to quantify risk based solely on human data, EPA cancer potency factors for inhalation and oral exposure have been calculated based on animal studies. An oral slope factor for continuous lifetime exposure from birth was estimated by EPA (2000) to be 1.5 per mg/kg/day based on the incidence of liver tumors in rats in the study by Feron et al. (1981). An oral slope factor of 7.5×10^{-1} per mg/kg/day for continuous lifetime exposure during adulthood was also estimated by EPA (2000). Oral doses associated with excess cancer risks of 10^{-4}, 10^{-5}, and 10^{-6} are 1.33×10^{-4}, 1.33×10^{-5}, 1.33×10^{-6}, and 1.33×10^{-7} mg/kg/day, respectively, and are plotted in Figure 3-2. These risks were calculated using PBPK modeling, which is discussed in further detail in Section 3.4. The lowest doses tested that produced a tumorigenic response (CEL) in rats chronically exposed to vinyl chloride by the oral route are recorded in Table 3-2 and plotted in Figure 3-2.

3.2.3 Dermal Exposure

Dermal exposure to vinyl chloride may occur by skin contact with either gaseous or liquid vinyl chloride. Negligible amounts of gaseous vinyl chloride are absorbed through the skin (see also Section 3.4 regarding absorption by the dermal route). However, dermal exposure can also occur by direct contact of gaseous vinyl chloride with the eyes. Only studies that specifically relate to dermal contact of liquid vinyl chloride or adverse ocular effects occurring with inhalation exposure to gaseous vinyl chloride are discussed below.

3.2.3.1 Death

No studies were located regarding lethal effects in humans or animals after dermal exposure to vinyl chloride.

3.2.3.2 Systemic Effects

No studies were located regarding adverse respiratory, cardiovascular, gastrointestinal, hematological, musculoskeletal, hepatic, renal, or endocrine effects in humans or animals after dermal exposure to vinyl chloride.

Dermal Effects. Vinyl chloride exists as a liquid when stored under pressure. However, when it is released from pressurized containers, it rapidly vaporizes into gas. Thus, the adverse dermal effects observed after exposure to vinyl chloride are not unique to vinyl chloride but can be expected as a result of a rapidly evaporating liquid on the skin. The effects are due to tissue freezing rather than direct toxicity of vinyl chloride. A man who had liquid vinyl chloride sprayed on his hands developed second degree burns. At first, the man reported that his hands felt numb. Within a short period, the hands had developed marked erythema and edema (Harris 1953).

No studies were located regarding adverse dermal effects in animals after dermal exposure to vinyl chloride.

Ocular Effects. Local burns on the conjunctiva and cornea were observed in a man who died after exposure to an unknown quantity of vinyl chloride escaping from an open valve (Danziger 1960).

No adverse ocular effects were noted in guinea pigs exposed for 30 minutes to up to 400,000 ppm vinyl chloride in inhalation chambers (Mastromatteo et al. 1960).

3.2.3.3 Immunological and Lymphoreticular Effects

No studies were located regarding adverse immunological and lymphoreticular effects in humans or animals following dermal exposure to vinyl chloride.

3.2.3.4 Neurological Effects

A man who had liquid vinyl chloride sprayed on his hands initially reported that his hands felt numb (Harris 1953).

No studies were located regarding adverse neurological effects in animals after dermal exposure to vinyl chloride.

No studies were located regarding the following adverse health effects in humans or animals after dermal exposure to vinyl chloride:

3.2.3.5 Reproductive Effects

3.2.3.6 Developmental Effects

3.2.3.7 Cancer

3.3 GENOTOXICITY

Vinyl chloride has been shown to be mutagenic and clastogenic in both *in vivo* and *in vitro* test systems. Tables 3-3 and 3-4 list the key *in vivo* and *in vitro* genotoxicity studies for vinyl chloride.

Genotoxicity studies of vinyl chloride in humans include a large number of assays for chromosomal aberrations in the cultured lymphocytes of occupationally exposed workers. Studies completed through the mid-1980s generally found a statistically significant increase in the frequency of chromosomal aberrations, usually of the chromatid type (i.e., affecting only one of the two strands formed upon deoxyribonucleic acid [DNA] replication), but also including some chromosomal-type defects such as inversions, rings, and translocations, which affect the entire chromosome (Anderson et al. 1981; Anderson 1999, 2000; Fleig et al. 1978; Fucic et al. 1990; Heath et al. 1977). An increase in chromosomal aberrations was also observed following an accidental environmental exposure to vinyl chloride (Becker et al. 2001; Huttner and Nikolova 1998; Huttner et al. 1998, 1999). Workers exposed to vinyl chloride for an average of 15 years were shown to have elevated levels of micronuclei and chromosomal aberrations when compared to the unexposed controls (Garaj-Vrhovac et al. 1990). An increase in chromosome aberrations and micronuclei was correlated with the air concentration of vinyl chloride at a plastics plant and the excretion of thiodyglycolic acid in the urine of exposed workers (Vaglenov et al. 1999). Micronuclei counts were also increased in a group of 52 workers exposed to vinyl chloride levels of 1.3–16.7 ppm compared to those of controls, but these increases were not observed in workers exposed to 0.3–7.3 ppm (Sinues et al. 1991). Micronuclei were also increased in lymphocytes from 19 workers exposed to 50 ppm vinyl chloride for approximately 15 years (Fucic et al. 1990).

Table 3-3. Genotoxicity of Vinyl Chloride *In Vivo*

Species (test system)	End point	Results	Reference
Mouse	Dominant lethal	–	Anderson et al. 1976
	Micronuclei	+	Richardson et al. 1983
Rat	Dominant lethal	–	Short et al. 1977
		–	Anderson et al. 1976
		–	Purchase et al. 1975
	Chromosomal aberration	+	Anderson and Richardson 1981
Hamster	Chromosomal aberration	+	Fleig et al. 1978
Human lymphocyte	Sister chromatid exchange	–	Hansteen et al. 1978
		+	Kucerova et al. 1979
		+	Sinués et al. 1991
		+	Fucic et al. 1990a
		+	Fucic et al. 1992
		+	Fucic et al. 1995
		+	Fucic et al. 1996a
		+	Fucic et al. 1996b
		+	Zhao et al. 1994
	DNA damage	+	Awara et al. 1998
		+	Du et al. 1995
	Micronuclei	+	Fucic et al. 1990a
		+	Garaj-Vrhovac et al. 1990
		+	Sinués et al. 1991
		+	Vaglenov et al. 1999
	Chromosomal aberration	+	Hansteen et al. 1978
		+	Heath et al. 1977
		+	Kucerova et al. 1979
		–	Picciano et al. 1977
		+	Purchase et al. 1978
		+	Ducatman et al. 1975
		+	Anderson 1980, 1981
		+	Fleig et al. 1978
		+	Fucic et al. 1990a, 1990b
		+	Fucic et al. 1995
		+	Fucic et al. 1996a

Table 3-3. Genotoxicity of Vinyl Chloride *In Vivo*

Species (test system)	End point	Results	Reference
		+	Fucic et al. 1996b
Human lymphocyte *(cont.)*	Chromosomal aberration	+	Funes-Cravioto et al. 1975
		+	Hrivnak et al. 1990
		+	Garaj-Vrhovac et al. 1990
		+	Anderson, 1999
		+	Becker et al. 2001
		+	Huttner et al. 1998
		+	Huttner et al. 1999
		+	Huttner and Nikolova 1998
		+	Fucic et al. 1992
		+	Vaglenov et al. 1999
Rat	DNA alkylation	+	Laib et al. 1989
		+	Green and Hathway 1978
		+	Gwinner et al. 1983
		+	Singer et al. 1987
		+	Bolt et al. 1986
		+	Ciroussel et al. 1990
		+	Eberle et al. 1989
Mouse	DNA alkylation	+	Osterman-Golkar et al. 1977
	DNA damage	+	Walles et al. 1988
Rat	DNA adduct	+	Fedtke et al. 1990
		+	Ciroussel et al. 1990
		+	Swenberg et al. 1992
		+	Bolt et al. 1986
		+	Morinello et al. 2002a, 2002b
		+	Eberle et al. 1989

– = negative result; + = positive result; DNA = deoxyribonucleic acid

Table 3-4. Genotoxicity of Vinyl Chloride *In Vitro*

Species (test system)	End point	Result		Reference
		With activation	Without activation	
Salmonella typhimurium	Reverse mutation	+	–	Rannug et al. 1974
		+	+	Bartsch et al. 1975, 1976
		+	+	Andrews et al. 1976
		+	+	Simmon et al. 1977
		Not tested	–	Elmore et al. 1976
		+	+	Poncelet et al. 1980
		+	+	De Meester et al. 1980
		+	+	Victorin and Stahlberg 1988a
		+	Not tested	McCann et al. 1975
		+	+	Rannug et al. 1976
TA100, TA1535	Base-pair substitution	+	+	duPont 1992a, 1992b
		+	Not tested	Malaveille et al. 1975
TA98, TA1537, TA1538	Frameshift mutation	–	–	
Escherichia coli		Not applicable	+	Jacobsen et al. 1989
Saccharomyces cerevisiae		Not tested	–	Shahin 1976
	Gene conversion	+	Not tested	Loprieno et al. 1976
Schizosaccharomyces pombe	Forward mutation	+	–	Loprieno et al. 1977
		+	Not tested	Loprieno et al. 1976
D7RAD yeast	Gene conversion	+	–	Ekardt et al. 1981
Chinese hamster ovary cells		Not applicable	+	Huberman et al. 1975
		+	Not tested	Drevon et al. 1978
		+	–	duPont 1992c
Bacillus subtilis	Rec-repair	Not tested	–	Elmore et al. 1976
Rat liver microsomes	RNA alkylation	Not applicable	+	Laib and Bolt 1977
QT6 (avian cells)	Inhibition of DNA synthesis	Not applicable	+	Kandala et al. 1990

– = negative result; + = positive result; DNA = deoxyribonucleic acid; RNA = ribonucleic acid

Increased sister chromatid exchanges have also been reported in occupationally exposed workers (Fucic et al. 1990, 1992, 1995; Kucerova et al. 1979; Sinues et al. 1991; Zhao et al. 1996). Sister chromatid exchange frequencies were significantly increased compared to those of the controls at 0.003–7.3 ppm vinyl chloride (Sinues et al. 1991). A positive correlation between frequency of chromosomal aberrations and length of exposure and history of exposure to excursion levels (up to 2,000 ppm) was reported by Purchase et al. (1978), who examined a cohort of 57 vinyl chloride workers, 19 on-site controls, and five off-site controls. The exposures for this cohort ranged from 1,000 ppm between 1945 and 1955 to 5 ppm since 1975. These authors also reported an effect on chromosomal aberrations from smoking. Smoking and the presence of an aldehyde dehydrogenase 2 genotype was associated with an increase in the frequency of sister chromatid exchange in vinyl chloride workers (Wong et al. 1998). DNA damage in lymphocytes of plastic industry workers was also demonstrated by a single-cell gel electrophoresis technique. A correlation was observed between the severity of DNA damage and the duration of exposure (Awara et al. 1998). DNA single strand breaks present in human lymphocytes from exposed workers were quickly repaired following cessation of exposure (Du et al. 1995). Induction of single-strand breaks in liver DNA was also observed in mice after inhalation of vinyl chloride (Walles et al. 1988).

The reversibility of chromosome damage has been reported for several populations of workers following a cessation or reduction of exposure to vinyl chloride. The increase of chromosome aberrations observed in workers exposed to 50 ppm returned to normal within 42 months after exposure levels had been reduced to <5 ppm (Anderson et al. 1980). Another study demonstrated a statistically significant increase in aberrations in workers exposed to concentrations of approximately 25 ppm. Following a reduction in exposure to 1 ppm, vinyl chloride chromosomal aberrations had returned to control values (Hansteen et al. 1978). A 9-year follow-up study of an occupationally exposed population demonstrated a decrease in chromosome aberrations and sister chromatid exchange frequencies over time, corresponding to a decrease in vinyl chloride air concentrations at the plant (Fucic et al. 1996a, 1996b).

The reversibility of clastogenic effects was not seen in another study of 12 current and 3 retired plastics industry workers who had been exposed to vinyl chloride while employed for 1.5–35 years (Fucic et al. 1992). Sister chromatid exchange frequencies were significantly higher in workers exposed to concentrations up to 2,000 ppm than in the controls. These findings showed no significant decrease in sister chromatid exchange frequencies from 8 days to 10 years after exposure (Fucic et al. 1992).

Other papers on human subjects have focused on specific mechanisms involved in the clastogenic effects of vinyl chloride. A cohort of 67 workers exposed to approximately 5 ppm for an average of 15 years was reported to have a nonrandom distribution of chromatid and bichromatid breaks (Fucic et al. 1990b). The most frequently affected areas of the genome were the terminal segments of the A, B, and C group chromosomes, suggesting that vinyl chloride or its metabolites interact more frequently with specific sites along the chromosome than would be expected. The study authors presented no correlation with particular fragile sites (gene sequences more prone to breakage than normal) or oncogene locations known to occur at these terminal segments. The implication is that the carcinogenicity of vinyl chloride could be at least partially explained by its nonrandom interaction with particular genes. These workers were also periodically exposed to 2,000 ppm for short periods. No specific information was given as to the frequency or duration of these events.

Male workers (n=20) who had been employed for 2–14 years at a vinyl chloride polymerization plant exposed to concentrations of vinyl chloride of 1 ppm (with occasional peaks of 300 ppm) underwent cytogenetic testing (Fucic et al. 1995). The test results were compared to those from 20 unexposed control men. Exposed individuals had higher percentages of chromosome aberrations, primarily chromatid breaks. Sister chromatid exchange frequencies were also increased in exposed workers (4–22 per cell) compared to controls (4–7 per cell). Significant changes in mitotic activity were noted among exposed workers; values for second mitosis were lower than controls and values for third mitosis were higher than controls (Fucic et al. 1995, 1997). Chromosome aberrations were not increased in workers exposed to <5 ppm vinyl chloride; however, the average exposure duration for this study was <1 year (Picciano et al. 1977).

Genetic polymorphisms of metabolic and DNA repair genes have been associated with the sister chromatid exchange frequency in exposed workers (Wong et al. 2003b). Metabolic genotypes for CYP2E1, aldehyde dehydrogenase 2 (ALDH2) and the DNA repair genotype for x-ray repair cross complementing group1 (XRCC1) were associated with an increased risk of DNA damage in humans.

Animal studies of rats and mice exposed via inhalation to vinyl chloride have concentrated on identifying the direct effects of vinyl chloride and its metabolites on DNA. Vinyl chloride is metabolized by mixed function oxidases (MFO) to form an epoxide intermediate, 2-chloroethylene oxide, which spontaneously rearranges to form 2-chloroacetaldehyde. Reactive metabolites of vinyl chloride can be transported intercellularly from parenchymal cells to the nonparenchymal cells (Kuchenmeister et al. 1996). Many studies have characterized the mutation profile associated with DNA adducts formed by the reactive

metabolites of vinyl chloride (Akasaka et al. 1997; Chiang et al. 1997; Dosanjh et al. 1994; Guichard et al. 1996; Matsuda et al. 1995; Pandya and Moriya 1996; Zhang et al. 1995; Zielinski and Hergenhahn 2000;). Four primary mutagenic DNA adducts are formed by the reactive metabolites of vinyl chloride. These are cyclic etheno-adducts that include 1,N^6-ethenoadenine, 3,N^4-ethenocytosine, N^2,3-etheno-guanine, and 1,N^2-ethenoguanine. These adducts can induce base-pair (i.e., purine-to-purine or pyrimidine-to-pyrimidine exchange) transitions during transcription (Cullinan et al. 1997; Oesch and Doerjer, 1982; Pandya and Moriya 1996; Singer et al. 1987, 1996). 1,N^6-Ethenoadenine adducts have been demonstrated to trap topoisomerase I, affecting DNA replication and transcription (Pourquier et al. 1998). DNA crosslinks can also be formed because chloracetaldehyde is bifunctional (Singer 1994). The adduct 7-(2'-oxoethyl)guanine is also extensively formed in mammalian liver (Laib et al. 1981); however, it is quickly recognized and removed by DNA repair mechanisms. Etheno-adducts are less abundant, but more persistent because they are poorly repaired (Brandt-Rauf et al. 2000b; Whysner et al. 1996).

The identification of the etheno-nucleosides has been reported following inhalation exposure to vinyl chloride in rats (Bolt et al. 1986; Ciroussel et al. 1990; Eberle et al. 1989; Fedtke et al. 1990; Morinello et al. 2002a, 2002b; Swenberg et al. 1992). Immature rats exposed *in vivo* formed 6 times more of this nucleoside adduct, which correlated with the age-related sensitivity to carcinogenesis in these animals (Ciroussel et al. 1990). This age-related sensitivity to DNA adduct formation was also noted in an inhalation study of lactating rats and their 10-day-old pups exposed 4 hours/day, for 5 days to 600 ppm of vinyl chloride (Fedtke et al. 1990). Concentrations of two adducts found in the liver of the pups were 4-fold higher than those found in the liver of the dams. Increased alkylation of liver DNA and increased cell proliferation were reported by Laib et al. (1989) following exposure to 600 ppm vinyl chloride for 6 hours. Young rats were apparently more susceptible to the effects of vinyl chloride, but only three male adults and two female adults were used for comparison. The concentration of ethenoguanine adducts was 2–3-fold greater in weanling rats as compared to adult rats exposed at the same dose for the time period (0, 10, 100, or 1,100 ppm, 6 hours/day for 5 days) (Morinello et al. 2002a). Rats exposed to 2,000 ppm vinyl chloride for 8 hours/day, 5 days/week, for 3 weeks beginning at 7 days of age demonstrated hepatocellular ATPase-deficient foci and alkylation of liver DNA (Gwinner et al. 1983). A study in rats exposed to 1,100 ppm vinyl chloride for 6 hours/day, 5 days/week for 1 or 4 weeks demonstrated that ethenoguanine adducts are not formed in the adult rat brain (Morinello et al. 2002b). This differential induction of DNA adducts (brain vs. liver) may relate to the direct effect of reactive intermediates at the site of metabolite generation.

The role of etheno-adducts in the carcinogenesis of vinyl chloride has been recently reviewed (Albertini et al. 2003; Barbin 1998, 1999, 2000; Kielhorn et al. 2000; Laib 1986; Nivard and Vogel 1999; Whysner et al. 1996). Both 2-chloroethylene oxide and 2-chloroacetaldehyde can react with DNA nucleotide bases; however, 2-chloroethylene oxide is a more potent mutagen and may be the ultimate carcinogenic metabolite of vinyl chloride (Chiang et al. 1997). Etheno-adducts generate mainly base pair substitution mutations. Mutations in specific genes (i.e., *ras* oncogenes, p53 tumor suppressor gene) have been identified in vinyl chloride-induced liver tumors in rats and humans and are discussed in further detail below. Exocylic DNA adducts are excised from the DNA by glycosylase enzymes that contribute to genetic stability (Laval and Saparbaev 2001). The four primary cyclic adducts formed in DNA by the vinyl chloride metabolite chloroacetaldehyde are released by human glycosylase enzymes (Dosanjh et al. 1994; Singer and Hang 1999). The expression of the DNA repair enzyme N-methylpurine-DNA-glycosylase was shown to be deficient in nonparenchymal cells of the rat liver, which are the target cells for vinyl chloride-induced angiosarcoma (Holt et al. 2000; Swenberg et al. 1999). However, there was no difference observed in the formation of ethenoguanine adducts in hepatocytes and nonparenchymal cells immediately following vinyl chloride exposure (Morinello et al. 2002a). Together, these data suggest that cellular differences in DNA repair capacity may play a role in vinyl chloride-induced carcinogenesis. It is important to note that endogenously formed etheno-adducts are also present in humans and laboratory animals due to a reaction between DNA and lipid peroxidation by-products. This background incidence of etheno-adducts should be taken into account when evaluating exposure to chemicals like vinyl chloride (Albertini et al. 2003; Bartsch and Nair 2000; Gonzalez-Reche et al. 2002; Swenberg et al. 2000; Watson et al. 1999; Yang et al. 2000; Zielinski and Hergenhahn 2001).

It has been suggested that members of the *ras* gene family, including Ha-*ras*, Ki-*ras*, and N-*ras*, are responsible for the control of cell proliferation and differentiation (Froment et al. 1994). DNA adducts formed by vinyl chloride metabolites can produce point mutations in these genes. Mutations of the Ki-*ras*-2 gene has been found in hepatic angiosarcomas of workers exposed to high levels of vinyl chloride; this specific gene was shown to be activated by a GC-AT transition at codons 12 and 13 (Brandt-Rauf et al. 1995; Marion et al. 1991; Weihrauch et al. 2002). Similar mutations of Ki-*ras*-2 have been found in hepatocellular carcinomas of workers exposed to vinyl chloride (Weihrauch et al. 2001a, 2001b). Hypermethylation of the p16 gene was also associated with Ki-*ras*-2 mutation in hepatocellular carcinomas from exposed workers (Weinhrauch 2001b).

Mutation of the Ki-*ras*-2 gene results in the expression of a mutant p21 protein. This mutant oncoprotein was detected in serum samples taken from vinyl chloride workers with angiosarcoma of the liver (DeVivo

et al. 1994; Marion 1998). Mutant p21 protein was also detected in the serum or plasma of exposed workers without liver tumors and a relationship between the frequency of the mutant protein in serum and the intensity of vinyl chloride exposure was demonstrated in several studies (Brandt-Rauf et al. 1995; DeVivo et al. 1994; Li et al. 1998; Luo et al. 1998, 2003; Marion 1998).

