2. HEALTH EFFECTS

2.1 INTRODUCTION

The primary purpose of this chapter is to provide public health officials, physicians, toxicologists, and other interested individuals and groups with an overall perspective of the toxicology of 2-butoxyethanol and 2-butoxyethanol acetate. It contains descriptions and evaluations of toxicological studies and epidemiological investigations and provides conclusions, where possible, on the relevance of toxicity and toxicokinetic data to public health.

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

2.2 DISCUSSION OF HEALTH EFFECTS BY ROUTE OF EXPOSURE

To help public health professionals and others address the needs of persons living or working near hazardous waste sites, the information in this section is organized first by route of exposure─inhalation, oral, and dermal; and then by health effect─death, systemic, immunological, neurological, reproductive, developmental, genotoxic, and carcinogenic effects. These data are discussed in terms of three exposure periods─acute (14 days or less), intermediate (15-364 days), and chronic (365 days or more).
Levels of significant exposure for each route and duration are presented in tables and illustrated in Figures. The points in the figures showing no-observed-adverse-effect levels (NOAELs) or lowest-observedadverse-effect levels (LOAELs) reflect the actual doses (levels of exposure) used in the studies. LOAELs have been classified into “less serious” or “serious” effects. “Serious” effects are those that evoke failure in a biological system and can lead to morbidity or mortality (e.g., acute respiratory distress or death). “Less serious” effects are those that are not expected to cause significant dysfunction or death, or those whose significance to the organism is not entirely clear. ATSDR acknowledges that a considerable amount of judgment may be required in establishing whether an end point should be classified as a NOAEL, “less serious” LOAEL, or “serious” LOAEL, and that in some cases, there will be insufficient data to decide whether the effect is indicative of significant dysfunction. However, the Agency has established guidelines and policies that are used to classify these end points. ATSDR believes that there is sufficient merit in this approach to warrant an attempt at distinguishing between “less serious” and “serious” effects. The distinction between “less serious” effects and “serious” effects is considered to be important because it helps the users of

the profiles to identify levels of exposure at which major health effects start to appear. LOAELs or NOAELs should also help in determining whether or not the effects vary with dose and/or duration, and place into perspective the possible significance of these effects to human health.

The significance of the exposure levels shown in the Levels of Significant Exposure (LSE) tables and figures may differ depending on the user's perspective. Public health officials and others concerned with appropriate actions to take at hazardous waste sites may want information on levels of exposure associated with more subtle effects in humans or animals (LOAEL) 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.

Estimates of exposure levels posing minimal risk to humans (Minimal Risk Levels or MRLs) have been made for 2-butoxyethanol. Insufficient data were available to estimate MRLs for 2-butoxyethanol acetate. Since 2-butoxyethanol acetate is metabolized to the same toxic metabolite as is 2-butoxyethanol, it seems likely that the MRLs for 2-butoxyethanol acetate would be similar to those of 2-butoxyethanol. 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 these levels (Barnes and Dourson 1988; EPA 1990a), 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.

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

2.2.1 inhalation Exposure

Occupational or environmental exposure to 2-butoxyethanol or 2-butoxyethanol acetate usually occurs through inhalation or dermal contact. Both routes of exposure are important in the industrial setting. Occupational exposure to 2-butoxyethanol usually involves co-exposure to other solvents and chemicals. In several NIOSH Health Hazard Evaluations, effects reported by workers included eye, nose, and throat irritation; coughing; runny nose; headache; dizziness; lightheadedness; and nausea (Apol 1981,1986; Lee 1988). Since personal breathing zone and workplace air samples analyzed for solvents and other clremicals, such as toluene, xylene, methyl ethyl ketone, methyl isobutyl ketone, styrene, along with 2-butoxyethanol, indicated that exposure levels for each chemical were below the NIOSH, ACGIH, and OSHA criteria, NIOSH concluded that the effects were probably due to the additive combination of the solvents. Experimental studies in which humans were exposed to 2-butoxyethanol by the inhalation route, in addition to experimental animal studies, are discussed in this section (Section 2.2.1) and have been summarized in several publications (Browning and Curry 1994; EPA 1984; NIOSH 1990; Tyler 1984). Although one study was located regarding oral exposure of animals to the major metabolite of 2-butoxyethanol, 2-butoxyacetic acid (discussed in Section 2.2.2), no studies were located describing the effects of 2-butoxyacetic acid after inhalation exposure.

2.2.1.1 Death

No studies were located regarding death in humans after inhalation exposure to 2-butoxyethanol or 2-butoxy-ethanol acetate.

The amount of 2-butoxyethanol in air required to produce death in animals has been measured under a variety of conditions. Acute 4-hour LC_{50} (lethal concentration, 50% kill) values for Fischer 344 rats after wholebody exposure were determined to be 486 ppm for males and 450 ppm for females (Dodd et al. 1983). In another study, a 4-hour LC_{50} for rats was reported to be 500 ppm (Nelson et al. 1984). In addition, in a preliminary concentration-finding study, in which non-pregnant female Sprague-Dawley rats were exposed to 2-butoxyethanol for 6.5 or 7 hours, death occurred in three of four rats at 450-500 ppm, in two of three rats at 350 ppm, and in two of three rats at 250 ppm for 7 hours (Nelson et al. 1984). Male Fischer 344 rats exposed to 2-butoxyethanol by nose-only inhalation at 438 ppm 2-butoxyethanol for 6 hours exhibited 50% (two of four) mortality (Sabourin et al. 1992a). Carpenter et al. (1956) observed death in rats at ≥375 ppm for l-3 days (7 hours per day) and at 314 ppm for up to 30 days (7 hours per day, and 5 days per week). No