Rat liver tumors induced by exposure to 500 ppm vinyl chloride were examined for mutations of the Ha-*ras*, Ki-*ras*, and N-*ras* genes (Boivin-Angele et al. 2000; Froment et al. 1994; Marion and Boivin-Angele 1999). In contrast with the studies in humans, Ki-*ras* gene mutation does not occur in rats or mice with angiosarcoma of the liver induced by vinyl chloride exposure. Rats with hepatocellular carcinoma demonstrated a AT–TA transversion of base 2 of codon 61 of the Ha-*ras* gene. This mutation was not detected in rodent angiosarcoma of the liver suggesting that there might be cell-specific factors that affect the *ras* gene. Other mutations in codons 13 and 36 of the N-*ras* A gene were found in two out of five of the liver angiosarcomas examined (Froment et al. 1994). These studies suggest differing molecular mechanisms of carcinogenesis in humans and rodents.

The p53 tumor suppressor gene is mutated in a variety of human cancers (Staib et al. 2003; Trivers et al. 1995). A study was performed to examine the p53 tumor suppressor genes and the murine double min-2 (MDM2) proto-oncogenes from tumors of five vinyl chloride workers; four with angiosarcoma of the liver and one with hepatocellular carcinoma (Hollstein et al. 1994). The p53 tumor suppressor gene was being tested for mutation, while the MDM2 proto-oncogene was being tested for amplification. No amplification of the MDM2 gene was detected; however, adenosine-to-thymidine missense mutations were found in exons 5–8 (codons 249 and 255) of the p53 gene in two of the angiosarcoma cases. In another study, tumors (angiosarcoma of the liver) from three of six vinyl chloride workers also had adenosine-to-thymidine missense mutations in the p53 gene (codons 249, 255, and 179) (Trivers et al. 1995). Data from a study of angiosarcoma of the liver resulting from endogenous or unknown sources (i.e., no vinyl chloride exposure) indicated that p53 mutations were uncommon, providing support for the specificity of p53 mutations with vinyl chloride exposure in cases of angiosarcoma of the liver (Soini et al. 1995). The p53 gene mutation pattern in rat liver tumors (angiosarcoma and hepatocellular carcinoma) was shown to be similar to that observed in human tumors from vinyl chloride-exposed workers (Barbin et al. 1997; Marion and Boivin-Angele 1999). Mutations of the p53 gene were found in hepatocellular carcinomas from workers exposed to vinyl chloride; however, no correlation with vinyl chloride exposure occurred and the mutation pattern was thought to reflect endogenous mechanisms rather that chemical mutagenesis (Weihrauch et al. 2000). A p53 mutation at codon 179 was detected in myofibroblast-type cells isolated from a liver tumor in an exposed worker (Boivin et al. 1997). Ki-*ras* mutations were not

observed in these cells. Vinyl chloride mutations of the p53 gene produce conformational effects in the expressed p53 protein that affect its function (Chen et al. 1999).

Mutant p53 protein and/or anti-p53 antibodies have been detected in the serum and plasma of vinyl chloride-exposed workers (Luo et al. 1999; Marion 1998; Smith et al. 1998; Trivers et al. 1995). A relationship between the frequency of the mutant protein or p53 antibodies in serum/plasma and the intensity of vinyl chloride exposure was demonstrated in these studies. Polymorphisms of the genes for vinyl chloride metabolism (CYP2E1) and DNA repair (x-ray cross-complementing group 1) are associated with a greater risk of p53 gene mutation and over-expression of p53 mutant protein (Li et al. 2003; Wong et al. 2002b).

Rat studies suggest that gap junctional intercellular communication mediated by connexin 37 is disturbed in angiosarcoma of the liver; however, mutation of the connexin 37 gene is considered rare (Saito et al. 1997). The incidence of hypoxanthine-guanine-phosphoribosyl-transferase (HPRT) mutants was not consistently elevated in workers exposed to vinyl chloride (Huttner and Holzapfel 1996; Liber et al. 1999). HPRT mutants were also not increased in humans accidentally exposed to vinyl chloride (Becker et al. 2001).

Vinyl chloride has not been shown to be positive for dominant lethal effects in rats exposed to up to 30,000 ppm, for 6 hours/day for 5 days (Anderson et al. 1976; Purchase et al. 1975; Short et al. 1977). The studies showed no evidence of pre- or postimplantation loss among the untreated females mated to the exposed males. These results indicate that no germinal mutations were produced by these acute exposures. Vinyl chloride induces somatic and sex-linked recessive lethal mutations in *Drosophila*, but does not induce dominant lethal mutations (Ballering et al. 1996; Giri 1995; Magnusson and Ramel et al. 1978).

Vinyl chloride is mutagenic in *S. typhimurium* (Andrews et al. 1976; Bartsch et al. 1975, 1976; de Meester et al. 1980; Elmore et al. 1976; Malaveille et al. 1975; Poncelet et al. 1980; Simmon et al. 1977), but only in strains reverted by base-pair substitution by alkylating agents rather than by frameshift mutations (Bartsch et al. 1976; duPont 1992a, 1992b). Metabolic activation is necessary for any mutagenic activity in this system (Rannug et al. 1974) or for a maximal response (Simmon et al. 1977). In addition, vinyl chloride is mutagenic in the gaseous phase, but not when it is dissolved in water (Poncelet et al. 1980). The negative findings for vinyl chloride dissolved in water are most likely due to

methodological problems associated with rapid evaporation and therefore do not reflect a lack of mutagenic potential.

Concentrations of vinyl chloride tested *in vitro* range from 0.275% (Shahin 1976) to 40% (duPont 1992a). Shahin (1976) reported negative results for 0.275 and 0.55% vinyl chloride in *Saccharomyces cerevisiae*. In *S. typhimurium*, a doubling of revertants has been reported to occur at about 5% vinyl chloride (Victorin and Stahlberg 1988a). Vinyl chloride was found to be mutagenic in Chinese hamster ovary cells and yeast (Drevon et al. 1978; duPont 1992c; Eckardt et al. 1981; Loprieno et al. 1976). A 5-hour exposure to 4,600 ppm vinyl chloride did not cause mutatgenicity in the mammalian spot test (Peter and Ungvary 1980). Workers exposed to vinyl chloride have been shown to have increased chromosomal aberrations, micronucleic counts, and sister chromatid exchange frequencies (Anderson et al. 1980; Fucic et al. 1992, 1995, 1997; Garaj-Vrhovac et al. 1990; Kucerova et al. 1979; Sinues et al. 1991; Zhao et al. 1996).

There is evidence that in *S. typhimurium*, *E. coli*, and *B. subtilis,* it is the oxidation of vinyl chloride to the reactive intermediates 2-chloroethylene oxide and 2-chloroacetaldehyde that is responsible for the mutagenicity of vinyl chloride (Bartsch et al. 1976, 1979; Hussain and Osterman-Golkar, 1976; Jacobsen et al. 1989; Laumbach et al. 1977; McCann et al. 1975; Rannug et al. 1976). The S-9 fraction from surgically obtained human liver specimens was shown to metabolize vinyl chloride to electrophiles that were mutagenic to *S. typhimurium* TA1530 (Sabadie et al. 1980). Mutagenicity assays were performed by exposing the plates containing *S. typhimurium* and 150 µL human S-9 fraction to a gaseous mixture of 20% vinyl chloride in air for 4 hours. Vinyl chloride was removed after the exposure. The vinyl chloride concentration in the aqueous phase of the plates was $4x10^{-3}$ M. Incubation was continued for an additional 48 hours. When compared with the number of revertants per plate resulting from identically prepared S-9 fractions from female strain BD IV rats, human S-9 fractions induced mutations (and presumably metabolism to a reactive electrophile) to an average 84% of the extent mediated by rat S-9. However, a 9-fold individual variation was observed.

Chloroacetaldehyde appears to be less genotoxic in yeast and Chinese hamster V79 cells than 2-chloroethylene oxide (Huberman et al. 1975; Loprieno et al. 1977) and has been shown to inhibit DNA synthesis in avian cells (Kandala et al. 1990). However, 2-chloroacetaldehyde has been shown to react directly with single-stranded DNA, which then produced base changes and subsequent reversion in *E. coli* when the DNA was inserted via phage (Jacobsen et al. 1989). Recent data have also shown

2-chloroacetaldehyde to be mutagenic in human fibroblast cells using shuttle vectors (Matsuda et al. 1995).

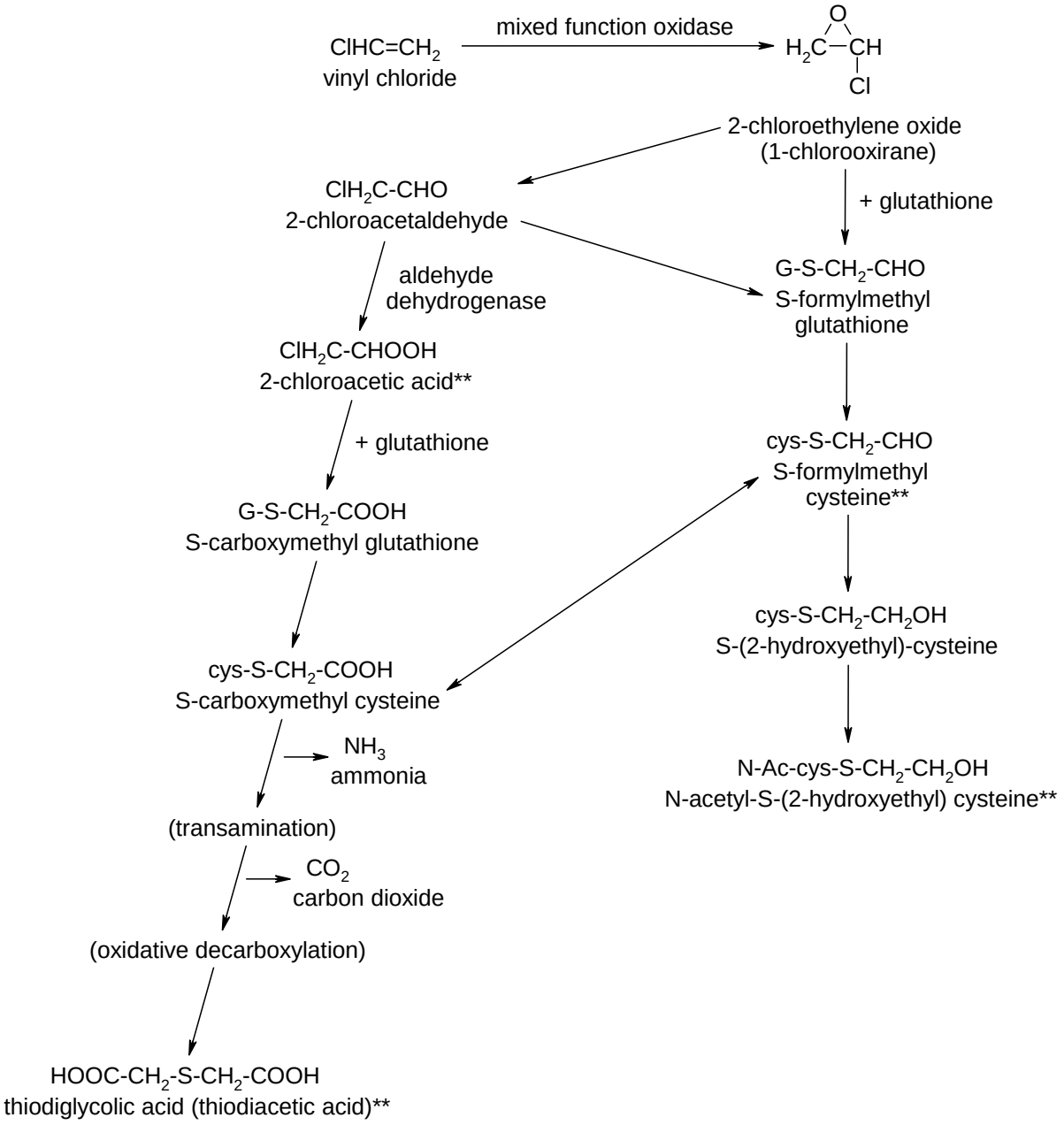
3.4 TOXICOKINETICS

Vinyl chloride is volatile and exposure occurs largely by inhalation. Studies in humans and animals have shown that vinyl chloride is readily absorbed through the lungs (Krajewski et al. 1980; Withey, 1976). Animal studies demonstrate that vinyl chloride is rapidly and almost completely absorbed from the gastrointestinal tract after oral exposure (Watanabe et al. 1976a; Withey 1976). A single study in monkeys, suggests that dermal absorption of vinyl chloride gas is not likely to be significant (Hefner et al. 1975a). No studies were located that reported the absorption of vinyl chloride in humans after oral or dermal exposure.

Animal studies indicate that the distribution of vinyl chloride is rapid and widespread; however, storage in the body is limited because of rapid metabolism and excretion. Metabolites of vinyl chloride have been found in the liver, kidney, spleen, skin, and brain, but tissue concentrations do not increase following repeated exposure (Bolt et al. 1976a; Butcher et al. 1977; Duprat et al., 1977; Watanabe 1978a, 1976b). Vinyl chloride has been shown to cross the placenta after inhalation exposure (Ungvary et al. 1978). No studies were located that reported tissue distribution after inhalation, oral, or dermal exposure to vinyl chloride in humans or after dermal exposure in animals. Vinyl chloride distribution may be affected by differences in gender, age, and nutritional status.

Vinyl chloride metabolism in humans is attributed to the cytochrome P-450 monooxygenases in the liver (Ivanetich et al. 1977; Sabadie et al. 1980; Salmon 1976). The proposed metabolic pathways for vinyl chloride are shown in Figure 3-3. Data obtained in rats indicate that metabolic pathways are consistent for both inhalation and oral exposure (Bartsch et al., 1976, 1979; Green and Hathway 1975, 1977; Hathway 1977; Watanabe and Gehring 1976; Watanabe et al. 1976a). Metabolism occurs via the oxidation of vinyl chloride by mixed function oxidases (MFO) to form an epoxide intermediate, 2-chloroethylene oxide, which spontaneously rearranges to form 2-chloroacetaldehyde. Intermediates are detoxified primarily via glutathione conjugation and conjugates are excreted in urine as substituted cysteine derivatives. Metabolism has been shown to follow Michaelis-Menten kinetics in rats, with enzyme saturation near 100 ppm in air or between 1 and 100 mg/kg/day for a single gavage dose (Hefner et al. 1975b; Watanabe et al. 1976a). Macromolecular binding has been attributed to the reactive intermediate 2-chloroethylene oxide, which binds to DNA and RNA (ribonucleic acid), and its reaction

Figure 3-3. Proposed Metabolic Pathways for Vinyl Chloride*



*Derived from Bolt et al. (1980); Cogliano and Parker (1992); Hefner et al. (1975b); Park et al. (1993); and Plugge and Safe (1977).

**Excreted in urine.

product, 2-chloroacetaldehyde, which binds to protein molecules (Barbin et al., 1975; Guengerich and Watanabe 1979; Guengerich et al. 1979, 1981; Kappus et al. 1976; Watanabe et al. 1978a, 1978b). No studies were located regarding vinyl chloride metabolism in humans after oral or dermal exposure or in animals after dermal exposure. It should be noted that the toxicokinetics of vinyl chloride could be affected by compromised liver function or exposure to alcohol and other drugs and chemicals.

Animal studies have demonstrated that the primary route of excretion is dose-dependent (Watanabe and Gehring 1976; Watanabe et al. 1978a, 1976b). Vinyl chloride metabolites are excreted primarily in the urine following oral or inhalation exposure to low doses. At higher doses where metabolic saturation has been exceeded, vinyl chloride is exhaled as the parent compound. This was also demonstrated in humans exposed by inhalation, where exhalation of vinyl chloride was a minor pathway of elimination at low concentrations (Krajewski et al. 1980). No studies were located regarding excretion in humans after oral or dermal exposure to vinyl chloride. After dermal exposure in monkeys, most of the little vinyl chloride absorbed was excreted in exhaled air (Hefner et al. 1975a).

3.4.1 Absorption

3.4.1.1 Inhalation Exposure

Inhalation absorption of vinyl chloride is rapid in humans. Young adult male volunteers were exposed to vinyl chloride concentrations of 2.9, 5.1, 11.7, or 23.5 ppm by gas mask for 6 hours (Krajewski et al. 1980). Retention was estimated by measuring the difference between inhaled and exhaled concentrations. An average retention of 42% was estimated. Although the results varied among the individuals tested, the percentage retained was independent of the concentration inhaled. Since retention did not change with increasing vinyl chloride concentrations, it appears that saturation of the major pathway of overall metabolism did not occur in this exposure regimen.

Animal data demonstrate that the inhalation absorption of vinyl chloride occurs readily and rapidly. PBPK models that have been developed to provide quantitative estimates of uptake are discussed in Section 3.4.5. Peak blood levels occurred at 30 minutes in rats exposed (head only) to 7,000 ppm (Withey 1976). On removal from the vinyl chloride atmosphere, blood levels fell rapidly. After 2 hours, concentrations were barely detectable.

3.4.1.2 Oral Exposure

No studies were located regarding absorption in humans after oral exposure to vinyl chloride.

Several studies in rats indicate that vinyl chloride is rapidly and virtually completely absorbed from the gastrointestinal tract. Peak blood levels of vinyl chloride were observed within 10–20 minutes after dosing in rats administered single oral doses (44–92 mg/kg) of vinyl chloride in aqueous solution (Withey 1976). Peak blood levels varied from 6 to >40 µg/mL. Data from another study in which rats were administered single gavage doses of 0.05, 1, and 100 mg/kg vinyl chloride labelled with radioactive carbon (^{14}C-vinyl chloride) (in corn oil) suggested that almost complete absorption of vinyl chloride occurred (Watanabe et al. 1976a). The fraction of the administered dose recovered in the feces, roughly indicative of the proportion unabsorbed, ranged from 0.47 to 2.39%; total recovery ranged from 82.3 to 91.3%. Loss of radioactivity might be attributed either to experimental error or to incomplete sampling of the carcass. Fecal excretion was measured in rats fed 0, 1.8, 5.6, and 17.0 mg/kg/day of vinyl chloride monomer (from powdered PVC containing a high level of the monomer) (Feron et al. 1981). Fecal excretion accounted for 8, 10, and 17% of the vinyl chloride present in the low-, middle-, and high-dose groups, respectively. The investigators hypothesized that the vinyl chloride recovered from the feces was encapsulated by PVC and was not available to the rats for absorption, and that absorption of available vinyl chloride was virtually complete.

3.4.1.3 Dermal Exposure

No studies were located regarding absorption in humans after dermal exposure to vinyl chloride.

Animal data suggest that dermal absorption of vinyl chloride gas is not likely to be significant. Dermal absorption was measured in two rhesus monkeys that received full body (except head) exposure to vinyl chloride gas. It was estimated that 0.031 and 0.023% of the total available vinyl chloride was absorbed at 800 and 7,000 ppm, respectively, after a 2–2.5-hour exposure (Hefner et al. 1975a). The investigators concluded that, after short-term exposure to high concentrations, dermal absorption was far less significant than inhalation absorption. No information is available regarding dermal absorption of vinyl chloride from liquid or solid mediums.

3.4.2 Distribution

Representative vinyl chloride partition coefficients for humans, rats, mice, and hamsters can be found in Table 3-5. These partition coefficients were obtained for use in PBPK models. They were estimated using a vial equilibration technique (Air Force 1990b). Further details about how the values were obtained, including the number of experiments completed and whether the errors shown are standard deviations or standard errors, were not provided. In general, concentrations of vinyl chloride found in fat are higher than would be found in other tissues. Partition coefficients for vinyl chloride range from 10 to 20 (fat/air) and from 1 to 3 (muscle/air, blood/air, and liver/air). In animal studies, females have shown greater partitioning from air to fat than males.

Tissue/blood partition coefficients in male Sprague-Dawley rats, measured using a vial equilibration method, have been reported as 10±3 for fat/blood, 0.4±0.2 for muscle/blood, 0.7±0.3 for liver/blood, and 0.7±0.4 for kidney/blood (Barton et al. 1995).

3.4.2.1 Inhalation Exposure

No studies were located regarding tissue distribution in humans after inhalation of vinyl chloride.