deaths occurred in rats similarly exposed to ≤203 ppm for 30 days Similarly, when Fischer 344 rats were exposed to concentrations of 0,20,86, or 245 ppm of 2butoxyethanol for 9 days, 6 hours per day, 4-5 days per week, no rats died during the exposure period or the 14-day observation period (Dodd et al. 1983). Sprague-Dawley rats exposed to 682 ppm 2-butoxyethanol for 3 hours showed no mortality during the exposure or the 14-day postexposure observation period (Khmisch et al. 1988). Similarly, no Fischer 344 rats died after 13 weeks of exposure to 77 ppm 2-butoxyethanol for 5 days per week, 6 hours per day (Dodd et al. 1983). An inhalation LC_{50} of 700 ppm was reported for mice exposed to 2-butoxyethanol for 7 hours (Werner et al. 1943a). C3H mice exhibited 20% (2 of 10) mortality after continuous exposure to 376 ppm 2-butoxyethanol and 70% (7 of 10) mortality after continuous exposure to 494 ppm for 30 days (no controls were used), although the same strain of mice exposed to ≤400 ppm for 90 days exhibited no mortality (Carpenter et al. 1956). Guinea pigs seem to be less sensitive to the lethal effects of 2-butoxyethanol and exhibited no mortality after exposure to 691 ppm for 1 hour followed by a 14-day observation period (Nachreiner 1994) or to 411 ppm for up to 5 days (Dow 1986). However, Carpenter et al. (1956) reported death in 1 of 10 guinea pigs exposed to 376 ppm and in 2 of 10 guinea pigs exposed to 494 ppm for 7 hours per day, 5 days per week, for a total of 30 days. No deaths occurred at ≤203 ppm. Some studies in dogs have reported mortality at concentrations of 385-617 ppm, with exposures of 2-8 days or 28 days (Carpenter et al. 1956), whereas other reports indicate no mortality in dogs at concentrations that are similar or lower (Dow 1972,1986). The studies in dogs used only one or two dogs per exposure level and sometimes did not include a control dog. Rabbits exhibited mortality at concentrations ≥ 200 ppm after exposure for 6 or 7 hours per day for 1-13 days (Dow 1986; Tyl et al. 1984).

No deaths were noted in Wistar rats exposed to a saturated vapor-air mixture of approximately 400 ppm 2-butoxyethanol acetate for 4 hours; 400 ppm for 1 month, 5 days per week, 4 hours per day; or 100 ppm for 10 months using the same intermittent exposure regimen (Truhaut et al. 1979). No deaths were observed in New Zealand white rabbits exposed to 400 ppm 2-butoxyethanol acetate for 4 hours or to 100 ppm 2-butoxy-ethanol acetate for 10 months under the same regimen as the rats. However, two of four rabbits died after exposure to 400 ppm 2-butoxyethanol acetate for 1 month, 5 days per week, 4 hours per day (Truhaut et al. 1979).

The LC_{50} values for rats and all LOAEL values from each reliable study for death for each species and duration category for 2-butoxyethanol and 2-butoxyethanol acetate are recorded in Tables 2- 1 and 2-2 and plotted in Figures 2-l and 2-2, respectively.

Table 2-1. Levels of Significant Exposure to 2-Butoxyethanol - Inhalation

Key to figure[a]	Species/ (strain)	Exposure/ duration/ frequency	System	NOAEL (ppm)	LOAEL Less serious (ppm)	LOAEL Serious (ppm)	Reference
ACUTE EXPOSURE							
Death							
1	Rat (NS)	1-6 d 2-9 hr/d				375 (death of 11/13 males and 23/23 females after 7 hrs)	Carpenter et al. 1956
2	Rat (Sherman)	3 d 7 hr/d				432 F (15/15 died)	Carpenter et al. 1956
3	Rat (Fischer- 344)	4 hr				486 M (LC_{50}) 450 F (LC_{50})	Dodd et al. 1983
4	Rat (Sprague-Dawley)	1 d 6.5 -7hr				250 F (2/3 died)	Nelson et al. 1984
5	Rat (Fischer- 344)	6 hr				438 M (2/4 died)	Sabourin et al. 1992a
6	Mouse (Swiss)	7 hr				700 (LC_{50})	Werner et al. 1943a
7	Rabbit (albino)	1-2d 7 hr/d				400-411 M (25-100% death)	Dow 1986
8	Rabbit (New Zealand)	Gd 6-18 6 hr/d				200 F (4/24 died)	Tyl et al. 1984
Systemic							
9	Human	4-8 hr	Resp	98	113 (nasal irritation, slight increase in nasal mucus discharge)		Carpenter et al. 1956
			Cardio	195 M			
			Gastro		98 F (emesis)		
			Hemato	195			
			Ocular	98	113 (ocular irritation)		
10	Human	2 hr	Resp	20 M			Johanson et al. 1986a
			Cardio	20 M			

Table 2-1. Levels of Significant Exposure to 2-Butoxyethanol - Inhalation (continued)

Key to figure[a]	Species/ (strain)	Exposure/ duration/ frequency	System	NOAEL (ppm)	LOAEL Less serious (ppm)	LOAEL Serious (ppm)	Reference
11	Rat (NS)	2-8 hr	Hemato			432 F (hemolysis in 2 hrs; hemoglobinuria in 3 hrs; hemin crystals in urine in 4 hrs)	Carpenter et al. 1956
			Renal		432 F (slight cloudy swelling of convoluted tubules in 2 hrs)		
12	Rat (NS)	9 d 7 hr/d	Hemato			200 F (50% decrease in erythrocyte count and 25% decrease in HGB level)	Carpenter et al. 1956
13	Rat (NS)	4 hr	Hemato	32 F	62 F (significant osmotic fragility of RBCs)		Carpenter et al. 1956
14	Rat (Fischer- 344)	4 hr	Resp	523	867 (rapid and shallow breathing)		Dodd et al. 1983
			Renal	202		523 (red discharge around urogenital area and bladder [hematuria]; enlarged kidneys)	

Table 2-1. Levels of Significant Exposure to 2-Butoxyethanol - Inhalation (continued)

Key to figure[a]	Species/ (strain)	Exposure/ duration/ frequency	System	NOAEL (ppm)	LOAEL Less serious (ppm)	LOAEL Serious (ppm)	Reference
15	Rat (Fischer- 344)	9 d 5 d/wk 6 hr/d	Resp	86 M 245 F	245 M (audible respiration and nasal discharge)		Dodd et al. 1983
			Hemato	20		86 M (HGB decreased 5%, significant increases in MCV of 11%) 86 F (HGB decreased 8%, MCHC decreased 18%, significant increases in MCV of 17%)	
			Hepatic	86 M 20 F	245 M (increased liver weights of about 5.4%) 86 F (increased liver weights of about 4.5%)		
			Renal	86	245 (transient red-stained urine [hematuria])		
			Ocular	245			
			Bd Wt	86 M 20 F	245 M (13% decrease in body weight gain) 86 F (10% decrease in body weight gain)		
16	Rat (Alpk/Ap)	3 hr	Renal			800 M (hematuria)	Doe 1984
17	Rat (Sprague-Dawley)	4 d 7 hr/d	Bd Wt	57-58 M			Dow 1972
18	Rat (Sprague-Dawley)	Gd 7-15 7 hr/d	Renal		150 F (slight hematuria)		Nelson et al. 1984
19	Rat (Sprague-Dawley)	1 d 6.5 -7 hr	Renal			250 F (hematuria)	Nelson et al. 1984
			Dermal		250 F (necrotic tail tip)		

Table 2-1. Levels of Significant Exposure to 2-Butoxyethanol - Inhalation (continued)

Key to figure[a]	Species/ (strain)	Exposure/ duration/ frequency	System	NOAEL (ppm)	LOAEL Less serious (ppm)	LOAEL Serious (ppm)	Reference
20	Rat (Fischer- 344)	6 hr	Hemato			438 M (hemoglobinuria)	Sabourin et al. 1992a
21	Rat (Fischer- 344)	Gd 6-15 6 hr/d	Resp	50 F	100 F (perinasal encrustation)		Tyl et al. 1984
			Hemato	50[b] F		100 F (reduced RBC and MCHC; increased MCH and MCV)	
			Hepatic	200 F			
			Renal	50 F		100 F (hematuria)	
			Dermal	100 F	200 F (necrosis of the tail tip, stained fur)		
			Ocular		25 F (periocular wetness)		
			Bd Wt	50 F		100 F (29% decrease in body weight gain)	
			Other	50 F	100 F (13% reduction in food consumption)		
				100 F	200 F (14% reduction in water consumption)		
22	Mouse (NS)	7 hr	Hemato		100 (increased osmotic fragility)		Carpenter et al. 1956
23	Mouse (Swiss-Webster)	10 min	Resp		153M (20% decrease in respiratory rate)		Kane et al. 1980
24	Gn Pig (NS)	8 hr	Hemato	665 M			Carpenter et al. 1956
25	Rabbit (NS)	7 hr	Hemato		125 (increased osmotic fragility of RBCs)		Carpenter et al. 1956

Table 2-1. Levels of Significant Exposure to 2-Butoxyethanol - Inhalation (continued)

Key to figure[a]	Species/ (strain)	Exposure/ duration/ frequency	System	NOAEL (ppm)	LOAEL Less serious (ppm)	LOAEL Serious (ppm)	Reference
26	Rabbit (albino)	1-2d 7 hr/d	Resp			400-411 M (congestion of lungs and turbinates, nasal discharge)	Dow 1986
			Gastro			400-411 M (hemorrhagic gastric ulcers)	
			Hepatic		400-411 M (mottled livers in surviving animals)		
			Renal			400-411 M (darkened kidneys in surviving animals, hematuria)	
			Ocular		400-411 M (ocular discharge, yellowing of sclerae)		
27	Rabbit (New Zealand)	Gd 6-18 6 hr/d	Resp	100 F	200 F (perinasal wetness and discharge)		Tyl et al. 1984
			Hemato	50 F	100 F (increased hemoglobin and hematocrit)		
			Hepatic	200 F			
			Renal	50 F		100 F (hematuria)	
			Dermal	100 F	200 F (stained fur)		
			Ocular	50 F	100 F (periocular wetness)		
			Bd Wt	100 F	200 F (9.7% reduction in maternal body weight)		
Immunological/Lymphoreticular							
28	Rat (Fischer- 344)	Gd 6-15 6 hr/d		100 F	200 F (20-24% increase in absolute and relative maternal spleen weights)		Tyl et al. 1984
Neurological							
29	Human	4-8 hr			98 (headache)		Carpenter et al. 1956
				98	113 M (disagreeable metallic taste)		
30	Rat (Fischer- 344)	4 hr		202		523 (loss of coordination)	Dodd et al. 1983

Table 2-1. Levels of Significant Exposure to 2-Butoxyethanol - Inhalation (continued)

Key to figure[a]	Species/ (strain)	Exposure/ duration/ frequency	System	NOAEL (ppm)	LOAEL Less serious (ppm)	LOAEL Serious (ppm)	Reference
31	Dog (Beagle)	1-5 d 7 hr/d			400-411 M (salivation)		Dow 1986
32	Rabbit (albino)	1-2 d 7 hr/d				400-411 M (poor coordination of extremities and loss of equilibrium)	Dow 1986
	Reproductive						
33	Rat (Fischer- 344)	Gd 6-15 6 hr/d		100 F		200 F (50% decrease in viable implants & in live fetuses per litter; 8-fold increase in nonviable implants; reduced maternal gravid uterine weight)	Tyl et al. 1984
34	Rabbit (New Zealand)	Gd 6-18 6 hr/d		100		200 F (14% decrease in total implants; 20% decrease in viable implants)	Tyl et al. 1984
	Developmental						
35	Rat (Sprague-Dawley)	Gd 7-15 7 hr/d		200			Nelson et al. 1984
36	Rat (Fischer- 344)	Gd 6-15 6 hr/d		50	100 (retarded skeletal ossification)		Tyl et al. 1984
37	Rabbit (New Zealand)	Gd 6-18 6 hr/d		100	200 (22% reduction in gravid uterine weight, reduced ossification in fetuses)		Tyl et al. 1984
	INTERMEDIATE EXPOSURE						
	Death						
38	Rat (Sherman)	30 d 5 d/wk 7 hr/d				432 M (12/15 died) 314 F (15/15 died)	Carpenter et al. 1956
39	Mouse (C3H)	30 d				376 M (2/10 died)	Carpenter et al. 1956

Table 2-1. Levels of Significant Exposure to 2-Butoxyethanol - Inhalation (continued)