Data from rat studies suggest that the distribution of inhaled vinyl chloride is rapid and widespread, but storage of vinyl chloride in the body is limited by rapid metabolism and excretion. In rats exposed to ^{14}C-vinyl chloride and pretreated with 6-nitro-1,2,3-benzothiadiazole to block metabolism of vinyl chloride by microsomal cytochrome P-450 oxidation pathways, the highest levels of radiolabel were located in the fat, with lesser amounts in the blood, liver, kidney, muscle, and spleen. When metabolism was not blocked, the highest levels of radiolabelled metabolites were located in the liver and kidney (Buchter et al. 1977). Immediately after a 5-hour exposure to ^{14}C-vinyl chloride at 50 ppm, tissue levels of ^{14}C-activity, expressed as the percentage incorporated per gram of tissue, were highest in the kidney (2.13%) and liver (1.86%), with lower levels in the spleen (0.73%) and brain (0.17%) (Bolt et al. 1976a). Radioactivity in tissue was measured in rats 72 hours after exposure to 10 or 1,000 ppm ^{14}C-vinyl chloride for 6 hours. In order of decreasing concentration for rats exposed to 10 ppm, ^{14}C-labeled compounds (expressed as percentage), present as nonvolatile metabolites, were measured in the liver (0.14), kidney (0.08), skin (0.07), lung (0.07), muscle (0.05), carcass (0.05), plasma (0.05), and fat (0.03). For rats exposed to 1,000 ppm, radiolabel (expressed as percentage) was measured in the liver (0.15), skin

Table 3-5. Vinyl Chloride Partition Coefficients

			Partition coefficient			
Species	Strain	Sex	Blood/air	Liver/air	Muscle/air	Fat/air
Rat	CDBR[a]	M	1.8±0.22	3.0±0.41	2.2±0.70	14.6±0.92
		F	2.1±0.44	1.7±0.43	1.3±0.25	19.2±0.96
	F-344[a]	M	1.6±0.33	2.0±2.0	2.1±0.40	11.8±0.81
		F	1.6±0.11	2.1±0.17	2.4±0.46	21.1±1.3
	Wistar[a]	M	2.1±0.31	2.7±0.56	2.7±0.58	10.2±1.6
		F	1.6±0.07	1.5±0.28	1.6±0.22	22.3±0.54
	Sprague-Dawley[b]	M	2.4±0.5	–	–	–
Mouse	$B6C3F_1$[a]	M	2.8±0.22	–	–	–
		F	2.6±0.14	–	–	–
	CD-1[a]	M	2.3±0.07	–	–	–
		F	2.4±0.16	–	–	–
Hamster	Golden Syrian[a]	M	2.7±0.15	3.4±0.36	2.6±0.46	14.3±5.3
		F	2.2±0.47	1.3±0.28	2.0±0.28	21.1±2.0
Human[c]	NA	NR	1.16	–	–	–

[a]Air Force 1990b; values determined using vial equilibration method.
[b]Barton et al. 1995
[c]EPA 1987g

– = no data; F = female; M = male; NA = not applicable; NR = not reported

(0.12), kidney (0.06), carcass (0.05), lung (0.05), muscle (0.04), fat (not detected), and plasma (not detected) (Watanabe et al. 1976b). There was no difference in the routes or rate of excretion between repeated-dose versus single-dose exposure of rats to 5,000 ppm of ^{14}C-vinyl chloride (Watanabe et al. 1978a). The concentration of radiolabel detected in tissues 72 hours after exposure revealed no statistically significant difference between rats exposed once or repeatedly to vinyl chloride. Percentages of radioactivity after 72 hours measured in tissues are as follows (for single and repeated doses, respectively): liver (0.12 and 0.16), kidney (0.06 and 0.07), skin (0.05 and 0.08), carcass (0.03 and 0.04), and fat (not detected and not detected).

Placental transfer of vinyl chloride can occur rapidly in rats. Female rats exposed to approximately 0, 2,000, 7,000, or 13,000 ppm vinyl chloride for 2.5 hours on Gd 18 showed high concentrations of vinyl chloride in maternal and fetal blood and amniotic fluid (Ungvary et al. 1978). Vinyl chloride concentrations in maternal blood were 19.02, 32.40, and 48.43 µg/mL, respectively, while fetal blood concentrations were 12.80, 22.67, and 30.52 µg/mL, respectively. Vinyl chloride concentrations in amniotic fluid were 0, 4.27, 4.93, and 13.50 µg/mL, at 2,000, 7,000, and 13,000 ppm vinyl chloride, respectively (Ungvary et al. 1978).

3.4.2.2 Oral Exposure

No studies were located regarding tissue distribution in humans after oral exposure to vinyl chloride.

The level of ^{14}C-nonvolatile metabolites was measured in tissues of rats 72 hours after single gavage doses (0.05–100 mg/kg) of ^{14}C-vinyl chloride in corn oil (Watanabe et al. 1976a). The highest levels of radioactivity for each dose level occurred in the liver. These levels were 2–5 times higher than in the other tissues examined (skin, plasma, muscle, lung, fat, and carcass).

3.4.2.3 Dermal Exposure

No studies were located regarding tissue distribution for humans or animals after dermal exposure to vinyl chloride.

3.4.3 Metabolism

3.4.3.1 Inhalation Exposure

Metabolism can be quantitatively estimated from gas uptake experiments in which, after initial absorption of vinyl chloride, continued absorption is largely attributed to metabolism. Krajewski et al. (1980) exposed young men to vinyl chloride at concentrations of 2.9, 5.1, 11.7, and 23.5 ppm by gas mask for 6 hours. Retention was independent of the inhaled concentration and did not change with increasing vinyl chloride concentrations, suggesting that saturation of the major metabolic pathway did not occur over this exposure range.

The major metabolic pathway of vinyl chloride involves oxidation by mixed-function oxidases to form a highly reactive epoxide intermediate, 2-chloroethylene oxide, which spontaneously rearranges to form 2-chloroacetaldehyde (Guengerich et al. 1979, 1981; Gwinner et al. 1983; Laib 1982). These intermediates are detoxified mainly through conjugation with glutathione catalyzed by glutathione *S*-transferase. The conjugated products are excreted in urine as substituted cysteine derivatives and include thiodiglycolic acid, *S*-formylmethylcysteine, and *N*-acetyl-*S*-(2-hydroxyethyl) cysteine (Bolt et al. 1980; Hefner et al. 1975b). Urinary metabolites identified in rats exposed by inhalation include polar compounds at low exposure concentrations (Hefner et al. 1975b; Watanabe et al. 1976b) and 2-chloroacetic acid at high exposure concentrations (Hefner et al. 1975b).

Early work on the metabolism of vinyl chloride in animals indicated that metabolism is a dose-dependent, saturable process. Rats were exposed to vinyl chloride in a closed chamber at concentrations of about 50–1,000 ppm for 52.5–356.3 minutes (Hefner et al. 1975b). Additional rats pretreated with ethanol (to inhibit alcohol dehydrogenase activity) or SKF 525-A (to inhibit microsomal oxidase activity) were similarly exposed. Metabolism, estimated by measuring the rate of disappearance of vinyl chloride from the closed system, followed first-order kinetics with a half-life of 86 minutes at <100 ppm. At >220 ppm, metabolism was slowed to a half-life of 261 minutes, suggesting saturation of the pathway predominant at 100 ppm. Pretreatment with ethanol depressed the rate of metabolism by approximately 83% at <100 ppm but by approximately 47% at >1,000 ppm. Pretreatment with SKF 525-A, however, had no effect at <100 ppm but depressed metabolism by 19% at >1,000 ppm. The study authors postulated alternative pathways for vinyl chloride metabolism. They suggested that at low concentrations sequential oxidation to 2-chloroethanol, 2-chloroacetaldehyde, and 2-chloroacetic acid involving alcohol dehydrogenase (inhibited by pretreatment with ethanol) appeared to be the predominant pathway. Little 2-chloroacetic acid was formed, however, possibly because 2-chloroacetaldehyde conjugated rapidly with

ubiquitous sulfhydryl groups. The authors further speculated that when the alcohol dehydrogenase pathway became saturated, 2-chloroethanol could be oxidized by catalase in the presence of hydrogen peroxide (H_2O_2) to a peroxide, which could undergo subsequent dehydration to form 2-chloro-acetaldehyde. However, it appears that the only support for this proposed metabolism of vinyl chloride by alcohol dehydrogenase comes from studies demonstrating metabolic inhibition by alcohol. This is not recognized as a direct pathway for metabolism of vinyl chloride in modern PBPK modeling studies. It is possible that ethanol exerts its effects by inhibiting specific P-450 enzymes involved in the metabolic activation of vinyl chloride.

Isolated rat liver cells converted ^{14}C-vinyl chloride into nonvolatile metabolites (Hultmark et al. 1979). Using this *in vitro* technique, it was determined that metabolism was NADPH-dependent, located in the microsomal fraction of the liver, and probably involved an MFO. Pretreatment with 6-nitro-1,2,3-benzo-thiadiazole, an inhibitor of some microsomal cytochrome P-450 oxidation pathways, was sufficient to totally block the metabolism of vinyl chloride in rats exposed to 0.45 ppm in a closed system for 5 hours (Bolt et al. 1977). This observation suggests that metabolism of vinyl chloride proceeds primarily through an MFO pathway with likely production of an epoxide intermediate.

Inhalation exposure to high concentrations of vinyl chloride has also been associated with a reduction in the liver nonprotein sulfhydryl concentration in the rat (Barton et al. 1995). These results are consistent with conjugation of the metabolites of vinyl chloride with limited reserves of glutathione and/or cysteine (Bolt et al. 1976b; Hefner et al. 1975b; Jedrychowski et al. 1984; Watanabe et al. 1978b).

Saturation of metabolic pathways was observed in rats and monkeys that were exposed in a closed system to ^{14}C-vinyl chloride (Bolt et al. 1977; Buchter et al. 1980; Filser and Bolt 1979). In Wistar rats, metabolic saturation was determined to occur at approximately 250 ppm, and a metabolic rate (V_{max}) of 110 µmol/hour/kg was estimated (Bolt et al. 1977; Filser and Bolt 1979). Kinetic constants of 58 µmol/hour/kg for V_{max} and 1 µM for the K_m in male Sprague-Dawley rats have also been reported (Barton et al. 1995). In an experiment using rhesus monkeys, metabolic saturation occurred at 200 ppm, with a V_{max} of 50 µmol/hour/kg (Buchter et al. 1980). The V_{max} of 50 µmol/hour/kg that was estimated using rhesus monkeys was suggested as a closer approximation of metabolism in humans than the value of 110 µmol/hour/kg estimated for rats by Filser and Bolt (1979).

Kinetic constants for vinyl chloride metabolism have also been derived from *in vitro* studies in rat liver microsomes (El Ghissassi et al. 1998). Vinyl chloride metabolism to reactive species followed Michaelis-

Menton kinetics with a K_m of 7.42 μM and a V_{max} of 4,674 pmol/mg protein/minute. Inhibitor studies using chemical and immunological inhibitors demonstrate that vinyl chloride is metabolized primarily by CYP2E1.

Several investigators have observed the binding of nonvolatile metabolites of ^{14}C-vinyl chloride to liver macromolecules *in vitro* and in rats exposed by inhalation (Guengerich and Watanabe 1979; Guengerich et al. 1979, 1981; Kappus et al. 1976; Watanabe et al. 1978a, 1978b). In single-exposure experiments at different concentrations, the extent of macromolecular binding increased proportionately to the amount of vinyl chloride metabolized and disproportionately to the exposure concentration (Watanabe et al. 1978b). The extent of macromolecular binding was increased by repeated exposure to vinyl chloride (Watanabe et al. 1978a) and by pretreatment with phenobarbital (Guengerich and Watanabe 1979). Macromolecular binding has been attributed to the reactive intermediate 2-chloroethylene oxide, which has been shown to bind to DNA and RNA, and to its rearrangement product, 2-chloroacetaldehyde, which has been shown to bind to protein molecules (Guengerich and Watanabe 1979; Guengerich et al. 1979, 1981; Kappus et al. 1976; Watanabe et al. 1978a, 1978b).

3.4.3.2 Oral Exposure

No studies were located regarding metabolism in humans after oral exposure to vinyl chloride.

Urinary metabolites identified from rats ingesting ^{14}C-vinyl chloride are consistent with the metabolic pathways postulated for inhalation exposure, in particular with the formation of 2-chloroethylene oxide and 2-chloroacetaldehyde. Metabolites identified include *N*-acetyl-*S*-(2-hydroxyethyl)cysteine, 2-chloroacetic acid, and thiodiglycolic acid (Green and Hathway 1975, 1977; Watanabe and Gehring 1976; Watanabe et al. 1976a). Metabolic saturation appears to occur with a single gavage dose of between 1 and 100 mg/kg/day (Watanabe et al. 1976a).

3.4.3.3 Dermal Exposure

No studies were located regarding metabolism in humans or animals after dermal exposure to vinyl chloride.

3.4.4 Elimination and Excretion

3.4.4.1 Inhalation Exposure

Human data suggest that exhalation of unmetabolized vinyl chloride is not an important pathway of elimination at low exposure concentrations. The mean concentration in expired air for humans exposed for 6 hours to air containing 2.9–23.5 ppm ranged from 0.21 to 1.11 ppm, representing from 7.23 to 4.73% of the inhaled amounts, respectively (Krajewski et al. 1980).

Animal studies indicate that the importance of exhalation of vinyl chloride as a major route of excretion varies with the exposure concentration. The mode of excretion of vinyl chloride and its metabolites following inhalation exposure of animals to different concentrations reflects the saturation of metabolic pathways. The cumulative excretion of radioactivity over a 72-hour postexposure period was measured in rats exposed to 10–1,000 ppm (Watanabe and Gehring 1976; Watanabe et al. 1976b) or 5,000 ppm (Watanabe et al. 1978a) ^{14}C-vinyl chloride for 6 hours. Radioactivity expired as carbon dioxide or vinyl chloride, excreted in the urine and feces, and retained in the carcass was expressed as a percentage of the total radioactivity recovered. The results suggest that metabolism was nearly complete at 10 ppm because less than 2% of the recovered radioactivity occurred as unchanged parent compound. The predominant route for excretion of radioactive metabolites was through the urine, accounting for about 70% of the recovered radioactivity. Metabolism became saturated at 1,000 ppm, since unchanged vinyl chloride increased to 12.3% and urinary radioactivity decreased to 56.3%. At 5,000 ppm, more than half the recovered radioactivity appeared as unchanged vinyl chloride in expired air, and urinary excretion accounted for about 27% of the recovered activity. Generally, there was little change in the proportion of recovered radioactivity excreted in the feces or exhaled as carbon dioxide. The percentage of the radioactivity retained in the carcass and tissues appeared to be somewhat decreased at 5,000 ppm compared with 10 and 1,000 ppm, suggesting preferential retention of metabolites rather than unchanged vinyl chloride. It should be noted that the trend of a greater percentage of vinyl chloride being exhaled at higher concentrations in animals is the opposite of what was observed in humans in Krajewski et al. (1980). In humans, a higher percentage of unmetabolized vinyl chloride was found in expired air at lower concentrations (Krajewski et al. 1980). However, it is possible that a reversal of this trend would occur in humans if concentrations were increased to those used in the animal studies or to concentrations closer to the K_m for human metabolism.

Pulmonary excretion of unaltered vinyl chloride in rats followed first-order kinetics regardless of exposure concentrations, with half-lives of 20.4, 22.4, and 30 minutes following 6-hour exposures at 10,

1,000, and 5,000 ppm, respectively. The urinary excretion of radioactivity was biphasic, with the second or slow phase accounting for less than 3% of the total urinary excretion. Estimated half-lives for the rapid (first-order) phase were 4.6, 4.1, and 4.5 hours, at 10, 1,000, and 5,000 ppm, respectively. Urinary metabolites included *N*-acetyl-*S*- (2-hydroxyethyl)cysteine, thiodiglycolic acid, and possibly *S*-(2-hydroxyethyl)cysteine (Watanabe et al. 1976b). Identification of these metabolites of vinyl chloride in the urine indicates that vinyl chloride is transformed in the body to a reactive metabolite, which is then detoxified by reaction with glutathione (GSH, gamma-glutamylcysteinylglycine). Subsequently the glutamic acid and glycine moieties of the tripeptide are cleaved, and the cysteine conjugate of the reactive metabolite of vinyl chloride is either acetylated or further oxidized and excreted. Thiodiglycolic acid is the major metabolite of vinyl chloride detected in the urine of exposed workers (Cheng et al. 2001). Urinary thiodiglycolic acid levels were correlated with vinyl chloride levels in air at concentrations >5 ppm.

3.4.4.2 Oral Exposure

No studies were located regarding excretion in humans after oral exposure to vinyl chloride.

Single oral doses of ^{14}C-vinyl chloride (0.05, 0.25, 1.0, 20, 100, and 450 mg/kg) were administered to rats, and the excretion of radioactivity was monitored over a 72-hour period (Green and Hathway 1975; Watanabe and Gehring 1976; Watanabe et al. 1976a). A striking increase in exhalation of unchanged vinyl chloride and compensatory decreases in urinary and fecal excretion of radioactivity and exhalation of carbon dioxide were observed at >20 mg/kg, suggesting that metabolic saturation had occurred at that dosage. At less than 1.0 mg/kg, the predominant route of elimination was urinary excretion of polar metabolites.

Exhalation of unchanged vinyl chloride was generally complete within 3–4 hours, but excretion of metabolites continued for days (Green and Hathway 1975). Pulmonary excretion of vinyl chloride appeared to be monophasic at less than 1.0 mg/kg, with a half-life of about 55–58 minutes (Watanabe et al. 1976a). At 100 mg/kg, pulmonary excretion of vinyl chloride was biphasic, with half-lives of 14.4 and 40.8 minutes for the rapid and slower phases, respectively. Urinary excretion of radioactivity was biphasic, with the rapid phase accounting for more than 97% of total urinary radioactivity and having half-lives of 4.5–4.6 hours for dosages of 0.05–100 mg/kg.

Metabolites identified in the urine of orally treated rats were consistent with the formation of 2-chloroethylene oxide and 2-chloroacetaldehyde (Green and Hathway 1977; Watanabe et al. 1976a), as postulated for metabolism following inhalation exposure. The major metabolites were identified as thiodiglycolic acid and *N*-acetyl-*S*-(2-hydroxyethyl)cysteine (Watanabe et al. 1976a). *N*-Acetyl-*S*-(2-chloroethyl)cysteine and *S*-(2-chloroethyl)cysteine have also been identified as having smaller amounts of radiolabelled urea, glutamic acid, and 2-chloroacetic acid (Green and Hathway 1975).

3.4.4.3 Dermal Exposure

No studies were located regarding excretion in humans after dermal exposure to vinyl chloride.

When two rhesus monkeys received whole-body (except head) exposure to vinyl chloride gas (800 and 7,000 ppm) for 2–2.5 hours, although very little vinyl chloride was absorbed, most was excreted in expired air (Hefner et al. 1975a). The percentages of absorbed vinyl chloride that were exhaled were 0.028 and 0.014% at 700 and 8,000 ppm, respectively (Hefner et al. 1975a).

3.4.4.4 Other Routes of Exposure

The elimination of radioactivity following intraperitoneal administration of ^{14}C-vinyl chloride to rats resembles the pattern observed following inhalation or oral administration. Following an intraperitoneal dose of 0.25 mg/kg, exhalation of unchanged vinyl chloride, exhalation of carbon dioxide, and urinary and fecal excretion of radioactivity accounted for 43.2, 11.0, 43.1, and 1.8% of the administered dose, respectively (Green and Hathway 1975). At 450 mg/kg, exhaled vinyl chloride increased to 96.2% of the administered dose, carbon dioxide decreased to 0.7%, urinary radioactivity decreased to 2.6%, and fecal radioactivity decreased to 0.1%.

Doses administered intravenously were eliminated very rapidly and almost entirely by exhalation of unchanged vinyl chloride. Green and Hathway (1975) administered a 0.25-mg/kg intravenous dose of ^{14}C-vinyl chloride to rats and recovered 80% of the dose within 2 minutes and 99% within 1 hour as unchanged compound in expired air.

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 3-4 shows a conceptualized representation of a PBPK model.

If PBPK models for vinyl chloride 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.

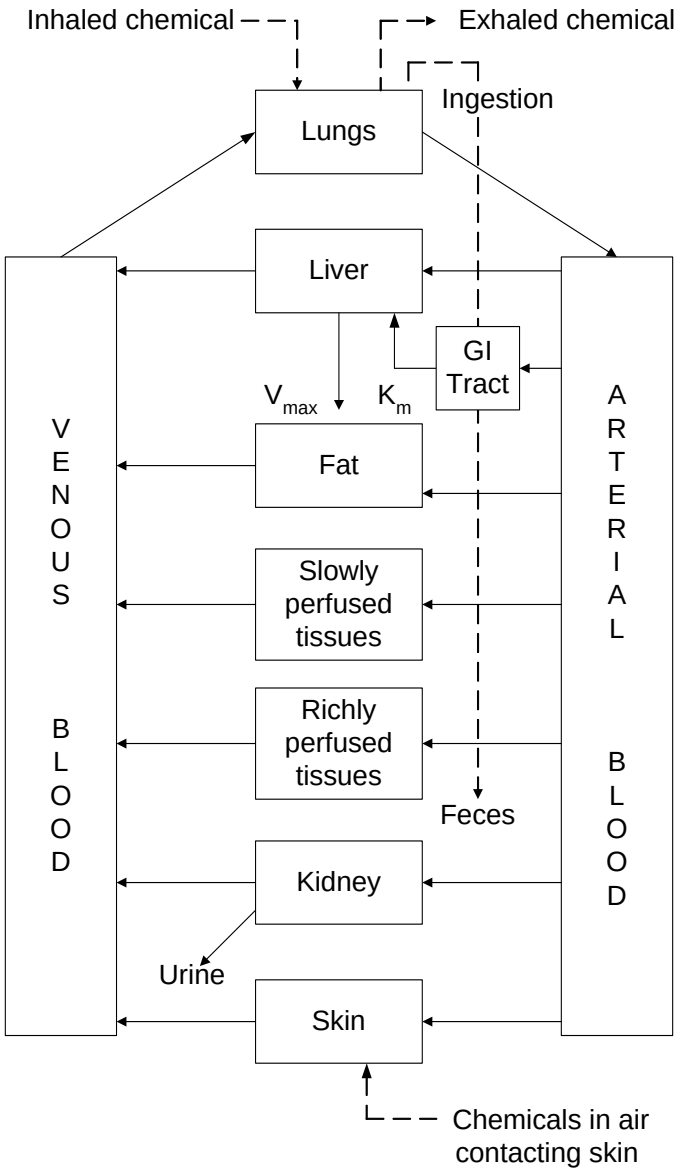
PBPK models are available for vinyl chloride. The overall results and individual models are discussed in this section in terms of their use in risk assessment, tissue dosimetry, and dose, route, and species extrapolations.

Summary of PBPK/PD Models

Models have been developed to predict the metabolism and distribution of vinyl chloride. EPA (1987g) developed a PBPK model to estimate the metabolized dose of vinyl chloride coupled to a multistage model to estimate cancer risk in animals. This PBPK model consists of four compartments, the liver, fat, highly perfused tissue, and poorly perfused tissue. All metabolism was assumed to occur in the liver by one saturable pathway (Michaelis-Menten kinetics) and by a first-order metabolism pathway. The physiologic parameters used were values from an EPA draft "Reference Physiologic Parameters in Pharmacokinetic Modeling" by Dr. Curtis Travis of the Oak Ridge National Laboratory.

The dose delivery of the vinyl chloride model developed by EPA (1987g) was further validated by the Air Force (1990b) study with additional vinyl chloride metabolism studies in rats. At low concentrations, this model fit *in vivo* data in rats by Gehring et al. (1978) well, but at concentrations above 25 ppm, the model predicted a greater amount of vinyl chloride metabolism than observed. The Air Force (1990b) then made modifications in the model to improve the fit with actual data. In the first modification, both vinyl

Figure 3-4. Conceptual Representation of a Physiologically Based Pharmacokinetic (PBPK) Model for a Hypothetical Chemical Substance



Source: adapted from Krishnan et al. 1994

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

chloride and the epoxide metabolite were assumed to react with glutathione. This model had difficulty predicting glutathione depletion at high doses; for example, it predicted glutathione depletions higher than observed at 4,600–5,800 ppm vinyl chloride. The second alternative model, in which only the product of the first-order metabolism was assumed to react with glutathione, also predicted glutathione depletions higher than observed at high concentrations. To improve the model, the investigators suggested the addition of a low-affinity glutathione pathway.

Using data obtained from Wright-Patterson Air Force Base, the Air Force (1990b) extended the first glutathione conjugation model, developed in rats, to different strains of rats, mice, and hamsters. Vinyl chloride gas uptake experiments were completed in which animals were exposed to various concentrations of vinyl chloride in closed chambers for up to 6 hours, and the disappearance of vinyl chloride was monitored. The glutathione content of the animals was also measured immediately after exposure. Using data from these studies and the physiologic parameters shown in Table 3-6, the investigators estimated metabolic parameters for vinyl chloride and the rate constant for the conjugation of vinyl chloride with glutathione (Table 3-7). Using the metabolic parameters determined from the gas uptake experiments, the model predictions showed good agreement with the actual data for all the strains tested. It does not appear that the investigators further validated the model with data from studies other than those used to determine the metabolic parameters. This model was not used to estimate metabolized doses for humans because the investigators indicated that human data to estimate all the required parameters were not available. They suggested that allometry may have to be used to estimate some of the parameters for humans.

Clewell et al. (1995) used PBPK modeling coupled with a linearized multistage model to predict human cancer risk. The model again had four compartments as described for the EPA (1987g) study, and the same EPA physiologic parameters were used. Partition coefficients were from *in vitro* experiments and are shown in Table 3-4. Metabolism was modeled by two saturable pathways: one high affinity, low capacity (P450 2E1), and one low affinity, high capacity (2C11/6 and 1A1/2). The metabolic parameters used were not provided, but they were estimated from the Air Force (1990b) model. This model assumed that the metabolites (chloroethylene oxide and chloroacetaldehyde) were further degraded to carbon dioxide, or reacted with glutathione, or reacted with DNA. The parameters (not stated) for reactions of the metabolites were estimated from vinylidene chloride data (D'Souza and Andersen 1988) using appropriate allometric scaling. Based on this PBPK model and a linearized multistage model using liver angiosarcoma data from animal studies, the human risk estimates for lifetime exposure to 1 ppb vinyl chloride ranged from 1.1 to 15.7/million persons (Clewell et al. 1995). Based on the incidence of liver

Table 3-6. Physiological Parameters Used to Estimate Parameters from Vinyl Chloride Gas Uptake Experiments[a]

Parameter	Rats	Mice	Hamsters
Ventilation rate (L/hour/body weight$^{0.74}$)	14	23–25[b]	13
Total cardiac output (L/hour/body weight$^{0.74}$)	14	23–25[b]	13
Blood flow to the liver (fraction of total cardiac output)	0.25	0.24	0.24
Blood flow to highly perfused tissue (fraction of total cardiac output)	0.51	0.52	0.52
Blood flow to fat (fraction of total cardiac output)	0.09[c]	0.05	0.09
Blood flow to poorly perfused tissue (fraction of total cardiac output)	0.15[c]	0.20	0.15
Volume of tissue (L/body weight)	0.04	0.04	0.04
Volume of highly perfused tissue (L/body weight)	0.04	0.05	0.05
Volume of fat tissue (L/body weight)	0.07–0.1[d]	0.04	0.07
Volume of poorly perfused tissue (L/body weight)	0.72–0.75[d]	0.78	0.75

[a]Air Force 1990b; units of body weight were not provided.
[b]Ventilation rates and total cardiac outputs were 23 for male B6C3F$_1$ mice, 25 for female B6C3F$_1$ mice, 28 for female CD-1 mice, and 35 for male CD-1 mice.
[c]Male Wistar rats blood flow to fat = 0.08 and blood flow to slowly perfused tissue = 0.16.
[d]Female F-344 and female Wistar rats had volume of fat tissue = 0.07 and volume of slowly perfused tissue = 0.75; male F-344 and female Wistar rats had volume of fat tissue = 0.08 and volume of slowly perfused tissue = 0.74; male Wistar rats and male CDBR rats had volume of fat tissue = 0.1 and volume of slowly perfused tissue = 0.72.

Table 3-7. Estimates of Metabolic Parameters Obtained from Gas Uptake Experiments

Species	Strain	Sex	V_{max}/body weight$^{0.7}$ (mg/hour/body weight$^{0.7}$)	Kfc (body weight$^{0.3}$/ hour)	Kgsc (body weight$^{0.3}$/hour/µmol/L GSH)
Rat	CDBR	M	2.5	0.63	ND
		F	2.47	1.0	0.000241
	F-344	M	3.17	1.08	0.000249
		F	2.95	1.03	0.000227
	Wistar	M	3.11	0.45	0.000093
		F	2.97	1.55	0.00040
Mouse	$B6C3F_1$	M	5.89	5.5	0.000827
		F	5.53	8.93	0.00167
	CD-1	M	6.99	5.1	0.000563
		F	5.54	6.62	0.000809
Hamster	Golden Syrian	M	4.94	1.67	ND
		F	4.76	2.06	0.000330

Source: Air Force 1990b

F = female; GSH = glutathione; Kfc = first order of epoxide formation; Kgsc = rate constant for conjugation of vinyl chloride with glutathione; M = male; ND = not determined; V_{max} = maximum velocity of reaction

angiosarcoma in human epidemiological studies, the risk estimates for lifetime exposure to 1 ppb vinyl chloride were 0.4–4.22/million persons. Clewell et al. (1995) indicated that the risk estimates in the occupational exposure range using PBPK modeling are about 30–50 times lower than estimates using external dose calculations based on the linearized multistage model.

Reitz et al. (1996) also developed a PBPK model that coupled measures of delivered dose in rats to a linearized multistage model to predict the incidence of hepatic angiosarcoma in mice and humans. The model incorporated four compartments—fat, muscle, rapidly perfused tissues, and liver. Physiological parameters in the model were based on similar ones used in an earlier multispecies PBPK model developed for methylene chloride. Partition coefficients were estimated by vial equilibration techniques similar to those described in the Air Force (1990b) study. Metabolic rate constants were obtained from *in vivo* gas uptake experiments performed at Wright-Patterson Air Force Base.

Based on the PBPK-based procedure utilized by Reitz et al. (1996), the predicted human risk estimates ranged from about 200 cases/100,000 (for workers employed 10 years at a plant where the TWA was 50 ppm) to almost 4,000 cases/100,000 in workers employed for 20 years in a plant where the TWA was 2,000 ppm. The predictions of human risk were compared with the data reported by Simonato et al. (1991). The predictions of angiosarcoma incidence in humans were almost an order of magnitude higher than actually observed in exposed human populations, and were more than two orders of magnitude lower than risk estimations that did not utilize pharmacokinetic data.

Clewell et al. (2001) futher refined the PBPK model for vinyl chloride and this model was applied by the EPA to develop quantitative toxicity values for vinyl chloride (i.e., reference dose [RfD], reference concentration [RfC], inhalation unit risk, oral slope factor) (EPA 2000). The model had four compartments and metabolism was modeled by two saturable pathways: one high affinity, low capacity (P450 2E1), and one low affinity, high capacity (2C11/6 and 1A1/2). A description of glutathione kinetics was also included in the model. Cancer risk estimates in the occupational exposure range calculated using the PBPK model were consistent with risk estimates from epidemiological studies and were approximately 80-fold lower than cancer risk estimates from animal studies without PBPK modeling. The inhalation portion of the PBPK model is well documented with experimental inhalation data sufficient to ensure a high degree of confidence in the derived dose metrics. Less confidence is associated with the oral dose metrics due to the limited experimental data available (EPA 2000).

The Clewell et al. (2001) model was also recently applied to evaluate the potential impact of age- and gender-specific pharmacokinetic differences on the dosimetry of vinyl chloride (Clewell et al. 2004). The rate of metabolite production per volume of liver was estimated to rise rapidly from birth until about age 16, after which it remains relatively constant before rising again late in life. Other factors that may affect vinyl chloride toxicity at early life stages include the presence of fetal P450s and the level of glutathione transferase.

The PBPK model described in Clewell et al. (2001) and EPA (2000) was used to derive the chronic-duration oral MRL. The chronic oral MRL for vinyl chloride is based on the same critical effect as that used by EPA (2000) to derive the RfD for vinyl chloride (i.e., the NOAEL for liver cell polymorphism in the oral rat study of Til et al. 1983, 1991). Source code and parameter values for running the rat and human models in Advance Continuous Simulation Language (ACSL) were transcribed from Appendix C of EPA (2000). Exposures in the Til et al. (1983, 1991) rat dietary study were simulated as 4-hour oral exposures with the NOAEL dose for liver effects of 0.17 mg/kg/day. A 4-hour feeding period was used in the study due to the rapid evaporative loss of vinyl chloride from the food. The total amount of vinyl chloride metabolized in 24 hours per liter of liver volume was the rat internal dose metric that was used in determining the human dose that would result in an equivalent human dose metric. One kilogram of liver was assumed to have an approximate volume of 1 L. Dose metrics reflect the cumulative amount of vinyl chloride metabolized over the 24-hour period. The human model was run iteratively, until the model converged with the internal dose estimate for the rat (3.16 mg/L liver). The human dose was assumed to be uniformly distributed over a 24-hour period with the resulting human equivalent dose of 0.09 mg/kg/day. Therefore, the human equivalent dose of 0.09 mg/kg/day, associated with the rat NOAEL of 0.17 mg/kg/day (Til et al. 1983, 1991), served as the basis for the chronic-duration oral MRL for vinyl chloride. A total uncertainty factor of 30 (3 for extrapolating from animals to humans using a dose metric conversion and 10 for human variability) was applied to yield the chronic-duration oral MRL of 0.003 mg/kg/day (see Appendix A for more detailed information regarding the application of the PBPK modeling in deriving the chronic-duration oral MRL for vinyl chloride).

3.5 MECHANISMS OF ACTION

3.5.1 Pharmacokinetic Mechanisms

Absorption. Vinyl chloride appears to be rapidly and completely absorbed following inhalation and oral exposure (Bolt et al. 1977; Krajewski et al. 1980; Watanabe et al. 1976a; Withey 1976).

Distribution. Distribution of vinyl chloride in the body is rapid and widespread. Storage is limited by rapid metabolism and excretion (Bolt et al. 1976a).

Metabolism. Vinyl chloride is metabolized by mixed function oxidases (MFO) to form an epoxide intermediate, 2-chloroethylene oxide, which spontaneously rearranges to form 2-chloroacetaldehyde. Reactive metabolites of vinyl chloride are detoxified by a reaction with glutathione. The glutamic acid and glycine moieties of the tripeptide are cleaved, and the cysteine conjugate of the reactive metabolite is either acetylated or further oxidized and excreted.

Excretion. The primary route of excretion of metabolites of vinyl chloride is through urine. Urinary metabolites that have been identified include *N*-acetyl-*S*-(2-hydroxyethyl)cysteine, thiodiglycolic acid, and possibly *S*-(2-hydroxyethyl)cysteine (Watanabe et al. 1976b). Exhalation of unmetabolized vinyl chloride is not an important pathway of elimination by humans after exposure to low concentrations. The importance of exhalation of vinyl chloride varies with the exposure concentration. At low exposure concentrations, little vinyl chloride is excreted unchanged in exhaled air. However, vinyl chloride can be excreted unchanged in exhaled air if metabolic pathways become saturated at high exposure concentrations (Green and Hathway 1975; Watanabe and Gehring 1976; Watanabe et al. 1976a, 1978a).

3.5.2 Mechanisms of Toxicity

The mechanisms of toxicity for noncancer effects of vinyl chloride have not been completely elucidated. Vinyl chloride disease exhibits many of the characteristics of autoimmune diseases (Raynaud's phenomenon and scleroderma). B-cell proliferation, hyperimmunoglobulinemia, and complement activation, as well as increased circulating immune complexes or cryoglobulinemia, have been noted in affected workers, indicating stimulation of immune response (Bogdanikowa and Zawilska 1984; Grainger et al. 1980; Ward 1976). Mechanisms for the vascular changes, such as those occurring with Raynaud's phenomenon, have been proposed by Grainger et al. (1980) and Ward (1976). According to these mechanisms, a reactive vinyl chloride intermediate metabolite, such as 2-chloroethylene oxide or 2-chloroacetaldehyde, binds to a protein such as IgG. The altered protein initiates an immune response, with deposition of immune products along the vascular endothelium. Circulating immune complexes are proposed to precipitate in response to exposure to the cold, and these precipitates are proposed to produce blockage of the small vessels. Resorptive bone changes in the fingers, also characteristic of vinyl chloride disease, may be due to activation of osteoclast secondary to vascular insufficiency in the finger tips, but

this mechanism has not been conclusively demonstrated. Scleroderma is an autoimmune disease of unknown etiology. It is characterized clinically by cutaneous and visceral fibrosis and can range from limited skin involvement to extensive cutaneous sclerosis with internal organ changes. It has been proposed that fetal cells may be involved in the pathogenesis of scleroderma. An increase in the number of microchimeric cells of fetal origin was reportedly associated with dermal fibrosis in mice injected with vinyl chloride (Christner et al. 2001).

It has been hypothesized that cardiac arrhythmia reported after vinyl chloride exposure may result from sensitization of the heart to circulatory catecholamines, as occurs with other halogenated hydrocarbons. This was demonstrated in a dog study where the EC_{50} for cardiac sensitization was determined to be 50,000 ppm (Clark and Tinston 1973). Cardiac sensitization by halogenated hydrocarbons generally occurs at very high air concentrations (0.5–90%) (Brock et al. 2003). Therefore, it appears unlikely that persons exposed to low levels of vinyl chloride will experience these effects.

Peripheral nervous system symptoms such as paresthesia, numbness, weakness, warmth in the extremities, and pain in the fingers have been reported after vinyl chloride exposure (Langauer-Lewowicka et al. 1983; NIOSH 1977; Suciu et al. 1963, 1975). It is not known whether these effects represent direct adverse effects of vinyl chloride on peripheral nerves or whether they are associated with tissue anoxia due to vascular insufficiency.

Vinyl chloride is a known human and animal carcinogen. It has been associated with both an increased incidence of hepatic angiosarcomas and hepatotoxicity. The mechanism for these liver effects has been studied to some extent. Vinyl chloride is metabolized by MFO to form an epoxide intermediate, 2-chloroethylene oxide, which spontaneously rearranges to form 2-chloroacetaldehyde. Reactive metabolites of vinyl chloride can be transported intercellularly from parenchymal cells to the nonparenchymal cells (Kuchenmeister et al. 1996). Many studies have characterized the mutation profile associated with DNA adducts formed by the reactive metabolites of vinyl chloride (Akasaka et al. 1997; Chiang et al. 1997; Dosanjh et al. 1994; Guichard et al. 1996; Matsuda et al. 1995; Pandya and Moriya 1996; Zhang et al. 1995; Zielinski and Hergenhahn 2000). Four primary DNA adducts are formed by the reactive metabolites of vinyl chloride. These are cyclic etheno-adducts that include $1,N^6$-ethenoadenine, $3,N^4$-ethenocytosine, $N^2,3$-ethenoguanine, and $1,N^2$-ethenoguanine. These adducts can produce base-pair (i.e., purine-to-purine or pyrimidine-to-pyrimidine exchange) transitions during transcription (Cullinan et al. 1997; Pandya and Moriya 1996; Singer et al. 1987, 1996). DNA crosslinks can also be formed because chloracetaldehyde is bifunctional (Singer 1994).

The role of etheno-adducts in the carcinogenesis of vinyl chloride has been recently reviewed (Albertini et al. 2003, Barbin 1998, 2000; Kielhorn et al. 2000; Laib 1986; Whysner et al. 1996). 2-Chloroethylene oxide and 2-chloroacetaldehyde can both react with DNA nucleotide bases; however, 2-chloroethylene oxide is a more potent mutagen and may be the ultimate carcinogenic metabolite of vinyl chloride (Chiang et al. 1997). Etheno-adducts generate mainly base pair substitiution mutations. Mutations in specific genes (i.e., *ras* oncogenes, p53 tumor suppressor gene) have been identified in vinyl chloride-induced liver tumors in rats and humans and are discussed in further detail in Section 3.3.

The mechanisms for clastogenic effects of vinyl chloride exposure were examined by Fucic et al. (1990). Since chromatid and bichromatid breaks most frequently occurred in the terminal A, B, and C group chromosomes, these investigators suggested that vinyl chloride or its metabolites might interact with specific sites along the chromosome. This implies that the carcinogenicity induced by vinyl chloride can be explained in part by its nonrandom interaction with particular genes.

Liver toxicity has been demonstrated in workers exposed vinyl chloride (Berk et al. 1975; Falk et al. 1974; Gedigke et al. 1975; Ho et al. 1991; Jones and Smith 1982; Lilis et al. 1975; Liss et al. 1985; Marsteller et al. 1975; NIOSH 1977; Popper and Thomas 1975; Suciu et al. 1975; Tamburro et al. 1984; Vihko et al. 1984). The mechanism for liver toxicity is thought to be related to the production of reactive metabolites that covalently bind to liver proteins, resulting in cellular toxicity (Kappas et al. 1975). The intermediary metabolites, 2-chloroethylene oxide and 2-chloroacetaldehyde, bind to macromolecules in the body. 2-Chloroethylene oxide is believed to bind primarily to DNA and RNA, whereas 2-chloroacetaldehyde binds primarily to proteins (Bolt 1986; Guengerich and Watanabe 1979; Guengerich et al. 1979, 1981; Kappus et al. 1976; Watanabe et al. 1978a, 1978b).

3.5.3 Animal-to-Human Extrapolations

Limited information is available regarding the toxicokinetic differences between species. Toxicokinetic data in humans are limited (Krajewski et al. 1980; Sabadie et al. 1980), but a primate study suggested that metabolism may saturate at lower concentrations in primates than in rats (Buchter et al. 1980), which is suggestive of a lower saturation point in humans. Exposure concentrations greater than about 300–400 ppm in the primate study showed saturation characteristics (Buchter et al. 1980). PBPK models have been developed to predict the metabolism and distribution of vinyl chloride in laboratory animals and humans (see Section 3.4.5). The most recent PBPK model for vinyl chloride (Clewell et al. 2001) was

applied by the EPA to develop quantitative toxicity values for vinyl chloride (i.e., RfD, RfC, inhalation unit risk, oral slope factor) (EPA 2000). The model had four compartments and metabolism was modeled by two saturable pathways: one high affinity, low capacity (P450 2E1), and one low affinity, high capacity (2C11/6 and 1A1/2). A description of glutathione kinetics was also included in the model. Cancer risk estimates calculated using the PBPK model were consistent with risk estimates from epidemiological studies.

Correlation of toxic effects between humans and animals with regard to respiratory, cardiovascular, hematological, hepatic, dermal, immunological, neurological, reproductive, and cancer effects has been noted. Renal effects, including increased relative kidney weight and an increase in severity of tubular nephrosis, have been reported in several rat studies (Bi et al. 1985; Feron and Kroes 1979; Feron et al. 1979a), but no evidence of renal effects has been shown in humans. Thus, it is unclear whether the renal effects reported in rats represent a lesion that can be attributed to vinyl chloride exposure that is unique to rats or whether the effects represent an increase in severity of a naturally occurring lesion. From the limited data available, however, it does not appear that the rat is the most appropriate species for use in studies of renal toxicity.