Key to figure[a]	Species/ (strain)	Exposure/ duration/ frequency	System	NOAEL (ppm)	LOAEL Less serious (ppm)	LOAEL Serious (ppm)	Reference
40	Gn Pig (NS)	30 d 5 d/wk 7 hr/d				376 M (1/10 died)	Carpenter et al. 1956
	Systemic						
41	Rat (Sherman)	30 d 5 d/wk 7 hr/d	Resp	203		432 M (congestion & hemorrhage 314 F of the lungs)	Carpenter et al. 1956
			Gastro	203		432 M (congestion of the 314 F abdominal viscera)	
			Hemato		54 (erythrocyte fragility)	203 (hemoglobinuria)	
			Hepatic	54	107 (unspecified increase in liver weight)		
			Renal	54	107 (unspecified increase in kidney weight)		
			Bd Wt	203			
42	Rat (Fischer- 344)	13 wk 5 d/wk 6 hr/d	Resp	77			Dodd et al. 1983
			Cardio	77			
			Hemato	25[c]		77 (5%-13% decrease in RBC, in both sexes; 4% decrease in HGB; 11% increase in MCH, females)	
			Musc/skel	77			
			Hepatic	77			
			Renal	77			
			Endocr	77			
			Bd Wt	77			
43	Mouse (C3H)	30-90 d 7 hr/d	Hemato		100 M (increased erythrocyte osmotic fragility)		Carpenter et al. 1956
			Hepatic	100 M	200 M (unspecified increase ($p<0.05$) in liver weights)		
			Renal	400 M			
			Bd Wt	200 M	400 M (unspecified decrease ($p<0.05$) in body weights)		

Table 2-1. Levels of Significant Exposure to 2-Butoxyethanol - Inhalation (continued)

Key to figure[a]	Species/ (strain)	Exposure/ duration/ frequency	System	NOAEL (ppm)	LOAEL Less serious (ppm)	LOAEL Serious (ppm)	Reference
44	Gn Pig (NS)	30 d 5 d/wk 7 hr/d	Resp	203 M	376 M (lung congestion)		Carpenter et al. 1956
			Hemato	494 M			
			Hepatic	494 M			
			Renal	107 M	203 M (unspecified increase ($p<0.05$) in kidney weight)		
			Bd Wt	495 M			
45	Dog (Hybrid)	31 d	Resp		200 (slight capillary engorgement or breakdown in the lungs)		Carpenter et al. 1956
			Hemato			200 M (increased osmotic fragility of RBCs and 100% increased leukocyte counts)	
					200 F (RBC osmotic fragility increased slightly; slight decrease in RBC count and hemoglobin level)		
			Ocular	200			
46	Dog (Terrier)	90 d	Hemato		100 (decreased hematocrit in male, transitory doubling of the leukocyte count midway into the 90-day exposure in both sexes)		Carpenter et al. 1956

Table 2-1. Levels of Significant Exposure to 2-Butoxyethanol - Inhalation (continued)

Key to figure[a]	Species/ (strain)	Exposure/ duration/ frequency	System	NOAEL (ppm)	LOAEL Less serious (ppm)	LOAEL Serious (ppm)	Reference
47	Dog (NS)	12 wk 5 d/wk 7 hr/d	Resp		415 (slightly increased nasal secretions)		Werner et al. 1943b
			Cardio	415			
			Gastro	415			
			Hemato		415 (decreased Hgb, hematocrit; hypochromia, polychromatophilia, and microcytosis)		
			Hepatic	415			
			Renal	415			
			Ocular		415 (slight increased secretions in the eyes)		
Immunological/Lymphoreticular							
48	Rat (Fischer- 344)	13 wk 5 d/wk 6 hr/d		77			Dodd et al. 1983
Neurological							
49	Rat (Fischer- 344)	13 wk 5 d/wk 6 hr/d		77			Dodd et al. 1983
Reproductive							
50	Rat (Fischer- 344)	13 wk 5 d/wk 6 hr/d		77 M			Dodd et al. 1983
CHRONIC EXPOSURE							
Systemic							
51	Human	1-6 yr	Hemato	0.6[d] M			Haufroid et al. 1997
			Hepatic	0.75 M			
			Renal	0.75 M			

Table 2-1. Levels of Significant Exposure to 2-Butoxyethanol - Inhalation (continued)

Key to figure[a]	Species/ (strain)	Exposure/ duration/ frequency	System	NOAEL (ppm)	LOAEL		Reference
					Less serious (ppm)	Serious (ppm)	

[a]The number corresponds to entries in Figure 2-1. Differences in levels of health effects and cancer effects between males and females are not indicated in Figure 2-1. Where such differences exist, only the levels of effect for the most sensitive gender are presented.

[b]Used to derive an acute inhalation minimal risk level (MRL) of 6.0 ppm. Concentration converted to an equivalent concentration in humans, and divided by an uncertainty factor of 9 (3 for extrapolation from animals to humans and 3 for human variability). For further details, see MRL worksheets in Appendix A.

[c]Used to derive an intermediate inhalation MRL of 3.0 ppm. Concentration converted to an equivalent concentration in humans, and divided by an uncertainty factor of 9 (3 for extrapolation from animals to humans and 3 for human variability). For further details, see MRL worksheets in Appendix A.

[d]Used to derive a chronic inhalation MRL of 0.2 ppm. Concentration divided by an uncertainty factor of 3 for human variability. Hematocrit significantly [p=0.03] decreased to 43.9%, MCHC significantly [p=0.02] increased to 33.6 g/dL. Changes in hematocrit and MCHC were within the range of normal clinical values and therefore were considered NOAELs. These effects were consistent with hemolysis seen in animal studies and may be an early indicator of potential adverse effects in humans. For further details, see MRL worksheets in Appendix A.

Bd Wt = body weight; Cardio = cardiovascular; d = day(s); Endocr = endocrine; F = female; Gastro = gastrointestinal; Gd = gestational day; Gn Pig = guinea pig; Hemato = hematological; HGB = hemoglobin; hr = hour(s); LC_{50} = lethal concentration; 50% kill; LOAEL = lowest-observable-adverse-effect level; M = male; MCH = mean corpuscular hemoglobin; MCHC = mean corpuscular concentration; MCV = mean corpuscular volume; min = minute(s); Musc/skel = musculoskeletal; NOAEL = no-observable-adverse-effect level; NS = not specified; RBC = red blood cell; Resp = respiratory; wk = week(s).