3.6 TOXICITIES MEDIATED THROUGH THE NEUROENDOCRINE AXIS

Recently, attention has focused on the potential hazardous effects of certain chemicals on the endocrine system because of the ability of these chemicals to mimic or block endogenous hormones. Chemicals with this type of activity are most commonly referred to as *endocrine disruptors.* However, appropriate terminology to describe such effects remains controversial. The terminology *endocrine disruptors*, initially used by Thomas and Colborn (1992), was also used in 1996 when Congress mandated the EPA to develop a screening program for "...certain substances [which] may have an effect produced by a naturally occurring estrogen, or other such endocrine effect[s]...". To meet this mandate, EPA convened a panel called the Endocrine Disruptors Screening and Testing Advisory Committee (EDSTAC), and in 1998, the EDSTAC completed its deliberations and made recommendations to EPA concerning *endocrine disruptors.* In 1999, the National Academy of Sciences released a report that referred to these same types of chemicals as *hormonally active agents*. The terminology *endocrine modulators* has also been used to convey the fact that effects produced 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 produce 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 functions. 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).

Vinyl chloride has not been classified as an endocrine modulator; however, adverse reproductive and developmental effects have been reported in human and laboratory animal studies. Effects on the thyroid gland have also been reported.

A number of case studies of male workers occupationally exposed to vinyl chloride report sexual impotence, loss of libido, and decreased androgen secretion (Suciu et al. 1975; Veltman et al. 1975; Walker 1976). Preeclampsia (i.e., elevated blood pressure and edema during pregnancy) was reported in female workers exposed to vinyl chloride (Bao et al. 1988). Animal studies indicate that exposure to vinyl chloride can result in a decrease in testicular function (Bi et al. 1985; Sokal et al. 1980); however, these effect appears to be due to direct toxicity at the target organ and are not related to a hormone-mediated mechanism of action.

Reproductive capability was not affected in a 2-generation inhalation reproductive toxicity study in rats (Thornton et al. 2002). No effects were seen in body weight, feed consumption, ability to reproduce, gestation index or length, or pre- and postweaning developmental landmarks. Sperm counts, motility, and morphology were also unaffected by vinyl chloride exposure. Changes were observed in liver weights and/or histopathological alterations in the liver of F_0 and F_1 generation male and female rats. No effect was observed on male fertility or pre- or postimplantation loss in mice following an acute exposure to vinyl chloride (i.e., 30,000 ppm, 6 hours/day, 5 days/week) (Anderson et al. 1976). In contrast, exposure of male rats to concentrations as low as 250 ppm for 6 hours/day, 5 days/week for 11 weeks produced a decrease in the ratio of pregnant to mated females, indicating a decrease in male fertility; this effect was not observed at 50 ppm (Short et al. 1977).

Although evidence has been presented indicating that members of communities with nearby vinyl chloride polymerization facilities have significantly greater incidences of some forms of developmental toxicity, these studies failed to demonstrate a statistically significant correlation between the developmental toxicity and either parental occupation or proximity to the facility (Edmonds et al. 1978; Infante et al. 1976b; Rosenman et al. 1989; Theriault et al. 1983). Vinyl chloride did not correlate with changes in gender ratio, birth weight or height, perinatal mortality, or the incidence of congenital abnormalities in mothers occupationally exposed to vinyl chloride for more than 1 year (Bao et al. 1988).

There are inconsistencies in the developmental toxicity database for the vinyl chloride. In general, vinyl chloride produced minor developmental effects in laboratory animals (i.e., delayed ossification) only at concentrations that were significantly toxic to maternal animals. Maternal toxicity was evident in mice, rats, and rabbits exposed throughout the period of organogenesis. Adverse fetal effects included delayed ossification (all species), increased crown-rump length (mice and rats), and vertebral lumbar spurs (rats). Mice were the most sensitive species investigated (John et al. 1977, 1981). Ungvary et al. (1978) reported a significant increase in resorptions in rats exposed to vinyl chloride during the first trimester of pregnancy. Increased liver-to-body weight ratios were observed in maternal animals exposed during the first and second trimesters, but no histopathologic alterations were found. Continuous exposure of rats to vinyl chloride throughout gestation resulted in decreased fetal weight and increased early postimplantation loss, hematomas, and hydrocephaly with intracerebral hematoma. Weanling rats displayed hepatotoxic effects including decreased bile enzyme activity, decreased bile secretion, decreased cholic acid content, and increased hexobarbital sleep time. No histological data on the livers of pups, or information regarding maternal health were presented (Mirkova et al. 1978).

In contrast with previous studies, no adverse effects were reported in an embryo-fetal developmental toxicity study conducted in rats exposed to similar concentrations of vinyl chloride via inhalation (Thornton et al. 2002). Embryo-fetal developmental parameters including uterine implantation, fetal gender distribution, fetal body weight, and fetal malformations and variations were not affected by vinyl chloride exposure. Vinyl chloride produced a slight decrease in maternal body weight gain at all exposure levels; however, no changes were observed in feed consumption, clinical signs, or postmortem gross findings. Maternal liver and kidney weights were increased relative to total body weight.

The developmental toxicity of vinyl chloride was examined using an *in vitro* whole embryo culture system (Zhao et al. 1996). Vinyl chloride induced embryo growth retardation, but was not shown to be teratogenic in the rat *in vitro* whole embryo culture system.

A study of workers exposed to vinyl chloride in PVC manufacturing plants reported that most workers who presented with scleroderma were shown to have thyroid insufficiency (Suciu et al. 1963). No histopathological effects on the adrenals were reported in guinea pigs exposed to 400,000 ppm for 30 minutes (Mastromatteo et al. 1960). Rats exposed to 30,000 ppm vinyl chloride 5 days/week, 4 hours/day for 12 months, were found to have colloid goiter and markedly increased numbers of perifollicular cells (Viola 1970).

3.7 CHILDREN'S SUSCEPTIBILITY

This section discusses potential adverse 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 and the nature of their response to toxicants. 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 adverse 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).

No studies were located that specifically address the effects of vinyl chloride in children. The effects that have been reported to occur in humans come almost exclusively from studies of workers exposed to high concentrations of vinyl chloride by inhalation. Although the effects observed in human adults could also be observed in children, it is important to note that occupational exposure concentrations are likely to be much greater than environmental levels to which children might be exposed. The toxicological effects reported in adult vinyl chloride workers include cardiovascular, gastric, hematologic, musculoskeletal, hepatic, endocrine, dermal, ocular, immunologic, neurologic, and reproductive effects as well as cancer and death.

Some epidemiologic studies (Infante 1976; Infante et al. 1976a, 1976b; NIOSH 1977) have suggested an association between birth defects and vinyl chloride exposure of the parents of affected children.

However, the design and analysis of these studies has been criticized (Hatch et al. 1981; Stallones 1987). Some inhalation studies with animals have suggested that vinyl chloride is a developmental toxicant (i.e., produces delayed ossification) at doses that also produce maternal toxicity (John et al. 1977, 1981; Mirkova et al. 1978; Sal'nikova and Kotsovskaya 1980; Ungvary et al. 1978). However, no adverse effects on embryo-fetal development were noted in a recent inhalation study in rats conducted using similar concentrations of vinyl chloride (Thornton et al. 2002).

Carcinogenicity studies with animals indicate that some of the adverse health effects of vinyl chloride are dependent on the age of the animal at the time of the exposure. Thus, higher death rates were observed when 2-month-old female hamsters, mice, and rats (equivalent to adolescent humans) were exposed to vinyl chloride in the air for 12 months than when 8- or 14-month-old animals were exposed (Drew et al. 1983). Lifetime cancer risk was also dependent on the age of the animals at the time of exposure to vinyl chloride. The incidence of hemangiosarcoma of the liver, skin, and spleen, and angiosarcoma of the stomach was greater in animals exposed by inhalation for 12 months starting immediately after weaning than in animals that were 1 year older at the time of exposure (Drew et al. 1983). The incidence of mammary gland carcinoma was higher in 2- or 8-month-old hamsters exposed to 200 ppm vinyl chloride for 6 months than in 14- or 20-month-old hamsters exposed to the same concentration and for the same duration (Drew et al. 1983). These results demonstrate the importance of the latency period for vinyl chloride-induced carcinogenesis. Animals that were exposed at a younger age had a longer post-exposure period for the development of tumors. It is difficult to assess the sensitivity of younger animals to cancer in this study because the same exposure concentrations were used for each age group. Exposures were most effective in producing cancer when started early in life (Drew et al. 1983).

Maltoni et al. (1981) evaluated the effect of vinyl chloride dosing on liver carcinogenicity in Sprague-Dawley rats. Rats were exposed to 0, 6,000, or 10,000 ppm vinyl chloride for 100 hours, beginning either at 1 day or at 13 weeks of age. The incidence of angiosarcoma of the liver in newborn rats exposed for only 5 weeks was higher than the incidence observed in rats exposed for 52 weeks beginning at 13 weeks. Hepatoma incidence was approximately 50% in newborn rats exposed for 5 weeks, but did not occur in rats exposed for 52 weeks after maturity. The increased tumor incidence combined with the production additional tumor types (i.e., angiosarcomas and hepatomas) suggest that newborn rats may be more sensitive to vinyl-chloride induced carcinogenicity.

An age-related increase in DNA adduct formation was noted in an inhalation study of lactating rats and their 10-day-old pups exposed to 600 ppm of vinyl chloride, 4 hours/day for 5 days (Fedtke et al. 1990).

Concentrations of two adducts found in liver of pups were 4-fold higher than those found in liver of dams; however, pups were exposed to contaminated breast milk in addition to air concentrations vinyl chloride. In another study, immature rats exposed to vinyl chloride formed 6 times more etheno-nucleosides compared with adults (Ciroussel et al. 1990). The concentration of ethenoguanine adducts was 2–3-fold greater in weanling rats as compared to adult rats exposed at the same dose for the time period (0, 10, 100, or 1,100 ppm, 6 hours/day for 5 days) (Morinello et al. 2002a).

Vinyl chloride induced preneoplastic foci in newborn rats, but not in mature rats (Laib et al. 1985). A study with newborn male or female Wistar rats exposed to 2,000 ppm vinyl chloride indicated that the induction of preneoplastic hepatocellular lesions in rats by vinyl chloride is restricted to an early stage in the life of the animals. The early-life stage sensitivity to the induction of tumors in animals exposed to vinyl chloride appears to be related to the induction by vinyl chloride of hepatic adenosine-5'-triphos-phatase (ATPase) deficient enzyme altered foci, which are putative precursors of hepatocellular carcinoma.

Taken together, the studies cited above suggest an early life stage sensitivity to vinyl chloride carcinogenicity (Cogliano et al. 1996). EPA has recommended an adjustment of the cancer risk estimates to account for early life-stage sensitivity to vinyl chloride (EPA 2000; Ginsberg 2003).

No studies were located that specifically address the toxicokinetics of vinyl chloride in children; however, the toxicokinetic behavior of vinyl chloride in children is expected to be similar to that in adults. An evaluation of pharmacokinetic differences across life stages suggests that the largest difference in pharmacokinetics occurs during the perinatal period (Gentry et al. 2003). The most important factor appears to be the potential for decreased clearance due to immature metabolic enzymes systems; however, an analysis of CYP2E1 levels during development suggests that protein levels and enzyme activity in children between 1 and 10 years old are comparable to adults (EPA 2001). This enzyme is not expressed in the embryonic liver, but rapidly increases during the first 24 hours after birth. Young children appear capable of metabolizing vinyl chloride to reactive intermediates that form DNA adducts that lead to cancer. A PBPK model was also recently applied to evaluate the potential impact of age- and gender-specific pharmacokinetic differences on the dosimetry of vinyl chloride (Clewell et al. 2004). The rate of metabolite production per volume of liver was estimated to rise rapidly from birth until about age 16, after which it remains relatively constant before rising again late in life. The data on CYP2E1 levels in the developing organism suggests that early life stage sensitivity to vinyl chloride-induced cancer is not solely due to an increase in the production of reactive intermediates via this isozyme. Fetal CYP isoforms

may play a role in metabolism of vinyl chloride to reactive intermediates in the fetus and neonate. Glutathione conjugation may also differ in the developing organism. DNA repair capacity and other pharmacodynamic factors may also be associated with an early life stage susceptibility to cancer.

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 or 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 vinyl chloride 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 produced by vinyl chloride 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 Vinyl Chloride

Exposure to vinyl chloride may be monitored to some extent by the identification and quantitation of a number of parameters. For example, following acute exposure to moderate-to-high levels, vinyl chloride can be measured in expired air. The expiration of vinyl chloride follows first-order kinetics; therefore, this parameter may be directly correlated with exposure levels (Baretta et al. 1969). This measure may provide the most direct evidence of vinyl chloride exposure. However, measurement of exposure by this technique is limited by the rapidity of excretion of vinyl chloride in expired air. The half-life of vinyl chloride in expired air has been determined to be between 20 and 30 minutes following an inhalation exposure and to be approximately 60 minutes following oral dosing (Watanabe and Gehring 1976; Watanabe et al. 1976b, 1978a, 1978b). Thus, testing must be initiated as soon as possible following termination of exposure. Furthermore, measurement of vinyl chloride in expired air has limited utility for low-level exposures (<50 ppm) because of competition with absorption and rapid metabolic processes (Baretta et al. 1969). In addition, it provides no information on the duration of exposure.

Thiodiglycolic acid is a major metabolite of vinyl chloride that is excreted in the urine. Measurement of thiodiglycolic acid in urine has been used to monitor workers occupationally exposed to vinyl chloride (Cheng et al. 2001; Müller et al. 1979). However, although this metabolite is used to estimate levels of exposure, the amount of thiodiglycolic acid in the urine varies according to individual metabolic idiosyncracies. Also, metabolism of vinyl chloride to thiodiglycolic acid is a saturable process. Therefore, when exposure exceeds a certain level, the excretion of vinyl chloride as thiodiglycolic acid will plateau (Watanabe and Gehring 1976). Furthermore, the rate of metabolism of vinyl chloride to thiodiglycolic acid may be influenced by the presence of liver disease, ethanol, or certain other substances such as barbiturates (Hefner et al. 1975b) (also see Section 3.4). Similar to the measurement of vinyl chloride in expired air, the measurement of thiodiglycolic acid must take place shortly after exposure because of the rapidity of its excretion. The half-life for excretion of thiodiglycolic acid following an acute exposure is between 4 and 5 hours (Watanabe and Gehring 1976; Watanabe et al. 1978a, 1978b). Cheng et al. (2001) suggests that urinary thiodiglycolic acid levels should not be measured at the end of a

work shift, but are best detected at the beginning of the following work day. Finally, excretion of thiodiglycolic acid is not unique to exposure to vinyl chloride. For example, thiodiglycolic acid may be excreted in the urine as the result of exposure to vinylidene chloride, ethylene oxide, or 2,2-dichloro-ethylether (Norpoth et al. 1986; Pettit 1986). Also, infants delivered prematurely have been found to have high levels of urinary thiodiglycolic acid. A correlation was observed between the thiodiglycolic acid levels and the number of weeks that the infant was born prematurely. The origin of this thiodiglycolic acid is unknown, but is not believed to be associated with vinyl chloride exposure (Pettit 1986).

The intermediary metabolites, 2-chloroethylene oxide and 2-chloroacetaldehyde, bind to macromolecules in the body. 2-Chloroethylene oxide is believed to bind primarily to DNA and RNA, whereas 2-chloroacetaldehyde binds primarily to proteins (Bolt 1986; Guengerich and Watanabe 1979; Guengerich et al. 1979, 1981; Kappus et al. 1976; Watanabe et al. 1978a, 1978b). Two of the DNA adducts that are formed are 1,N^6-etheno-adenosine and 3,N^4-ethenocytidine. Monoclonal antibodies for these DNA adducts have been isolated and used in enzyme-linked immunosorbent assays (ELISA) to quantify these ethenoderivatives in biological samples (Eberle et al. 1989; Young and Santella 1988). Measurement of DNA adducts may be useful in estimating vinyl chloride exposure. However, this technique is of limited value for quantifying levels of exposure because formation of these products will be influenced by variability in vinyl chloride metabolism. Also, their persistence in tissues will be influenced by the rate of DNA metabolism and repair. Furthermore, the DNA adducts, for which monoclonal antibodies have been isolated, are also formed as a result of exposure to vinyl bromide, ethyl carbamate, acrylonitrile, 2-cyanoethylene, and 1,2-dichloroethane (Bolt et al. 1986; Svensson and Osterman-Golkar 1986). See Section 3.4 for additional information on the kinetics of vinyl chloride.

Ethenoguanine adducts have been quantified in human urine using high performance liquid chromatography and tandem mass spectrometry (Gonzalez-Reche et al. 2002). Etheno-adducts are removed from DNA through base excision repair and excreted in the urine where they can be measured using this technique. This method would also include the measurement of endogenously formed etheno-adducts; thus, it is critical to determine the background level of urinary adducts in a control population.

Vinyl chloride-induced genetic alterations have been identified in the Ki-*ras* oncogene and the p53 tumor suppressor gene, and oncoproteins and p53 antibodies have been detected in the serum of cancer patients with angiosarcoma (see Section 3.3). Immunological techniques have been used to detect the presence of Asp13p21 (oncoprotein for mutation of the Ki-*ras* gene), p53 mutant protein, and p53 antibodies in the serum of exposed workers (Brandt-Rauf et al. 2000a, 2000b; Marion 1998). Statistical analyses suggest a

relationship between vinyl chloride exposure and the presence of these serum biomarkers; however, the predictive value of these biomarkers for development of cancer is not known.

The micronucleus assay, performed using peripheral lymphocytes of 32 vinyl chloride workers, was used to indicate the time elapsed since the last vinyl chloride exposure occurred (Fucic et al. 1994, 1997). The study showed a decrease in the frequency of micronuclei and mitotic activity in proportion to the length of the interval after the last vinyl chloride exposure. For the group with 10 years of employment, the percentage of micronuclei decreased from 12.82 when exposure occurred on the day of blood sampling to 3.16 when the last exposure occurred 90 days before blood sampling (Fucic et al. 1994). Similar changes were noted when the mean duration of employment was 5 years. However, this use of the micronucleus assay must take into account the total duration of exposure. Micronucleus frequency was shown to be several times higher in binucleated lymphocytes as compared to mononuclear lymphocytes in 25 workers exposed to vinyl chloride for an average of 10 years (Fucic et al. 2004).

Exposure to vinyl chloride may also be estimated to some extent by the presence of certain symptoms known to be closely associated with vinyl chloride exposure. The exposure may have occurred even if the symptoms were not found upon examination, but their presence could be indicative of exposure. For example, a syndrome known as vinyl chloride disease has been identified in workers occupationally exposed to vinyl chloride. This syndrome includes Raynaud's phenomenon, acroosteolysis of the distal phalanges of the fingers, and scleroderma-like changes in the hands and forearms (also see Section 3.2). Although this syndrome resembles systemic sclerosis, a differential diagnosis may be made based on the absence of antinuclear antibodies from the blood of those afflicted with vinyl chloride disease (Black et al. 1983, 1986). The occurrence of vinyl chloride disease in highly exposed worker populations is about 3%, and susceptibility appears to be genetically related (Black et al. 1983, 1986). Symptoms of vinyl chloride disease are unlikely to occur in hazardous waste site conditions because of predicted low levels of exposure. Absence of these symptoms would not eliminate the possibility of exposure, but their presence may be a good indicator of exposure.

Angiosarcoma of the liver has been identified in workers occupationally exposed to vinyl chloride. This type of tumor is extremely rare in the general population (Heath et al. 1975); therefore, its diagnosis may indicate vinyl chloride exposure. However, other causes of angiosarcoma such as exposure to arsenicals and Thorotrast (thorium dioxide; formerly used in arteriography) should be considered as possible causative factors, if present, before correlating hepatic angiosarcoma with vinyl chloride exposure (Gedigke et al. 1975; Marsteller et al. 1975). Their elimination may depend upon such factors as the

magnitude of the vinyl chloride exposure and the frequency of the other causes of angiosarcoma in the population.

3.8.2 Biomarkers Used to Characterize Effects Caused by Vinyl Chloride

The realization that angiosarcoma of the liver is associated with vinyl chloride exposure prompted several investigators to try to identify assays that could be used to monitor those individuals considered to be at risk. Standard serum assays designed to detect the presence of hepatic enzymes in the blood were found to be of limited value in monitoring the progression of vinyl chloride-induced hepatic changes (Berk et al. 1975; Liss et al. 1985; Vihko et al. 1984). This may be because of the extent of hepatic damage produced by vinyl chloride and the late development of necrotic areas in the disease process (Popper et al. 1981). In contrast, studies indicate that clearance type assays, which measure liver function, are more sensitive indicators of the hepatic damage resulting from vinyl chloride exposure. These assays include the indocyanine clearance test, measurement of serum bile acids, and measurement of serum hyaluronic acid concentration (Berk et al. 1975; Liss et al. 1985; McClain et al. 2002; Vihko et al. 1984).

Liver biopsy may provide the most accurate identification of vinyl chloride-associated liver damage (Liss et al. 1985). This is because of the characteristic pattern of hepatic histopathology associated with vinyl chloride-induced damage (Popper et al. 1981). However, liver biopsy is an invasive procedure with attendant risks and, therefore, may not be justified.

Individual exposure to vinyl chloride has been linked to angiosarcoma and benign angiomatous lesions based on the monitoring of serum found to be positive for the presence of the mutant protein Asp 13 c-Ki-*ras* p21, which was not present in control individuals (DeVivo et al. 1994). Additionally, this protein was found in the serum of 49% of exposed workers who had no apparent liver lesions. It may be possible to utilize the presence of this mutant protein for the early detection of angiosarcoma of the liver.