Table 2-2. Levels of Significant Exposure to 2-Butoxyethanol Acetate - Inhalation

Key to figure[a]	Species/ (strain)	Exposure/ duration/ frequency	System	NOAEL (ppm)	LOAEL Less serious (ppm)	LOAEL Serious (ppm)	Reference
ACUTE EXPOSURE							
Systemic							
1	Rat (Wistar)	4 hr	Resp	400			Truhaut et al. 1979
			Cardio	400			
			Hemato	400			
			Hepatic	400			
			Renal	400			
			Endocr	400			
2	Rabbit (New Zealand)	4 hr	Resp	400			Truhaut et al. 1979
			Cardio	400			
			Hemato		400 (slight and transient hemoglobinuria)		
			Hepatic	400			
			Renal		400 (slight and transient hematuria)		
			Endocr	400			
INTERMEDIATE EXPOSURE							
Death							
3	Rabbit (New Zealand)	1 mo 5 d/wk 4 hr/d				400 (death in 2/4)	Truhaut et al. 1979

Table 2-2. Levels of Significant Exposure to 2-Butoxyethanol Acetate - Inhalation (continued)

Key to figure[a]	Species/ (strain)	Exposure/ duration/ frequency	System	NOAEL (ppm)	LOAEL Less serious (ppm)	LOAEL Serious (ppm)	Reference
	Systemic						
4	Rat (Wistar)	1 mo 5 d/wk 4 hr/d	Resp	400			Truhaut et al. 1979
			Cardio	400			
			Hemato			400 (hemoglobinuria)	
			Hepatic	400			
			Renal		400 M (slight hematuria)	400 F (slight hematuria; in females tubular nephrosis, cellular cloudy swelling to hemorrhagic necrosis)	
			Endocr	400			
			Bd Wt	400			
5	Rat (Wistar)	10 mo 5 d/wk 4 hr/d	Resp	100			Truhaut et al. 1979
			Cardio	100			
			Hemato	100			
			Hepatic	100			
			Renal			100 M (discrete and inconstant lesions - tubular nephritis; enlargement; atrophy; inflammatory fibrosis; dilatated Henle's loop, distal convoluted tubules; hyaline casts)	
				100 F			
			Endocr	100			
			Bd Wt	100			

Table 2-2. Levels of Significant Exposure to 2-Butoxyethanol Acetate - Inhalation (continued)

Key to figure[a]	Species/ (strain)	Exposure/ duration/ frequency	System	NOAEL (ppm)	LOAEL Less serious (ppm)	LOAEL Serious (ppm)		Reference
6	Rabbit (New Zealand)	1 mo 5 d/wk 4 hr/d	Resp	400				Truhaut et al. 1979
			Cardio	400				
			Hemato			400	(28-59% decrease in RBC, 24-62% decrease in HGB concentration; pronounced hemoglobinuria)	
			Hepatic	400				
			Renal			400	(hypertrophy, necrotizing tubular nephrosis, atrophic tubular dilatation, luminar granular deposits; hematuria)	
			Endocr	400				
			Bd Wt	400				
7	Rabbit (New Zealand)	10 mo 5 d/wk 4 hr/d	Resp	100				Truhaut et al. 1979
			Cardio	100				
			Hemato	100				
			Hepatic	100				
			Renal			100	(tubular nephritis with tubular enlargement, cortical atrophy, inflammatory fibrosis, dilatation of Henle's loop and distal convoluted tubules)	
			Endocr	100				
			Bd Wt	100				
Immunological/Lymphoreticular								
8	Rat (Wistar)	1 mo 5 d/wk 4 hr/d		400				Truhaut et al. 1979

Table 2-2. Levels of Significant Exposure to 2-Butoxyethanol Acetate - Inhalation (continued)

Key to figure[a]	Species/ (strain)	Exposure/ duration/ frequency	System	NOAEL (ppm)	LOAEL Less serious (ppm)	LOAEL Serious (ppm)	Reference
9	Rat (Wistar)	10 mo 5 d/wk 4 hr/d		100			Truhaut et al. 1979
10	Rabbit (New Zealand)	10 mo 5 d/wk 4 hr/d		100			Truhaut et al. 1979
11	Rabbit (New Zealand)	1 mo 5 d/wk 4 hr/d		400			Truhaut et al. 1979
	Neurological						
12	Rat (Wistar)	1 mo 5 d/wk 4 hr/d		400			Truhaut et al. 1979
13	Rat (Wistar)	10 mo 5 d/wk 4 hr/d		100			Truhaut et al. 1979
14	Rabbit (New Zealand)	10 mo 5 d/wk 4 hr/d		100			Truhaut et al. 1979
15	Rabbit (New Zealand)	1 mo 5 d/wk 4 hr/d		400			Truhaut et al. 1979
	Reproductive						
16	Rat (Wistar)	1 mo 5 d/wk 4 hr/d		400			Truhaut et al. 1979

Table 2-2. Levels of Significant Exposure to 2-Butoxyethanol Acetate - Inhalation (continued)

Key to figure[a]	Species/ (strain)	Exposure/ duration/ frequency	System	NOAEL (ppm)	LOAEL		Reference
					Less serious (ppm)	Serious (ppm)	
17	Rat (Wistar)	10 mo 5 d/wk 4 hr/d		100			Truhaut et al. 1979
18	Rabbit (New Zealand)	10 mo 5 d/wk 4 hr/d		100			Truhaut et al. 1979
19	Rabbit (New Zealand)	1 mo 5 d/wk 4 hr/d		400			Truhaut et al. 1979

[a]The number corresponds to entries in Figure 2-2.

Bd Wt = body weight; Cardio = cardiovascular; d = day(s); Endocr = endocrine; F = female; Hemato = hematological; HGB = hemoglobin; hr = hour(s); LOAEL = lowest-observable-adverse-effect level; M = male; mo = month(s); NOAEL = no-observable-adverse-effect level; RBC = red blood cell; Resp = respiratory; wk = week(s).

Figure 2-1. Levels of Significant Exposure to 2-Butoxyethanol - Inhalation

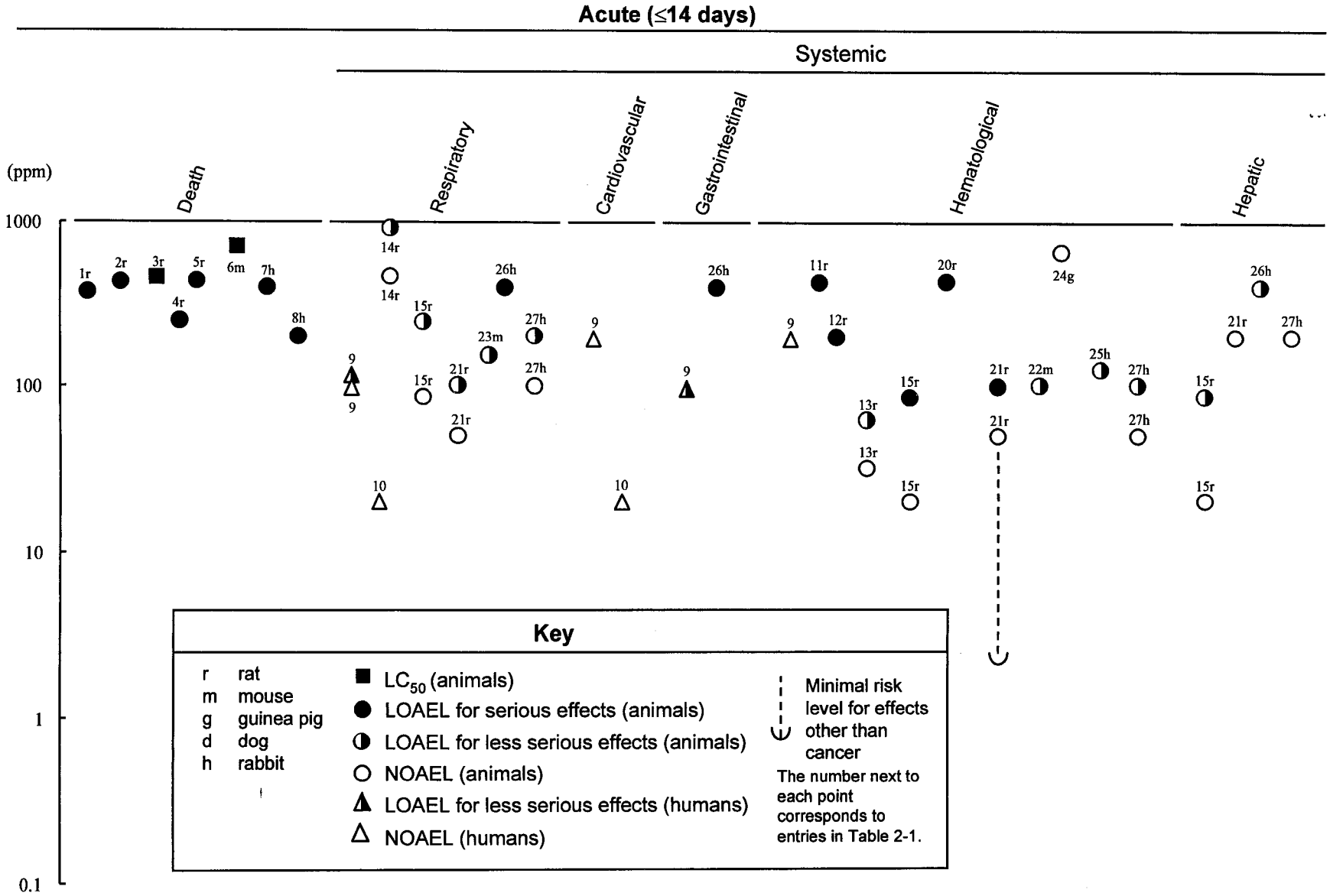


Figure 2-1. Levels of Significant Exposure to 2-Butoxyethanol - Inhalation (cont.)

Acute (≤14 days)

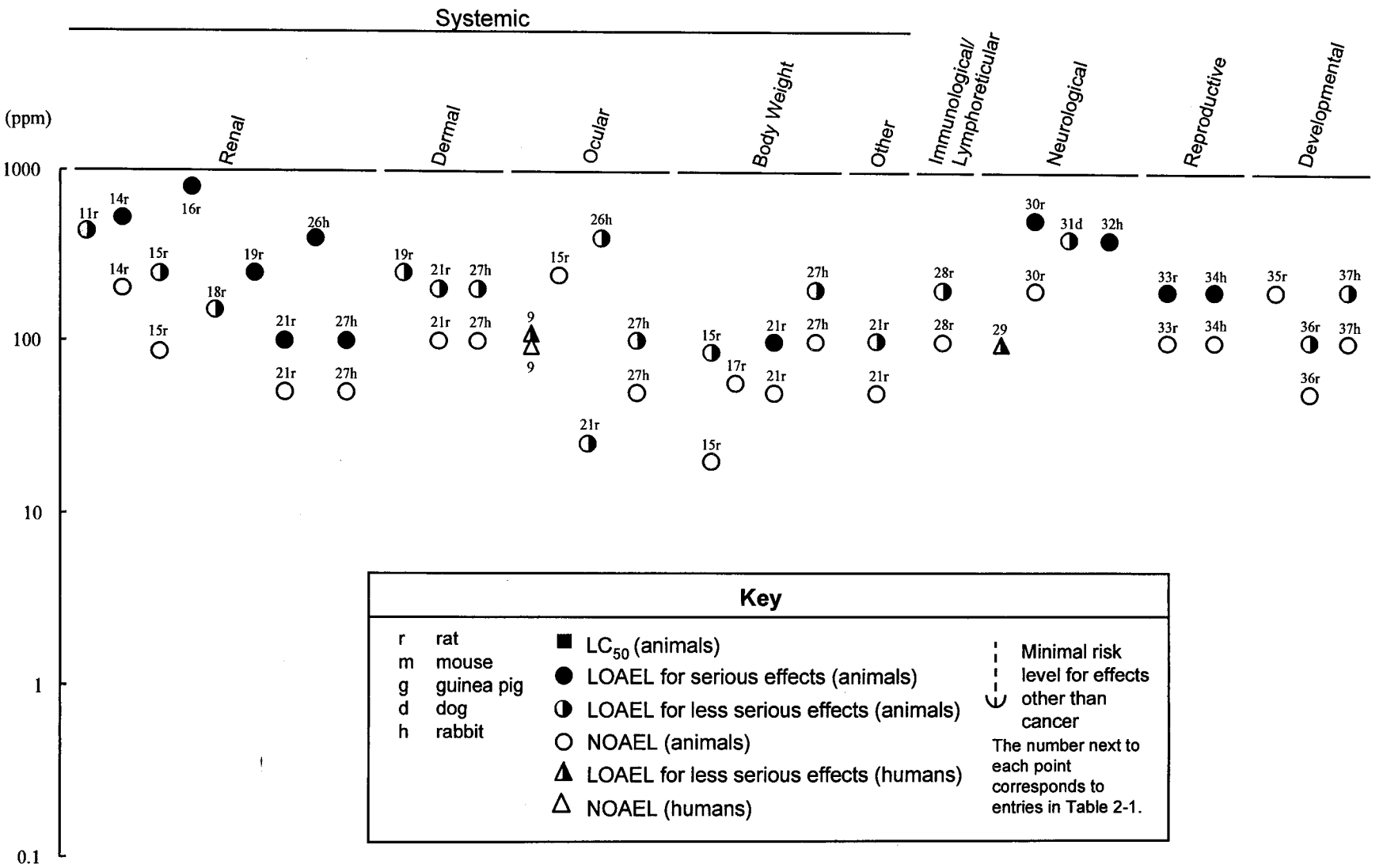


Figure 2-1. Levels of Significant Exposure to 2-Butoxyethanol - Inhalation

Intermediate (15-364 days)