Use of enzyme-linked immunoassay (EIA) to detect anti-p53 antibodies in serum of individuals exposed to vinyl chloride may provide an early method of screening for angiosarcoma of the liver (Trivers et al. 1995). Detection of serum anti-p53 antibodies has occurred in some, but not all, individuals exposed to vinyl chloride who later developed angiosarcoma of the liver (Trivers et al. 1995). However, not all individuals who developed angiosarcoma of the liver tested positive for anti-p53 antibodies. Also, anti-p53 antibodies are not specific to angiosarcoma of the liver but can be detected in the sera of patients with

other types of cancers such as leukemia; childhood lymphoma; breast, lung, and colon cancer; and hepatocellular carcinoma.

The symptoms and signs associated with vinyl chloride disease (Raynaud's phenomenon, scleroderma-like skin changes, and acroosteolysis) are similar to those observed in systemic sclerosis. Vinyl chloride disease may be differentiated from systemic sclerosis by the absence of antinuclear antibodies in the blood and association of vinyl chloride disease with vinyl chloride exposure (Black et al. 1983, 1986). Raynaud's phenomenon is an early symptom of vinyl chloride disease. However, cyanosis and blanching of fingers with exposure to cold may be the result of a number of other conditions such as connective tissue disorders, mechanical arterial obstruction, hyperviscosity of the blood, or exposure to drugs, chemicals, or vibrating tools (Freudiger et al. 1988). Thus, other potential causes must be eliminated before this syndrome can be used to identify vinyl chloride disease. The symptoms associated with vinyl chloride disease have been attributed to vinyl chloride-induced changes in the microvasculature (Grainger et al. 1980). Capillary abnormalities in the hands may be detected using wide-field capillary microscopy and have been proposed to represent an early manifestation of the effects of vinyl chloride (Maricq et al. 1976). Also, immunofluorescent examination of biopsy material from the skin may be used to identify circulating immune complexes and their deposition on the vascular endothelium (Ward 1976).

Chromosomal aberrations found in lymphocytes may be indicative of the genotoxic effects of vinyl chloride (Anderson 2000; Anderson et al. 1980; Ducatman et al. 1975; Fucic et al. 1990a, 1990b, 1992; Funes-Cravioto et al. 1975; Garaj-Vrhovac et al. 1990; Hansteen et al. 1978; Hrivnak et al. 1990; Kucerova et al. 1979; Purchase et al. 1978; Sinues et al. 1991). However, any of a number of genotoxic substances can produce chromosomal aberrations. Also, de Jong et al. (1988) have found that variability in the control population may obscure the observation of chromosomal aberrations in persons exposed to low levels of vinyl chloride. G-banding analysis appeared to provide a more sensitive indication of chromosomal alteration than sister chromatid exchanges (Zhao et al. 1996). DNA damage in lymphocytes can be directly assessed using a single-cell gel electrophoresis technique. The severity of the damage may correlate with the duration of exposure (Awara et al. 1998). The DNA adducts produced by the reactive intermediary metabolites of vinyl chloride, including 1,N^6-ethenoadenosine and 3,N^4-ethenocytidine, may be more specific indicators of vinyl chloride's genotoxic potential.

3.9 INTERACTIONS WITH OTHER CHEMICALS

A number of studies have been performed that examine the effect of agents intended to alter the metabolism of vinyl chloride on its toxicity. For example, the effects of phenobarbital pretreatment on vinyl chloride-induced hepatotoxicity have been examined by Jaeger et al. (1974, 1977), Jedrychowski et al. (1985), and Reynolds et al. (1975a, 1975b). Pretreatment of rats with phenobarbital for 7 days prior to a 4-hour vinyl chloride exposure produced an increase in microsomal cytochrome P-450 activity (Reynolds et al. 1975b) and enhanced hepatotoxicity (Jaeger et al. 1974, 1977; Jedrychowski et al. 1985; Reynolds et al. 1975a, 1975b). In these studies, in the absence of the phenobarbital pretreatment, a single exposure to approximately 50,000 ppm had no detectable adverse effect on the livers of exposed rats. However, following phenobarbital pretreatment, 50,000 ppm of vinyl chloride produced increased serum activity of hepatic enzymes (Jaeger et al. 1977; Jedrychowski et al. 1985), areas of hepatic necrosis (Reynolds et al. 1975a), or both (Jaeger et al. 1974; Reynolds et al. 1975b).

Another agent known to increase MFO activity, Aroclor 1254, was also tested for its ability to enhance vinyl chloride-induced hepatotoxicity (Conolly and Jaeger 1979; Conolly et al. 1978; Jaeger et al. 1977; Reynolds et al. 1975b). Pretreatment of rats with Aroclor 1254 for several days prior to exposure to vinyl chloride resulted in an increase in serum activity of hepatic enzymes (Conolly and Jaeger 1979; Conolly et al. 1978; Jaeger et al. 1977; Reynolds et al. 1975b) and areas of hepatic necrosis (Conolly et al. 1978; Reynolds et al. 1975b).

Additional support for a role for MFO in the enhanced toxicity of vinyl chloride was obtained using SKF525A, an MFO inhibitor. If SKF525A was administered following phenobarbital pretreatment and before vinyl chloride exposure, it blocked the ability of phenobarbital pretreatment to enhance vinyl chloride-induced hepatotoxicity (Jaeger et al. 1977).

The role of glutathione conjugation in vinyl chloride-induced toxicity was also examined (Conolly and Jaeger 1979; Jaeger et al. 1977). The investigators hypothesized that depletion of glutathione might enhance the toxicity of vinyl chloride by preventing the excretion of toxic intermediary metabolites. However, diethylmaleate, an agent known to deplete hepatic glutathione levels, had no effect on the toxicity produced by vinyl chloride following pretreatment with either phenobarbital (Jaeger et al. 1977) or Aroclor 1254 (Conolly and Jaeger 1979). Trichloropropene oxide (TCPO), another agent known to deplete hepatic glutathione, produced enhancement of the hepatic toxicity produced by Aroclor 1254 pretreatment and vinyl chloride exposure but only when the animals had been fasted prior to vinyl

chloride exposure (Conolly and Jaeger 1979). The study authors hypothesized that the enhancement of vinyl chloride toxicity was a result of the ability of TCPO to inhibit epoxide hydrase rather than its ability to deplete glutathione levels. The lack of the effect of glutathione depletion indicates that the glutathione pathway is not very important at normal levels of exposure.

Although the depletion of cellular glutathione levels did not appear to enhance vinyl chloride toxicity, treatment with cysteine, the rate-limiting precursor in hepatic glutathione synthesis, increased hepatic glutathione levels and provided partial protection against the toxic effects produced by Aroclor 1254 and vinyl chloride (Conolly and Jaeger 1979).

The effects of the interaction of ethanol with vinyl chloride on development were tested by John et al. (1977). In this study, animals were exposed to vinyl chloride in the presence and absence of 15% ethanol in the drinking water during pregnancy. Ethanol produced a decrease in maternal food consumption and maternal weight gain in mice, rats, and rabbits and enhanced incidence of skeletal abnormalities in mice, and to a lesser extent, in rats. Interpretation of these results is clouded by the absence of an ethanol-exposed control group and the current recognition of the adverse effects of ethanol on pregnancy outcome.

In the experiment by Radike et al. (1981), ethanol-consuming rats exposed to vinyl chloride for a year had an enhanced incidence of hepatic angiosarcomas, hepatomas, and lymphosarcomas, earlier onset of the tumors, and an enhanced death rate. The incidence of vinyl chloride-induced angiosarcomas was potentiated by ethanol, whereas the increased incidences of hepatoma and lymphosarcomas by ethanol were additive in nature.

The effects of smoking on chromosomal aberrations in vinyl chloride-exposed workers was examined by Hrivnak et al. (1990), who found no effect of smoking in 43 workers exposed for an average of 11.2 years to levels of vinyl chloride ranging from 0.8 to 16 ppm. Most cytogenetic studies of the effects of smoking in humans have reported no effect on chromosomal aberrations, although the sister chromatid exchange frequency is usually elevated (Wong et al. 1998).

A study that examined the interaction between vinyl chloride and trichloroethylene using both inhalation exposures of rats and pharmacokinetic modeling found that trichloroethylene exposure inhibited vinyl chloride in a competitive manner (Barton et al. 1995). This interaction was observed only at high

concentrations (both chemicals >10 ppm), and the study authors concluded that the interaction is not likely to be important for environmental exposures.

3.10 POPULATIONS THAT ARE UNUSUALLY SUSCEPTIBLE

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

Data suggest that the following subsets of the human population may be unusually susceptible to the toxic effects of vinyl chloride: fetuses; infants; young children; people with liver disease, irregular heart rhythms, impaired peripheral circulation, or systemic sclerosis; people with exposure to organochlorine pesticides; and people consuming ethanol or barbiturates or taking Antabuse for alcoholism. Also, persons who possess the HLA-DR5, HLA-DR3, and B8 alleles may be at increased risk.

Vinyl chloride can cross the placenta and enter the blood of the fetus (Ungvary et al. 1978). Studies by Drew et al. (1983), John et al. (1977, 1981), and Maltoni et al. (1981) have shown that animals exposed by inhalation prior to adolescence or during pregnancy may have a greater death rate and increased likelihood of developing cancer than adult animals exposed for similar periods. This may relate to the length of the induction period of hepatic angiosarcoma rather than to an increased susceptibility of the young, *per se*. It is also possible that there are explanations for these findings. Cogliano and Parker (1992) suggested that in the multistage model of carcinogenesis, carcinogens that induce an initial transition early in the life of an animal would be more effective since there would be a longer period of time remaining in the lifespan for completion of the remaining transitions. Their empirical model of the effect of age at exposure on the development of cancer suggests that there is an age-sensitive period of exposure to vinyl chloride.

Vinyl chloride is metabolized in the liver in a multistep process. The intermediary metabolites of vinyl chloride, 2-chloroethylene oxide and 2-chloroacetaldehyde, have been suggested to be responsible for some of the adverse effects produced by vinyl chloride. Thus, activation of the enzyme system

responsible for production of these toxic metabolites would be expected to increase the toxicity of vinyl chloride exposures. 2-Chloroethylene oxide is formed by action of the MFO system associated with cytochrome P-450. The barbiturate, phenobarbital, and the pesticide extender, Aroclor 1254, increased MFO activity and have been shown to greatly increase the hepatotoxicity of vinyl chloride (Conolly and Jaeger 1979; Conolly et al. 1978; Jaeger et al. 1974, 1977; Jedrychowski et al. 1985; Reynolds et al. 1975a, 1975b). Thus, persons taking barbiturates or who might be exposed to organochlorine pesticides that are known to induce microsomal enzymes (such as Aroclor 1254) would be expected to be at increased risk for developing vinyl chloride-induced hepatotoxicity.

Genetic polymorphisms related to vinyl chloride metabolism and DNA repair may increase the susceptibility of individuals to liver toxicity and cancer. CYP2E1 and glutathione *S*-transferase genotypes were associated with abnormal liver function, "vinyl chloride disease", and the incidence of angiosarcoma in exposed workers (El Ghissassi et al. 1995; Green et al. 2000; Huang et al. 1997). Genotypes for CYP2E1, the DNA repair gene, x-ray repair cross-complementing group 1 (XRCC1), and aldehyde dehydrogenase 2 (ALDH2) have been associated with increased sister chromatid exchange frequency and increased expression of p53 mutant protein and anti-p53 antibody in exposed workers (Li et al. 2003; Wong et al. 1998, 2002b, 2003b). The risk of developing liver cancer also appears elevated in those with a history of Hepatitis B viral infection (Du and Wang 1998; Wong et al. 2003b).

Radike et al. (1981) demonstrated that ethanol-consuming rats exposed to vinyl chloride had an increased incidence of cancer and an earlier death rate than animals exposed to vinyl chloride in the absence of ethanol.

Some persons consume the agent, Antabuse, to curb the desire for alcohol. In its role as a therapeutic agent, Antabuse blocks aldehyde dehydrogenase and causes a build-up of acetaldehyde, which is emetic, in the body when alcohol is consumed. If persons taking Antabuse are exposed to vinyl chloride, the alternative metabolic pathway for vinyl chloride metabolism will be blocked, causing more vinyl chloride to be metabolized to the toxic metabolite, 2-chloroethylene oxide. Thus, these persons may be at increased risk for hepatotoxicity, cancer, and death at an early age.

Very high levels of vinyl chloride have been demonstrated to cause cardiac arrhythmias in dogs (Carr et al. 1949; Oster et al. 1947). Persons with a propensity to develop cardiac arrhythmias because of heart disease or damage may be at an increased risk of having heart beat irregularities when exposed to high concentrations of vinyl chloride.

Vinyl chloride has been shown to produce decreased circulation in the hands and fingers of some people. Persons with impaired circulation due to some other cause such as connective tissue disorders, systemic sclerosis, hyperviscosity of the blood, or use of vibrating tools, may experience more severe impairment of the circulation.

Work by Black et al. (1983, 1986) has shown that persons with the HLA allele HLA-DR5 may have an increased likelihood of developing vinyl chloride disease, and those with the alleles HLA-DR3 and B8 may have an increased severity of the disease.

3.11 METHODS FOR REDUCING TOXIC EFFECTS

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

Bronstein AC, Currance PL, eds. 1988. Emergency care for hazardous materials exposure. St. Louis, MO: CV Mosby Company, 143-144.

Haddad LM, Winchester JF, eds. 1990. Clinical management of poisoning and overdose. Philadelphia, PA: W.B. Saunders Company, 516, 1209, 1214, 1224, 1227-1229.

Stutz DR, Ulin S, eds. 1992. Hazardous materials injuries. A handbook for pre-hospital care. 3rd ed. Beltsville, MD: Bradford Communications Corporation, 286-287.

3.11.1 Reducing Peak Absorption Following Exposure

Limited information from humans and results from animal studies indicate that vinyl chloride is rapidly and virtually completely absorbed following inhalation and oral exposure, but animal studies suggest that dermal absorption of vinyl chloride gas is not likely to be significant (see Section 3.3.1). Efforts to reduce absorption following acute exposure to vinyl chloride should focus on removing the individual from the site of exposure and decontaminating exposed areas of the body. Vinyl chloride gas is relatively dense and accumulates at ground level. Therefore, the subject should be moved from low-lying areas.

Contaminated skin may be washed with soap and water; however, this will most likely not prevent tissue damage produced by frostbite from the cooling caused by the rapid evaporation of vinyl chloride from the skin. It is suggested that eyes exposed to vinyl chloride be copiously irrigated with water or normal saline (Bronstein and Currance 1988; Haddad and Winchester 1990; Stutz and Ulin 1992). Because of its volatility, it is unlikely that vinyl chloride would be ingested unless it had been dissolved in a solvent. If such ingestion of vinyl chloride occurs, it is suggested that water or milk be administered for dilution if the patient can swallow, has a good gag reflex, and is not drooling (Bronstein and Currance 1988; Stutz and Ulin 1992). In addition, gastric lavage and administration of activated charcoal have been suggested as a means to reduce absorption of vinyl chloride. Induction of emesis is contraindicated (Bronstein and Currance 1988; Haddad and Winchester 1990; Stutz and Ulin 1992).

3.11.2 Reducing Body Burden

Because of its rapid metabolism and excretion, vinyl chloride does not tend to accumulate in the body. As discussed in Section 3.4.3, the metabolism of vinyl chloride is a dose-dependent, saturable process. Vinyl chloride is oxidized primarily by the microsomal MFO system (cytochrome P-450) to a reactive epoxide intermediate (2-chloroethylene oxide), which can rearrange to 2-chloroacetaldehyde or conjugate with glutathione to form *S*-formylmethyl glutathione. At exposure concentrations below about 1,000 ppm in air, very little vinyl chloride is excreted unchanged in the exhaled air. However, when metabolic saturation occurs at high exposure concentrations (approximately 1,000 ppm following inhalation exposure in rats [Watanabe and Gehring 1976; Watanabe et al. 1976b] and approximately 20 mg/kg following oral administration to rats [Green and Hathway 1975; Watanabe and Gehring 1976; Watanabe et al. 1976a]), vinyl chloride is excreted unchanged in expired air. Therefore, a possible means to enhance the elimination of vinyl chloride without allowing its biotransformation to toxic intermediates is to saturate this oxidative pathway by administration of substances known to be metabolized via this route. Saturation of the P-450 system may occur with drugs such as phenytoin or dicumerol (Goodman and Gilman 1980). However, the effectiveness of these agents in blocking the P-450 metabolism of vinyl chloride has not been tested, and it is unclear whether toxic doses would be necessary to overcome the relative affinities of the enzymes for vinyl chloride versus these agents. In addition, the potential toxicity of any side products of these substances would need to be considered in any protocol. Several agents induce activity of the microsomal enzymes and could potentially increase the toxicity of vinyl chloride. Administration of such substances would be contraindicated.

3.11.3 Interfering with the Mechanism of Action for Toxic Effects

Following acute, high-level exposure, vinyl chloride behaves as an anesthetic and produces central nervous system and respiratory depression (see Sections 3.2.1.4). Therefore, basic life support measures, such as supplemental oxygen and cardiopulmonary resuscitation, are suggested in such instances (Bronstein and Currance 1988; Haddad and Winchester 1990; Stutz and Ulin 1992). In addition, like other halogenated hydrocarbons, vinyl chloride may sensitize the heart to the effects of circulating catecholamines. Therefore, the patient's cardiac rhythm should be monitored, and the use of isoproterenol, epinephrine, or other sympathomimetic drugs should be avoided (Haddad and Winchester 1990).

Vinyl chloride is a known human and animal carcinogen; long-term exposure to this compound is associated with an increased incidence of hepatic angiosarcomas (see Section 3.2.1.7). Vinyl chloride is also hepatotoxic. The mechanism by which vinyl chloride induces its carcinogenic and toxic effect on the liver has been well studied. A reactive epoxide intermediate of vinyl chloride, 2-chloroethylene oxide, interacts directly with DNA and RNA producing cyclic etheno-adducts that include 1,N^6-ethenoadenine, 3,N^4-ethenocytosine, N^2,3-ethenoguanine, and 1,N^2-ethenoguanine. This alkylation results in highly efficient base-pair substitution, leading to neoplastic transformation (see Section 3.5.2). As discussed above, this epoxide intermediate is formed when vinyl chloride is oxidized by the P-450 isoenzymes. Interference with this metabolic pathway, therefore, could reduce the toxic and carcinogenic effects of vinyl chloride by reducing the amount of epoxide produced. A number of drugs, such a cobaltous chloride, SKF-535-A, and 6-nitro-1,2,3-benzothioadiazole, have been reported to inhibit P-450 enzymes. Pretreatment with 6-nitro-1,2,3-benzothioadiazole completely blocked the metabolism of vinyl chloride in rats exposed to 0.45 ppm in a closed system for 5 hours (Bolt et al. 1977). P-450 metabolism also results in products that can be more readily eliminated than can the parent compound. Hence, any side products of the drugs and their potential to increase the biological half-life of vinyl chloride would also need to be considered in any protocol. In fact, a study by Buchter et al. (1977) showed that substantial unmetabolized vinyl chloride accumulated in fatty tissue when 6-nitro-1,2,3-benzothioadiazole was used to block P-450 metabolism. The study did not examine the fate of vinyl chloride in fatty tissue after P-450 metabolism was reactivated, but it is likely that vinyl chloride would leave the fat slowly and be metabolized. Thus, while P-450 metabolism would probably reduce the generation of toxic metabolites in the short term, it is unclear whether the generation of toxic metabolites could be completely avoided. Further research to determine which isozymes are involved in the metabolism to the reactive intermediates, as well as which isozymes are involved in enhancing the elimination of vinyl chloride,

could lead to the development of strategies to selectively inhibit specific isozymes and thus reduce the toxic effects of vinyl chloride.

Because vinyl chloride is detoxified by conjugation with glutathione and/or cysteine (see discussion above and Section 3.4.3), ensuring sufficient glutathione stores in the body (e.g., by treatment with *N*-acetyl cysteine) may reduce the possibility of toxic effects following acute exposure to vinyl chloride.

Vinyl chloride disease has been reported in a small percentage of workers exposed to this compound. One of the symptoms of this condition is Raynaud's phenomenon (blanching, numbness, and discomfort of the fingers upon exposure to cold). Studies of these individuals demonstrated that vinyl chloride may produce blockage of the blood vessels supplying the hand, hypervascularity, and a thickening of the blood vessel walls (Harris and Adams 1967; Preston et al. 1976; Veltman et al. 1975; Walker 1976). Several investigators have suggested that the mechanism for vinyl chloride disease may be an autoimmune response similar to systemic sclerosis. Grainger et al. (1980) and Ward (1976) proposed that a reactive vinyl chloride intermediate metabolite, such as 2-chloroethylene oxide or 2-chloroacetaldehyde, binds to a protein such as IgG. The altered protein initiates an immune response, with deposition of immune products along the vascular endothelium. Cold temperatures could produce the precipitation of these immune complexes resulting in blockage of the blood vessels. Another characteristic of vinyl chloride disease is acroosteolysis, in which the terminal phalanges of the fingers are resorbed. This condition has been noted predominantly in workers who first had Raynaud's phenomenon (Dinman et al. 1971; Freudiger et al. 1988; Harris and Adams 1967; Magnavita et al. 1986; Markowitz et al. 1972; Preston et al. 1976; Sakabe 1975; Veltman et al. 1975; Wilson et al. 1967). The resorptive bone changes may be due to activation of osteoclasts secondary to vascular insufficiency in the finger tips, but this remains to be demonstrated conclusively. Other manifestations of vinyl chloride disease include joint and muscle pain, enhanced collagen deposition, stiffness of the hands, and scleroderma-like skin changes. Increased levels of circulating immune complexes and immunoglobulins have been observed in vinyl chloride workers, suggesting a stimulatory effect of vinyl chloride on the immune system (Bogdanikowa and Zawilska 1984). A correlation between the severity of the symptoms of vinyl chloride disease and the magnitude of the immune response was observed (Grainger et al. 1980; Langauer-Lewowicka et al. 1976; Ward 1976). Research on the genetic characteristics of workers with this disease has demonstrated that the susceptibility to vinyl chloride disease was increased in the presence of the HLA-DR5 allele or a gene in linkage disequilibrium with it, and progression of the disease to its more severe forms was favored by HLA-DR3 and B8 (Black et al. 1983, 1986). If vinyl chloride disease is mediated by an immune mechanism in individuals with a genetic predisposition, then the effects of this disease may be mitigated

by administration of drugs used to treat other similar autoimmune diseases (e.g., azathioprine, cyclophosphamide, and prednisone). However, the toxicity associated with the use of these drugs must also be considered.