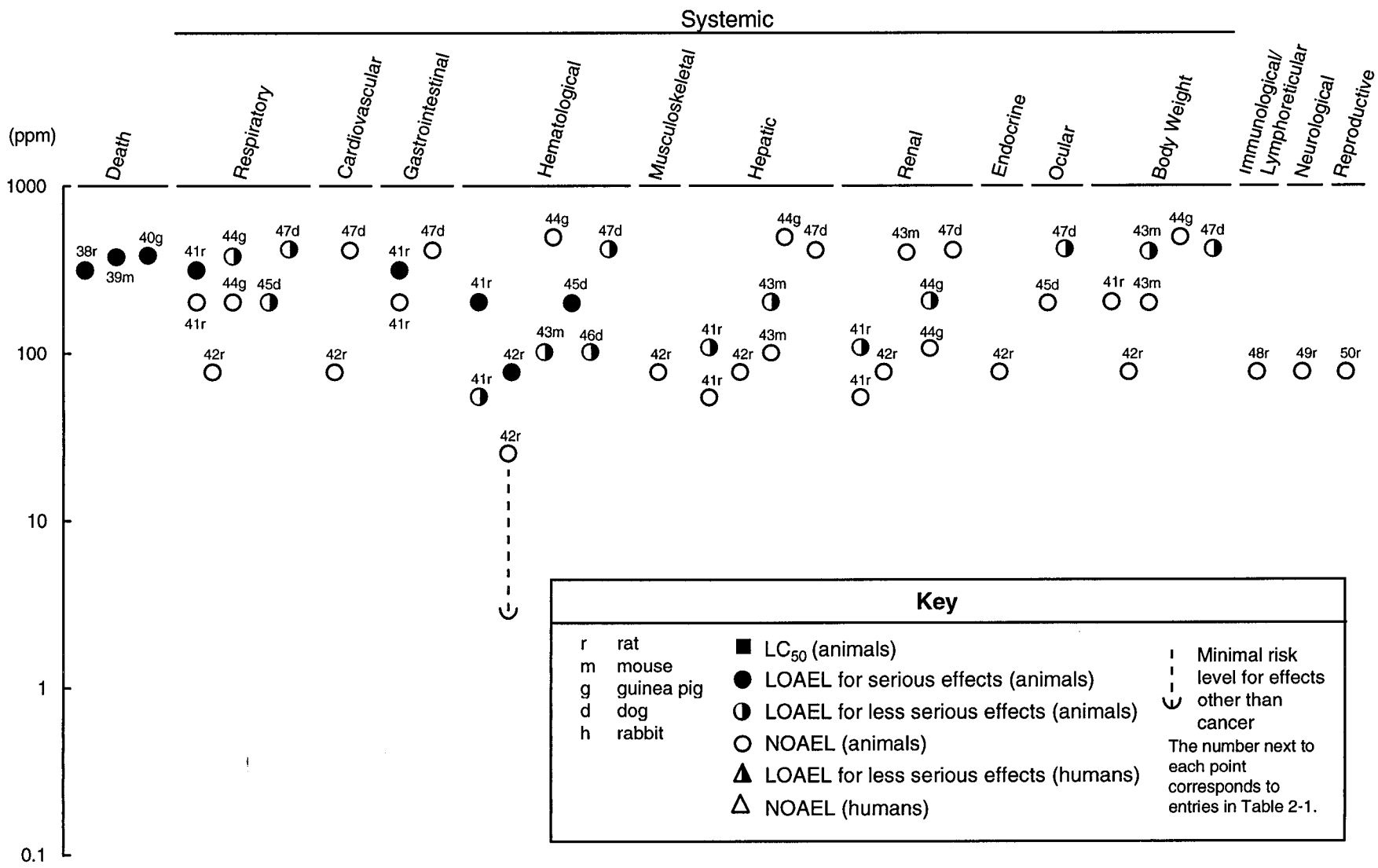


Figure 2-1. Levels of Significant Exposure to 2-Butoxyethanol - Inhalation

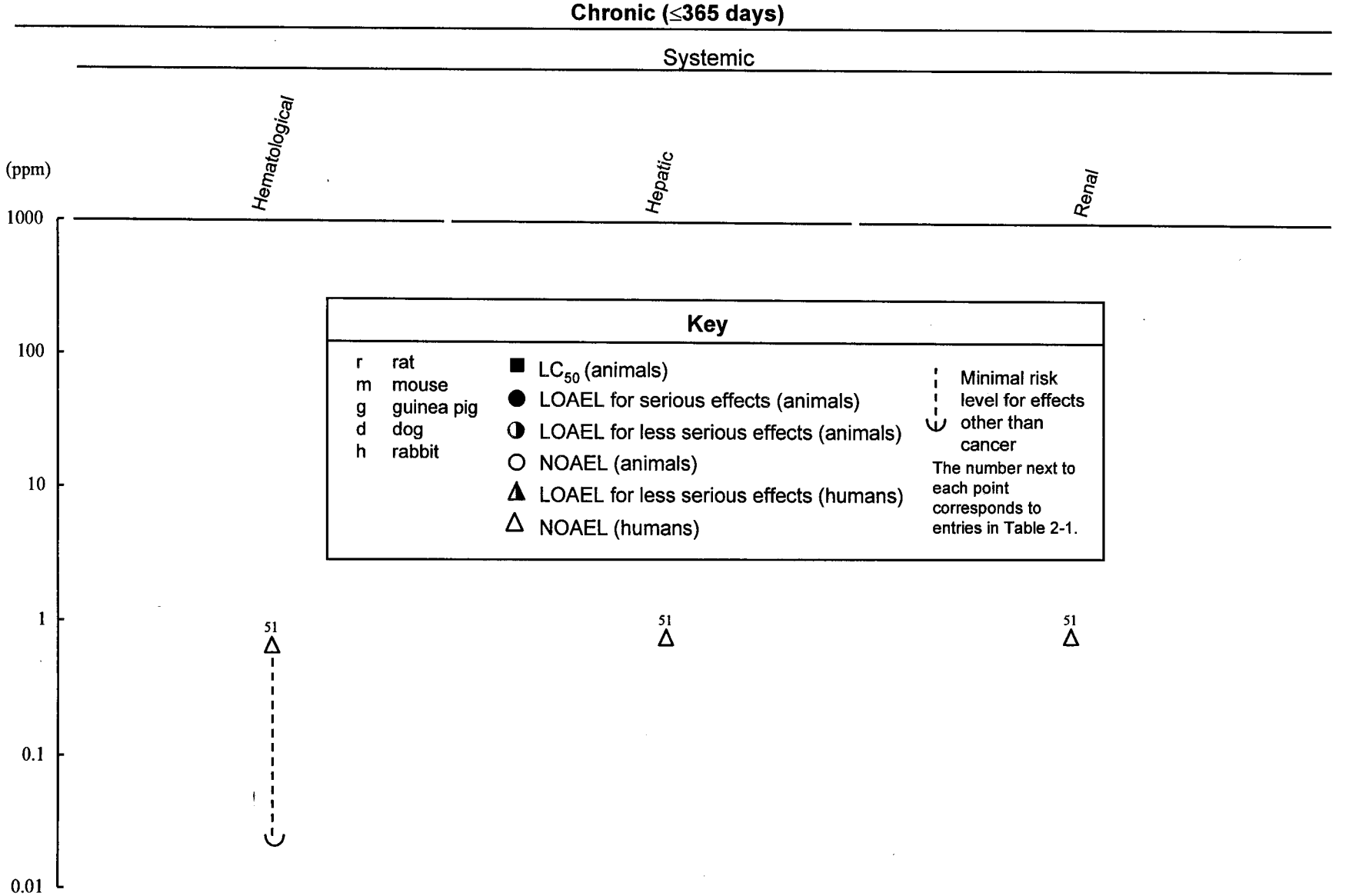