3.12 ADEQUACY OF THE DATABASE

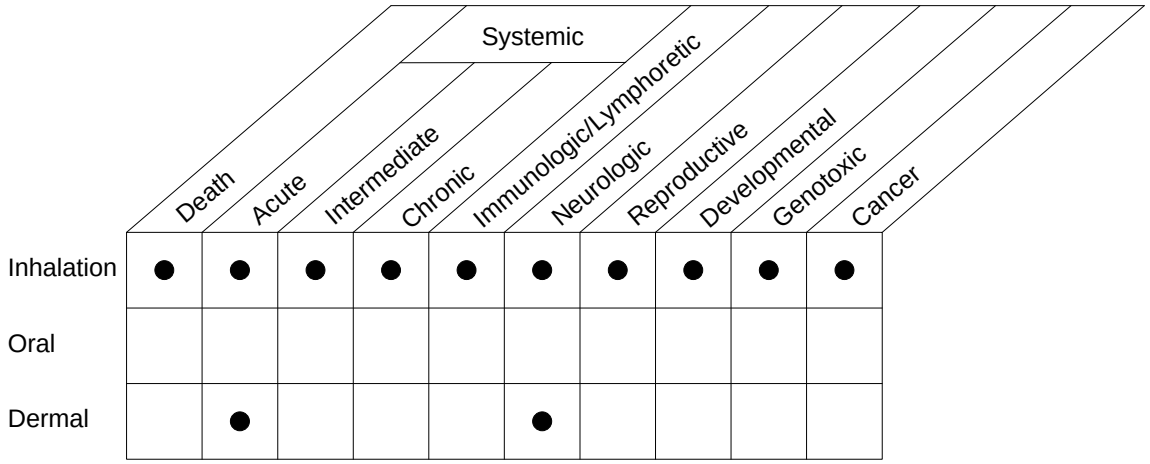
Section 104(i)(5) of CERCLA, as amended, directs the Administrator of ATSDR (in consultation with the Administrator of EPA and agencies and programs of the Public Health Service) to assess whether adequate information on the adverse health effects of vinyl chloride is available. Where adequate information is not available, ATSDR, in conjunction with the National Toxicology Program (NTP), is required to ensure the initiation of a program of research designed to determine the adverse health effects (and techniques for developing methods to determine such adverse health effects) of vinyl chloride.

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.

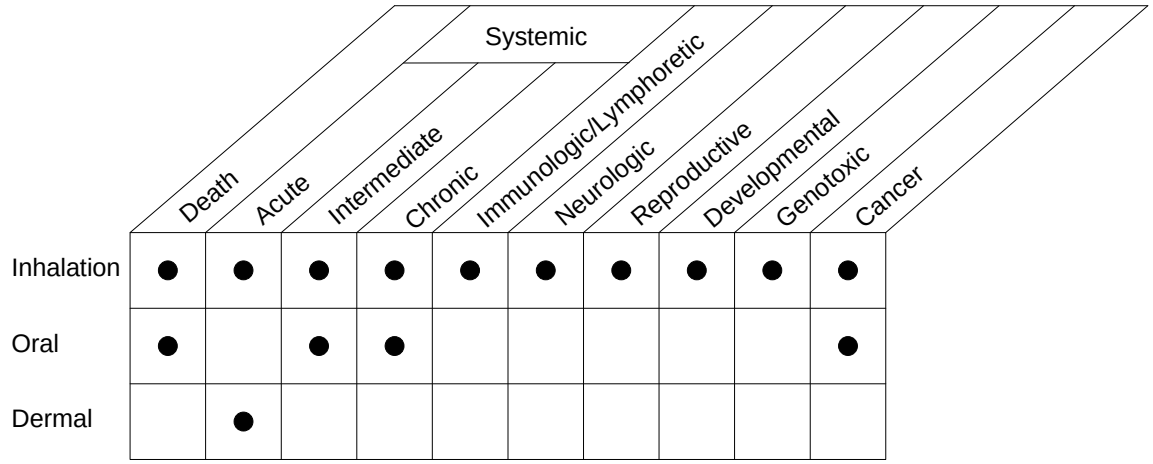
3.12.1 Existing Information on Adverse Health Effects of Vinyl Chloride

The existing data on health effects of inhalation, oral, and dermal exposure of humans and animals to vinyl chloride are summarized in Figure 3-5. The purpose of this figure is to illustrate the existing information concerning the adverse health effects of vinyl chloride. 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.

Figure 3-5. Existing Information on Health Effects of Vinyl Chloride



Human



Animal

● Existing Studies

Virtually all of the literature regarding adverse health effects in humans comes from studies of workers exposed to vinyl chloride during the production of PVC. Case reports and cohort studies describe some acute health effects and a wide range of long-term health effects. The predominant mode of exposure in these studies is via inhalation. These studies are limited by the lack of reliable data on individual exposure levels. No studies were found regarding the adverse health effects of oral exposure. One case report examined the effects of dermal exposure to liquid vinyl chloride, but exposure by this route is not expected to contribute significantly to producing adverse health effects because of the limited absorption of vinyl chloride through the skin.

A large number of studies examining the adverse health effects of inhaled vinyl chloride in animals were reviewed. As can be seen in Figure 3-5, no information is available on acute adverse systemic effects, immunologic, neurologic, reproductive, developmental, or genotoxic effects of exposure of animals by the oral route. One study examined the effects of dermal/ocular exposure to vinyl chloride gas, but toxicokinetic studies indicate that this route is not an important means of exposure.

3.12.2 Identification of Data Needs

Acute-Duration Exposure. Populations in areas that contain hazardous waste sites may be exposed to vinyl chloride for brief periods. Exposure most likely would occur by inhalation, but relatively brief oral and dermal exposures are also possible. There are acute inhalation exposure data in humans and animals that indicate that the central nervous system is a major target organ of vinyl chloride toxicity.

Symptoms of central nervous system depression ranging from dizziness and drowsiness to loss of consciousness have been observed in humans and animals as a result of brief exposure to very high levels of vinyl chloride (Hehir et al. 1981; Jaeger et al. 1974; Lester et al. 1963; Mastromatteo et al. 1960; Patty et al. 1930). A threshold for central nervous system effects appears to be approximately 8,000 ppm (Lester et al. 1963). Extremely high concentrations of vinyl chloride produce respiratory irritation and death in humans and animals by the inhalation route (Danziger 1960; Lester et al. 1963; Mastromatteo et al. 1960; Patty et al. 1930). Based on studies in animals, the threshold for these effects appears to be in the range of 100,000–400,000 ppm (Lester et al. 1963; Mastromatteo et al. 1960; Patty et al. 1930). However, increased rate of death was noted in pregnant mice at 500 ppm (John et al. 1977, 1981). Extremely high concentrations of vinyl chloride produced cardiac arrhythmias in dogs exposed by the inhalation route (Carr et al. 1949; Oster et al. 1947). Although no threshold was reported for these effects, concentrations of this magnitude would not likely be encountered by humans. Pharmacokinetic data indicate that similar end points might be expected if sufficiently high doses could be consumed by

the oral route. However, the solubility characteristics of vinyl chloride in aqueous media (1,100–2,763 mg/L at 25 °C) (Cowfer and Magistro 1983; EPA 1985b) indicate that achieving concentrations of vinyl chloride in excess of 5,000 ppm may be extremely difficult. Animal studies indicate that acute inhalation exposures to vinyl chloride can produce developmental effects at concentrations that also cause significant maternal toxicity (John et al. 1977, 1981; Ungvary et al. 1978). Concentrations of 500 ppm were observed to produce delayed ossification in the fetus and decreased food consumption, body weight gain, and increased rate of mortality in maternal mice (John et al. 1977, 1981). The NOAEL (50 ppm) in this study was used to derive an acute-duration inhalation MRL. Animal studies examining the developmental, neurological, and systemic effects of the highest doses achievable in drinking water would be helpful for determining whether any effects would occur when vinyl chloride-contaminated groundwater or food products are consumed. One report described severe frostbite with second degree burns on the hands of a man resulting from the rapid evaporation of spilled liquid vinyl chloride (Harris 1953). A toxicokinetic study using two monkeys indicates that absorption of vinyl chloride by the dermal route is exceedingly small (Hefner et al. 1975a); thus, studies examining the effects of acute-duration dermal exposure do not seem warranted. However, if further toxicokinetics studies contradict these findings, acute-duration dermal exposure studies in animals may be valuable.

A report was located regarding adverse hepatic and respiratory effects observed 18 months following a single 1-hour inhalation exposure to vinyl chloride (Hehir et al. 1981). However, limitations in the study diminished its reliability. Because of the implications of adverse chronic effects from acute exposure, confirmation of these results in another study would be valuable.

Intermediate-Duration Exposure. No studies in humans specifically address intermediate-duration effects by any route. Most epidemiological studies of occupationally exposed persons have concentrated on persons who have been employed over several years. A study with reliable quantification of exposure levels that examined the effects experienced by vinyl chloride workers in their first year of exposure would be helpful for predicting the effects that might be observed in populations exposed to hazardous waste sites for similar periods of time. However, at current low levels of exposure in the workplace, it may be difficult to demonstrate effects. There is a large database describing the effects of intermediate-duration inhalation exposures in animals (Adkins et al. 1986; Bi et al. 1985; Drew et al. 1983; Du et al. 1979; Feron et al. 1979a, 1979b; Hong et al. 1981; Lee et al. 1978; Lester et al. 1963; Maltoni et al. 1981; Mirkova et al. 1978; Sal'nikova and Kotsovskaya 1980; Schaffner 1978; Sharma and Gehring 1979; Short et al. 1977; Sokal et al. 1980; Suzuki 1978, 1981; Thornton et al. 2002; Torkelson et al. 1961; Wisniewska-Knypl et al. 1980). Animals exposed to vinyl chloride for more than 2 weeks and less than a

year have experienced effects on the liver, kidneys, lungs, and blood (Bi et al. 1985; Du et al. 1979; Feron et al. 1979a, 1979b; Lester et al. 1963; Sal'nikova and Kotsovskaya 1980; Schaffner 1978; Sokal et al. 1980; Torkelson et al. 1961; Wisniewska-Knypl et al. 1980). Data were sufficient to determine an intermediate-duration inhalation MRL based on adverse liver effects in rats. The MRL was based on a NOAEL of 15 ppm for hepatic centrilobular hypertrophy (Thornton et al. 2002). Extremely limited information was available regarding oral intermediate-duration effects. One chronic study presented interim sacrifice data that identified relative weight and histopathological changes in the liver (Feron et al. 1981). However, only a single-dose group was compared to controls, precluding determination of the dose-response of the effects observed. Thus, no MRL for oral intermediate-duration exposures could be determined. Additional studies examining the effects of oral exposure to vinyl chloride would be helpful for evaluating relevant biomarkers of exposure and effects in humans consuming contaminated drinking water or foods (see Section 3.8). As noted above, absorption of vinyl chloride through the skin is not expected to be significant (Hefner et al. 1975a); thus, additional dermal exposure studies do not seem warranted. However, if further toxicokinetics studies contradict these findings, other intermediate-duration dermal exposure studies may be valuable.

Chronic-Duration Exposure and Cancer. A large number of studies of workers exposed to vinyl chloride have identified a wide range of target organs that may be affected by chronic-duration inhalation of vinyl chloride (Bao et al. 1988; Bencko et al. 1988; Berk et al. 1975; Black et al. 1983, 1986; Bogdanikowa and Zawilska 1984; Brugnami et al. 1988; Byren et al. 1976; Creech and Johnson 1974; Dinman et al. 1971; Falk et al. 1974; Freudiger et al. 1988; Fucic et al. 1995; Gedigke et al. 1975; Grainger et al. 1980; Harris and Adams 1967; Jayson et al. 1976; Jones and Smith 1982; Langauer-Lewowicka et al. 1976; Laplanche et al. 1987; Lee et al. 1977b; Lilis et al. 1975; Liss et al. 1985; Lloyd et al. 1984; Magnavita et al. 1986; Maricq et al. 1976; Markowitz et al. 1972; Marsteller et al. 1975; Micu et al. 1985; Miller 1975; NIOSH 1977; Perticoni et al. 1986; Popper and Thomas 1975; Popper et al. 1981; Preston et al. 1976; Sakabe 1975; Spirtas et al. 1975; Suciu et al. 1963, 1975; Tamburro et al. 1984; Veltman et al. 1975; Vihko et al. 1984; Walker 1976; Ward 1976; Wilson et al. 1967; Wong et al. 1991). The target organs include the liver, lungs, blood, immune system, cardiovascular system, skin, bones, nervous system, and the reproductive organs. These studies are severely limited in that individual exposure levels have not been documented. In general, studies in animals provide supportive evidence for these effects and give indications of the exposure levels that may be associated with them (Bi et al. 1985; Feron and Kroes 1979; Feron et al. 1979a, 1979b; Lee et al. 1981; Thornton et al. 2002; Viola 1970; Viola et al. 1971).

No information was available regarding chronic-duration oral exposure in humans. However, studies in animals indicate that the liver, blood, and skin are target organs for oral exposure to vinyl chloride (Feron et al. 1981; Knight and Gibbons 1987; Til et al. 1983, 1991). A chronic-duration oral MRL of 0.003 mg/kg/day was derived from a human equivalent NOAEL of 0.09 mg/kg/day based on liver cell polymorphism in rats (Til et al. 1983, 1991).

No information was available regarding effects of chronic-duration dermal exposure in humans or animals, but absorption of vinyl chloride gas through the skin was not significant in an acute-duration exposure study in monkeys (Hefner et al. 1975a). However, only two animals were used, and this was the only study located that examined toxicokinetics after dermal exposure. No information is available regarding dermal absorption of vinyl chloride from liquid or solid media (i.e., water, soil). Dermal exposure from these media is expected to be minimal; however, a study confirming this assumption would be useful. If further toxicokinetic studies demonstrate significant dermal absorption of vinyl chloride, then other intermediate-duration dermal exposure studies may be needed.

There is sufficient evidence to indicate that vinyl chloride is carcinogenic to humans (Belli et al. 1987; Boffetta et al. 2003; Brugnami et al. 1988; Byren et al. 1976; Cheng et al. 1999; Chung and Keh 1987; Cooper 1981; Creech and Johnson 1974; Davies et al. 1990; Du and Wang 1998; Fitzgerald and Griffiths 1987; Fox and Collier 1977; Gelin et al. 1989; Geryk and Zudova 1986; Hagmar et al. 1990; Heldass et al. 1987; Infante et al. 1976b; Jones et al. 1988; Lelbach 1996; Lewis 2001; Lewis and Rempala 2003; Lewis et al. 2003; Monson et al. 1975; Mundt et al. 2000; Ojajarvi et al. 2001; Pirastu et al. 1990; Rhomberg 1998; Rinsky et al. 1988; Saurin et al. 1997; Simonato et al. 1991; Smulevich et al. 1988; Teta et al. 1990; Ward et al. 2001; Waxweiler et al. 1981; Weber et al. 1981; Weihrauch et al. 2000; Williamson and Ramsden 1988; Wong et al. 1991, 2002a, 2002b, 2003a, 2003b; Wu et al. 1989) and animals (Bi et al. 1985; Drew et al. 1983; Feron and Kroes 1979; Feron et al. 1979a; Froment et al. 1994; Lee et al. 1978; Maltoni et al. 1981; Viola et al. 1971) exposed via inhalation, and in animals exposed via the oral route (Feron et al. 1979a; Maltoni et al. 1981; Til et al. 1983, 1991). The mechanism for carcinogenicity appears to be associated with the formation of reactive intermediates that bind to DNA.

Genotoxicity. There are substantial data on clastogenesis in humans exposed to vinyl chloride that indicate that this chemical acts as a potent genotoxicant (Anderson 2000; Anderson et al. 1980; Awara et al. 1998; Becker et al. 2001; Ducatman et al. 1975; Fucic et al. 1990a, 1990b, 1992, 1995; Funes-Cravioto et al. 1975; Hansteen et al. 1978; Hrivnak et al. 1990; Huttner and Nikolova 1998; Huttner et al. 1998, 1999; Kucerova et al. 1979; Marion et al. 1991; Purchase et al. 1978; Sinues et al. 1991; Wong et al.

1998; Zhao et al. 1996). The reversibility of chromosome damage has been reported for several populations of workers following a cessation or reduction of exposure to vinyl chloride (Anderson et al. 1980; Fucic et al. 1996a, 1996b; Hansteen et al. 1978). Findings in humans are supported by both animal studies and *in vitro* studies that show positive genotoxicity in a variety of microbial organisms, cultured cell lines, and isolated nucleic acid assays (Anderson and Richardson 1981; Andrews et al. 1976; Bartsch 1976; Bartsch et al. 1976; Bolt et al. 1986; Ciroussel et al. 1990; de Meester et al. 1980; Eberle et al. 1989; Froment et al. 1994; Green and Hathway 1978; Gwinner et al. 1983; Hansteen et al. 1978; Huberman et al. 1975; Jacobsen et al. 1989; Kandala et al. 1990; Laib and Bolt 1977; Laib et al. 1989; Loprieno et al. 1977; McCann et al. 1975; Osterman-Golkar et al. 1977; Poncelet et al. 1980; Rannug et al. 1974, 1976; Simmon et al. 1977; Singer et al. 1987; Victorin and Stahlberg 1988a; Walles et al. 1988). The role of etheno-adducts in the carcinogenesis of vinyl chloride has been extensively studied (Albertini et al. 2003, Barbin 1998, 1999, 2000; Kielhorn et al. 2000; Nivard and Vogel 1999; Whysner et al. 1996). Both 2-chloroethylene oxide and 2-chloroacetaldehyde can react with DNA nucleotide bases; however, 2-chloroethylene oxide is a more potent mutagen and may be the ultimate carcinogenic metabolite of vinyl chloride (Chiang et al. 1997). Etheno-adducts generate mainly base pair substitution mutations. Mutations in specific genes (i.e., *ras* oncogenes, p53 tumor suppressor gene) have been identified in vinyl chloride-induced liver tumors in rats and humans (Barbin et al. 1997; Brandt-Rauf et al. 1995; Hollstein et al. 1994; Marion and Boivin-Angele 1999; Marion et al. 1991; Trivers et al. 1995; Weihrauch et al. 2002). Immunological techniques have been used to detect the presence of Asp13p21 (oncoprotein for mutation of the Ki-*ras* gene), p53 mutant protein, and p53 antibodies in the serum of exposed workers (Brandt-Rauf et al. 2000a, 2000b; Marion 1998). Statistical analyses suggest a relationship between vinyl chloride exposure and the presence of these serum biomarkers; however, the predictive value of these biomarkers for development of cancer is not known. Further mechanistic research would be helpful in identifying the specific gene mutations responsible for vinyl chloride-induced liver cancer.

Reproductive Toxicity. Data from a number of epidemiological studies provide suggestive evidence of adverse effects on male and female reproductive function. Sexual impotence and decreased androgen levels were found in men exposed occupationally to vinyl chloride (Suciu et al. 1975; Veltman et al. 1975; Walker 1976). In women exposed to vinyl chloride, menstrual disturbances and an increased incidence of elevated blood pressure and edema during pregnancy (preeclampsia) were observed (Bao et al. 1988). Animal studies indicate that exposure to vinyl chloride can result in a decrease in testicular weight, damage to the seminiferous tubules, and depletion of spermatocytes (Bi et al. 1985). A significant increase in damage to the spermatogenic epithelium and disorders of spermatogenesis were also observed (Sokal et al. 1980). Reproductive capability was not affected in a 2-generation inhalation

reproductive toxicity study in rats (Thornton et al. 2002). No effects were seen in body weight, feed consumption, ability to reproduce, gestation index or length, or pre- and postweaning developmental landmarks. Sperm counts, motility, and morphology were also unaffected by vinyl chloride exposure. Animal models of preeclampsia could be tested to determine the mechanism by which vinyl chloride might produce this effect. Well-designed and well-conducted epidemiological studies examining such changes would also be helpful. No data are available on the possible reproductive toxicity resulting from oral exposure to vinyl chloride. Oral studies that use drinking water as the vehicle of administration would be particularly useful because contaminated groundwater is a potentially significant source of human exposure. However, such studies would be technically difficult to perform due to the volatility of vinyl chloride and its low solubility in water. The PBPK model would be useful for assessing reproductive toxicity resulting from oral exposure to vinyl chloride.

Developmental Toxicity. The epidemiological studies that have addressed developmental toxicity in offspring of humans who have been exposed to vinyl chloride are controversial. Although some of these purport to show a significant association between birth defects and vinyl chloride exposure (Infante 1976; Infante et al. 1976a, 1976b; NIOSH 1977), their design and analysis have been severely criticized (Hatch et al. 1981; Stallones 1987). At this time, there are insufficient human data to provide a definitive answer to this question. A well-designed and well-conducted epidemiological study examining potential developmental end points would be helpful. There are also inconsistencies in the developmental toxicity data for vinyl chloride in laboratory animals. In general, vinyl chloride produced minor adverse developmental effects only at concentrations that were significantly toxic to maternal animals. Concentrations of 500 ppm were observed to produce delayed ossification in the fetus and decreased food consumption, body weight gain, and mortality in maternal mice (John et al. 1977, 1981). In contrast, no adverse effects were reported in an embryo-fetal developmental toxicity study conducted in rats exposed to vinyl chloride via inhalation (Thornton et al. 2002). Embryo-fetal developmental parameters including uterine implantation, fetal gender distribution, fetal body weight, and fetal malformations and variations were not affected by vinyl chloride exposure. Vinyl chloride produced a decrease in maternal body weight gain at all exposure levels; however, no changes were observed in feed consumption, clinical signs, or postmortem gross findings. Maternal liver and kidney weights were increased relative to total body weight. It would be helpful to determine whether pregnancy increases the susceptibility to vinyl chloride in the mother. There are no data for oral exposures. Because of this deficiency, oral studies examining a range of developmental end points would be useful in assessing the possibility of these effects in humans. However, such studies would be technically difficult to perform due to the volatility of

vinyl chloride and its low solubility in water. The PBPK model would be a useful tool in such risk assessment.

Immunotoxicity. Studies of workers occupationally exposed to vinyl chloride suggest that the immune system may be activated by vinyl chloride (Bogdanikowa and Zawilska 1984). Some data suggest that reactive intermediates may bind to proteins in the body, sufficiently altering them so that they become antigenic (Grainger et al. 1980). In some instances, an autoimmune-like syndrome develops. The likelihood of this may be associated with the possession by individuals of specific genetic determinants (HLA alleles) (Black et al. 1983, 1986). Because of the low incidence of the autoimmune response in humans, the immunotoxicity may be best further studied in one of the strains of mice known to have a propensity for developing autoimmune diseases. Also, additional epidemiological studies examining the immune response of exposed populations may be helpful.

Neurotoxicity. A number of studies in humans (Lester et al. 1963; Patty et al. 1930) and animals (Hehir et al. 1981; Jaeger et al. 1974; Lester et al. 1963; Mastromatteo et al. 1960; Patty et al. 1930) demonstrate that vinyl chloride is a central nervous system depressant following brief high-level inhalation exposures. Two studies in animals have also found degenerative effects in central nervous system tissue following chronic inhalation exposure to high levels of vinyl chloride (Viola 1970; Viola et al. 1971). It is unknown whether these degenerative changes might also occur at lower doses; thus, a study examining the effects of a range of lower doses would be informative. In addition, relatively recent studies present suggestive evidence that vinyl chloride may also produce peripheral nerve damage in humans exposed chronically via inhalation (Langauer-Lewowicka et al. 1976; Magnavita et al. 1986; Perticoni et al. 1986; Sakabe 1975; Walker 1976). Animal studies examining histopathological and electrophysiological end points in peripheral nerves would be helpful for assessing what doses may be associated with this effect. Epidemiological studies examining exposed populations for subclinical peripheral nerve damage would also be helpful.

Epidemiological and Human Dosimetry Studies. Virtually all of the data on effects in humans following inhalation exposure to vinyl chloride come from epidemiological studies of workers exposed during the production of PVC (Belli et al. 1987; Boffetta et al. 2003; Brugnami et al. 1988; Byren et al. 1976; Cheng et al. 1999; Chung and Keh 1987; Cooper 1981; Creech and Johnson 1974; Davies et al. 1990; Du and Wang 1998; Fitzgerald and Griffiths 1987; Fox and Collier 1977; Gelin et al. 1989; Geryk and Zudova 1986; Hagmar et al. 1990; Heldass et al. 1987; Infante et al. 1976b; Jones et al. 1988; Lelbach 1996; Lewis 2001; Lewis and Rempala 2003; Lewis et al. 2003; Monson et al. 1975; Mundt et al.

2000; Ojajarvi et al. 2001; Pirastu et al. 1990; Rhomberg 1998; Rinsky et al. 1988; Saurin et al. 1997; Simonato et al. 1991; Smulevich et al. 1988; Teta et al. 1990; Ward et al. 2001; Waxweiler et al. 1981; Weber et al. 1981; Weihrauch et al. 2000; Williamson and Ramsden 1988; Wong et al. 2002a, 2002b, 2003a, 2003b, 1991; Wu et al. 1989). These studies are limited by the absence of information on individual exposure levels. Also, in North America and Western Europe, only limited numbers of females have been studied.

For the most part, studies examining the carcinogenic potential of vinyl chloride have been adequate to distinguish an increased incidence of the rare cancer, angiosarcoma (Byren et al. 1976; Creech and Johnson 1974; Fox and Collier 1977; Infante et al. 1976b; Jones et al. 1988; Monson et al. 1975; Pirastu et al. 1990; Rinsky et al. 1988; Teta et al. 1990; Waxweiler et al. 1976; Weber et al. 1981; Wong et al. 1991; Wu et al. 1989). However, many studies have used cohorts that are too small to detect smaller increases in other types of cancer (respiratory, central nervous system, lymphatic, or hematopoietic). Epidemiological studies designed to investigate reproductive and developmental effects of vinyl chloride have not been useful, in part because of a poor choice of statistical analysis, inadequate controls, lack of effects due to current low levels of exposure, or failure to take into account nutritional status and other chemical exposures. Additional cohort studies of these end points would be useful for examining these effects in humans.

Clastogenic effects have been used as a dosimeter for exposures to radioactive substances, and work has been done to use this approach for chemical exposures as well. More data on quantified exposures and well-controlled cytogenetic studies would be useful in developing a method for monitoring populations living near hazardous waste sites.

Biomarkers of Exposure and Effect.

Exposure. Several potential biomarkers for exposure to vinyl chloride have been identified. Vinyl chloride measured in expired air is an adequate indicator of recent, moderate-to-high-level exposure (Baretta et al. 1969). However, for low-level exposures or exposures that occur over 1–2 hours prior to the time of measurement, this biomarker is not useful. Thiodiglycolic acid, a major urinary metabolite of vinyl chloride, has been used to monitor workers occupationally exposed to vinyl chloride (Müller et al. 1979). However, this biomarker is rapidly excreted, and therefore, the period of its utility is limited (Watanabe and Gehring 1976; Watanabe et al. 1979b). Also, thiodiglycolic acid is not specific for vinyl

chloride; it may also be produced as a result of the metabolism of 1,1-dichloroethene, ethylene oxide, or 2,2-dichloroethylether (Norpoth et al. 1986; Pettit 1986).

The DNA adducts 1,N^6-ethenoadenosine and 3,N^4-ethenocytidine may be used to indicate vinyl chloride exposure, although studies correlating the levels of these adducts with exposure levels are still lacking. These products remain in the body longer than free vinyl chloride or thiodiglycolic acid, thereby increasing the period after exposure that a potential exposure may be detected (Bolt 1986; Guengerich and Watanabe 1979; Guengerich et al. 1979, 1981; Kappus et al. 1976; Watanabe et al. 1987a, 1987b). However, the presence of these adducts cannot indicate how long it has been since exposure occurred. In addition, these adducts are formed as the result of binding of the intermediary metabolites with nucleic acids, and other compounds producing the same intermediary metabolites will also produce these adducts. For example, these adducts have been identified as a result of exposure to vinyl bromide, ethyl carbamate, acrylonitrile, 2-cyanoethylene, and 1,2-dichloroethane (Bolt et al. 1986; Svensson and Osterman-Golkar 1986). Studies attempting to identify a metabolite more specific to vinyl chloride may be helpful in developing a biomarker that may be used to facilitate future medical surveillance, which can lead to early detection and possible treatment.

Vinyl chloride-induced genetic alterations have been identified in the Ki-*ras* oncogene and the p53 tumor suppressor gene, and oncoproteins and p53 antibodies have been detected in the serum of cancer patients with angiosarcoma (see Section 3.3). Immunological techniques have been used to detect the presence of Asp13p21 (oncoprotein for mutation of the Ki-*ras* gene), p53 mutant protein, and p53 antibodies in the serum of exposed workers (Brandt-Rauf et al. 2000a, 2000b; Marion 1998). Statistical analyses suggest a relationship between vinyl chloride exposure and the presence of these serum biomarkers; however, the predictive value of these biomarkers for development of cancer is not known.

Effect. With regard to biomarkers of effect of vinyl chloride exposure, numerous indicators have been examined. The central nervous system depression associated with brief high-level exposures is easily determined by observation. The hepatic changes that may develop during longer term exposures are difficult to detect by standard biochemical liver function tests (Berk et al. 1975; Du et al. 1995; Liss et al. 1985; Vihko et al. 1984). In contrast, tests of clearance such as the indocyanine clearance test or measurement of serum bile acid levels are more specific and sensitive indicators of vinyl chloride-induced liver damage (Berk et al. 1975; Liss et al. 1985; Vihko et al. 1984). Angiosarcoma of the liver is a rare tumor type that has been shown to result from vinyl chloride exposure. However, other agents are known to produce angiosarcoma of the liver, such as arsenic and Thorotrast® (Gedigke et al. 1975; Marsteller et

al. 1975). Enzyme-linked immunoassay (EIA) has been used to detect anti-p53 antibodies in the serum of some individuals with angiosarcoma of the liver before clinical diagnosis of this lesion was made (Trivers et al. 1995). However, not all individuals who develop angiosarcoma of the liver test positive for anti-p53 antibodies; in addition, anti-p53 bodies are not specific only to angiosarcoma of the liver. Further investigation into the ability of this assay to predict individuals at increased risk for developing angiosarcoma of the liver would be useful. Measurement of chromosomal aberrations may indicate the genotoxic effects of vinyl chloride (Anderson et al. 1980; Ducatman et al. 1975; Fucic et al. 1990a, 1990b). However, these aberrations do not specifically indicate vinyl chloride-induced damage. Also, DNA adducts may signal the potential to develop genotoxic effects. Further work identifying the correlation between specific adducts and genotoxic effects would be useful. The cyanosis and blanching of the fingers in response to exposure to the cold may be an early indicator for the development of vinyl chloride disease. However, other conditions also known to produce these symptoms include connective tissue disorders, mechanical arterial obstruction, hyperviscosity of the blood, and exposure to drugs, chemicals, or vibrating tools (Black et al. 1983, 1986; Freudiger et al. 1988). The presence of basophilic stippled erythrocytes has been reported after inhalation exposure of mice to vinyl chloride (Kudo et al. 1990). Further study would be necessary to determine whether this parameter could be used as a biomarker of effect in humans.

Absorption, Distribution, Metabolism, and Excretion. There are few data on humans for all toxicokinetic parameters across all exposure routes (Krajewski et al. 1980; Sabadie et al. 1980). There are a number of animal studies describing the absorption, distribution, metabolism, and excretion of vinyl chloride administered via the oral route (Feron et al. 1981; Green and Hathway 1978; Watanabe and Gehring 1976; Watanabe et al. 1987a, 1987b; Withey 1976) and the inhalation route (Bolt et al. 1976a, 1977; Buchter et al. 1977, 1980; Filser and Bolt 1979; Guengerich and Watanabe 1979; Hefner et al. 1975b; Jedrychowski et al. 1984, 1985; Ungvary et al. 1978; Watanabe and Gehring 1976; Watanabe et al. 1978a, 1978b; Withey 1976) but few describing the toxicokinetics of vinyl chloride administered via the dermal route. One study in monkeys found an extremely limited absorption of vinyl chloride across the skin (Hefner et al. 1975a). However, only two animals were used, and this was the only study located that examined toxicokinetics after dermal exposure. No information is available regarding dermal absorption of vinyl chloride from liquid or solid media (i.e., water, soil). Dermal exposure from these media is expected to be minimal; however, a study confirming this assumption would be useful. Furthermore, the intermediary metabolites of vinyl chloride appear to be responsible for many of the toxic effects observed. Therefore, information regarding differences in the metabolic pattern according to

gender, age, nutritional status, and species and correlations to differences in health effects would also be useful.

Comparative Toxicokinetics. The absorption, distribution, metabolism, and excretion of vinyl chloride have been studied in animals (Bolt et al. 1976a, 1977; Buchter et al. 1977, 1980; Feron et al. 1981; Filser and Bolt 1979; Green and Hathway 1975; Guengerich and Watanabe 1979; Hefner et al. 1975b; Jedrychowski et al. 1984, 1985; Ungvary et al. 1978; Watanabe and Gehring 1976; Watanabe et al. 1976a, 1976b, 1978a, 1978b; Withey 1976), but information on toxicokinetics in humans is extremely limited (Krajewski et al. 1980; Sabadie et al. 1980). Human and animal data indicate that similar target organs (liver, central nervous system) for the toxic effects of vinyl chloride exist, suggesting some similarities of kinetics. Limited information is available regarding interspecies differences in kinetics. Most toxicokinetic studies have been conducted using rats (Bolt et al. 1976a, 1977; Buchter et al. 1977; Feron et al. 1981; Filser and Bolt 1979; Green and Hathway 1975; Guengerich and Watanabe 1979; Hefner et al. 1975b; Jedrychowski et al. 1984, 1985; Ungvary et al. 1978; Watanabe and Gehring 1976; Watanabe et al. 1976a, 1976b, 1978a, 1978b; Withey 1976), but one study in primates indicates that metabolism may saturate at lower concentrations in primates than rats (Buchter et al. 1980). This may suggest a lower saturation point in humans also. Modeling studies might continue to provide information on the toxicokinetics of vinyl chloride in humans.

Methods for Reducing Toxic Effects. Vinyl chloride appears to be rapidly and completely absorbed following inhalation and oral exposure (Bolt et al. 1977; Krajewski et al. 1980; Watanabe et al. 1976a; Withey 1976). Methods used to reduce absorption immediately after exposure include removal from the source of exposure, cleansing contaminated body parts, and in cases of ingestion, speeding the removal of unabsorbed material from the gastrointestinal tract (Bronstein and Currance 1988; Haddad and Winchester 1990; Stutz and Ulin 1992). No information was located regarding the mechanism of absorption. Additional experiments examining the mechanism of absorption and potential means of interfering with that mechanism would be useful. Distribution of vinyl chloride in the body is rapid and widespread, but storage is limited by rapid metabolism and excretion (Bolt et al. 1976a; Buchter et al. 1977; Watanabe et al. 1976a, 1976b, 1978a). The toxicity of vinyl chloride has been attributed to the formation of reactive epoxide metabolites. No information was located regarding removal of these toxic metabolites from the body once they have been formed, but information from toxicokinetic studies suggest that vinyl chloride metabolism to toxic metabolites may be reduced. Saturation of the metabolic pathways for vinyl chloride can result in the clearance of unmetabolized vinyl chloride in exhaled air (Green and Hathway 1975; Watanabe and Gehring 1976; Watanabe et al. 1976a, 1976b, 1978a). Studies

examining the effectiveness and endogenous toxicity of the agents used to block the metabolic pathways (cobaltous chloride, SKF-535-A, 6-nitro-1,2,3-benzothiadiazole) would provide useful information. Another strategy for reducing the formation of toxic metabolites includes increasing the pool of glutathione for use in metabolism to nontoxic metabolites. Studies examining the effectiveness of this procedure would also be helpful. Vinyl chloride disease may be mediated by an autoimmune mechanism (Grainger et al. 1980; Ward 1976). Further studies continuing to examine the role of autoimmune responses in vinyl chloride disease, the genetic factors resulting in greater susceptibility to the disease, and the effectiveness of drugs that block immune responses in reducing the symptoms of vinyl chloride disease would also provide valuable information.

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.

No studies were located that specifically address the effects of vinyl chloride in children. Some epidemiologic studies (Infante 1976; Infante et al. 1976a, 1976b; NIOSH 1977) have suggested an association between birth defects and vinyl chloride exposure of the parents of affected children. However, the design and analysis of these studies has been criticized (Hatch et al. 1981; Stallones 1987). Some inhalation studies with animals have suggested that vinyl chloride is a developmental toxicant (i.e., produces delayed ossification), but only at doses that produce significant maternal toxicity (John et al. 1977, 1981; Mirkova et al. 1978; Sal'nikova and Kotsovskaya 1980; Ungvary et al. 1978). No adverse effects on embryo-fetal development were noted in a recent inhalation study in rats conducted using similar concentrations of vinyl chloride (Thornton et al. 2002). There is no evidence that vinyl chloride has hormone-like effects. However, a developmental neurotoxicity study in rats in which pups are tested at various ages after being exposed *in utero* would be informative.

Carcinogenicity studies with animals suggest that younger animals may be more sensitive to the toxicity and carcinogenicity of vinyl chloride (Laib et al. 1985; Maltoni et al. 1981). An age-related sensitivity to DNA adduct formation was noted in rats (Ciroussel et al. 1990; Fedtke et al. 1990; Morinello et al. 2002a). Further mechanistic research may be useful in establishing the mechanism of early life stage sensitivity in laboratory animals and determining whether it is anticipated to be relevant to humans.

No studies were located that specifically address the toxicokinetics of vinyl chloride in children; however, the toxicokinetic behavior of vinyl chloride in children is expected to be similar to that in adults. Young

children appear capable of metabolizing vinyl chloride to reactive intermediates that form DNA adducts that lead to cancer. The data on CYP2E1 levels in the developing organism suggest that early life stage sensitivity to vinyl chloride-induced cancer is not solely due to an increase in the production of reactive intermediates via this isozyme. Fetal CYP isoforms may play a role in metabolism of vinyl chloride to reactive intermediates in the fetus and neonate. Glutathione conjugation may also differ in the developing organism. DNA repair capacity and other pharmacodynamic factors may also be associated with an early life stage susceptibility to cancer. Further information on the toxicokinetics and toxicodynamics of vinyl chloride and metabolites during pregnancy, lactation, and early childhood would be useful. The biomarkers of exposure and effects used in occupational worker populations should be evaluated for their relevance to human exposure at all age levels following acute or chronic exposure to vinyl chloride. There are no data on the interaction of vinyl chloride with other chemicals in children. The information available indicates that methods to reduce peak absorption of vinyl chloride are applicable to children.

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

The following ongoing studies concerning the adverse health effects associated with vinyl chloride have been identified in the Federal Research in Progress (FEDRIP 2005) database.

Dr. P.W. Brandt-Rauf at Columbia University proposes to investigate whether genetic polymorphisms in vinyl chloride-metabolizing enzymes are also related to the more specific biomarkers of mutagenic damage (mutant *ras*-p21 and/or mutant p53) in vinyl chloride-exposed workers. Restriction fragment length polymorphism techniques will be used to analyze DNA from sub-groups of vinyl chloride-exposed workers. It is anticipated that workers with genetic polymorphisms will be more likely to have the biomarkers of mutagenic damage than similarly exposed workers without the polymorphisms and thus will be more likely to suffer from the subsequent carcinogenic and other health effects of vinyl chloride exposure. If this proves to be correct, then such special populations at risk could be targeted for more stringent interventions to help prevent the occurrence of vinyl chloride-related occupational diseases. This research is sponsored by the National Institute for Occupational Safety and Health.

Dr. W.K. Kaufman at the University of North Carolina at Chapel Hill will investigate the role of DNA repair in the formation of hprt mutations in vinyl chloride-exposed workers. A subfraction of people

exposed to vinyl chloride in the workplace expressed high frequencies of hprt mutations in blood lymphocytes. The possible existence of a DNA repair defect in sensitive workers will be evaluated by studying chlorethylene oxide-induced genotoxicity in lymphoblastoid lines derived from sensitive and resistant people. This project will employ a functional assay for DNA repair capacity in peripheral lymphocytes that measures rejoining of radiation-induced chromatid breaks. This research is supported by the National Institute of Environmental Health Sciences (NIEHS).

Dr. G.E. Kisby at the Oregon Health Sciences University proposes experiments to examine the relationship between the formation of etheno base DNA adducts of chloroacetaldehyde and neurotoxicity or mutations. Neuronal and astrocyte cell cultures will be developed from different brain regions (e.g., cortex, hippocampus, midbrain, cerebellum) of DNA repair proficient and deficient mice (i.e., k N-methylpurine DNA glycosylase Aag). These cell lines will be examined for acute and delayed chloroacetaldehyde-induced neurotoxicity. Separate sets of astrocyte cell cultures will be developed from hprt heterozygous-deficient mice and examined to determine the spectrum of chloroacetaldehyde-induced mutations. Findings from these studies are expected to provide important information about the neurotoxic and mutagenic mechanisms of vinyl chloride. This research is sponsored by the NIEHS.

Dr. K.D. Thrall at the Oregon Health Sciences University will investigate the influence of route of exposure on the total body burden and internal target tissue dosimetry of vinyl chloride and other chemicals. Exposure assessment studies will be conducted with human volunteers using a novel real-time breath analysis system to determine the uptake of contaminants from tap water by each of three routes: inhalation, ingestion, and dermal contact. These data will be coupled with PBPK modeling to determine uptake kinetics and brain dosimetry. This research is sponsored by the NIEHS.