Figure 2-2. Levels of Significant Exposure to 2-Butoxyethanol Acetate - Inhalation

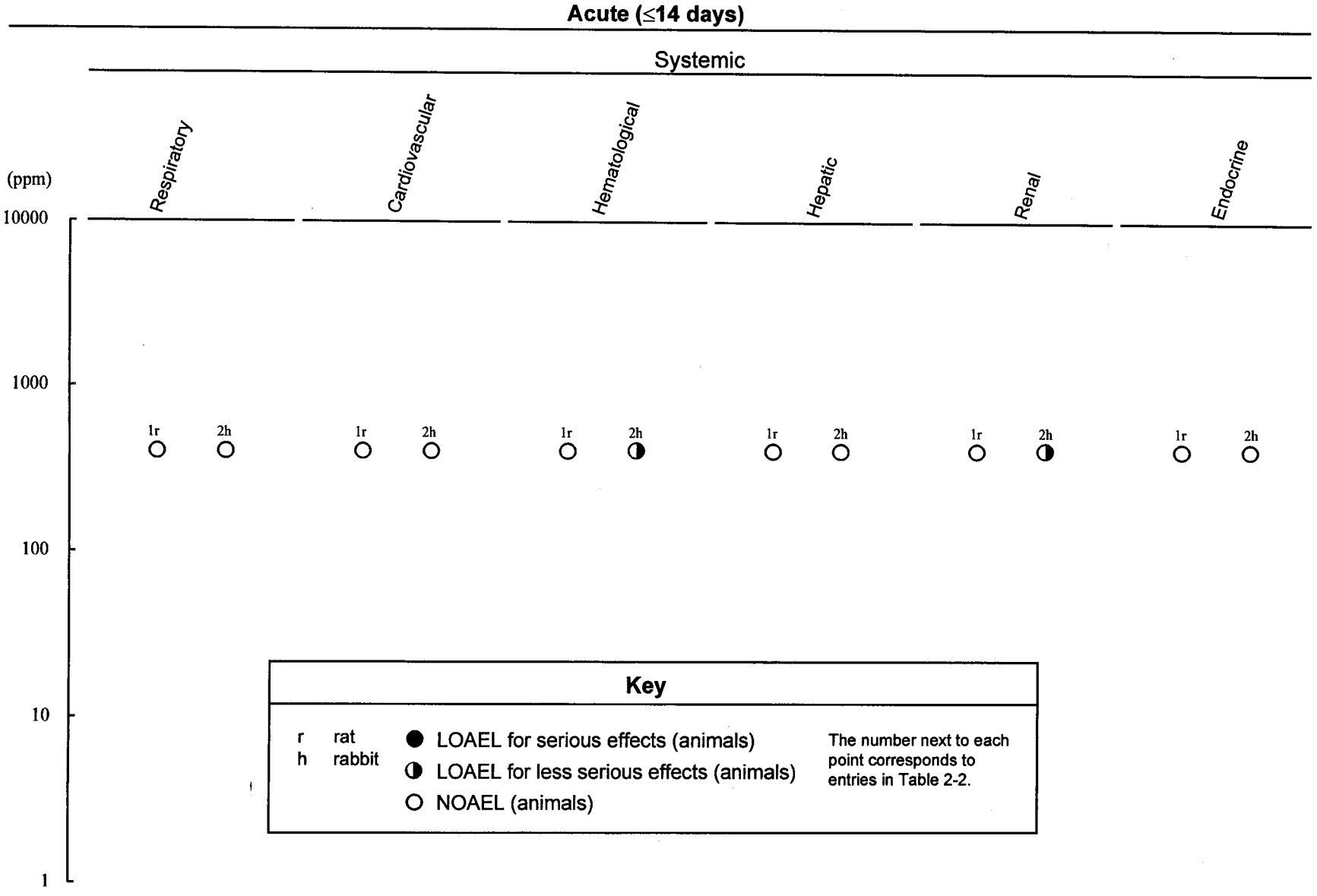


Figure 2-2. Levels of Significant Exposure to 2-Butoxyethanol Acetate - Inhalation (cont.)

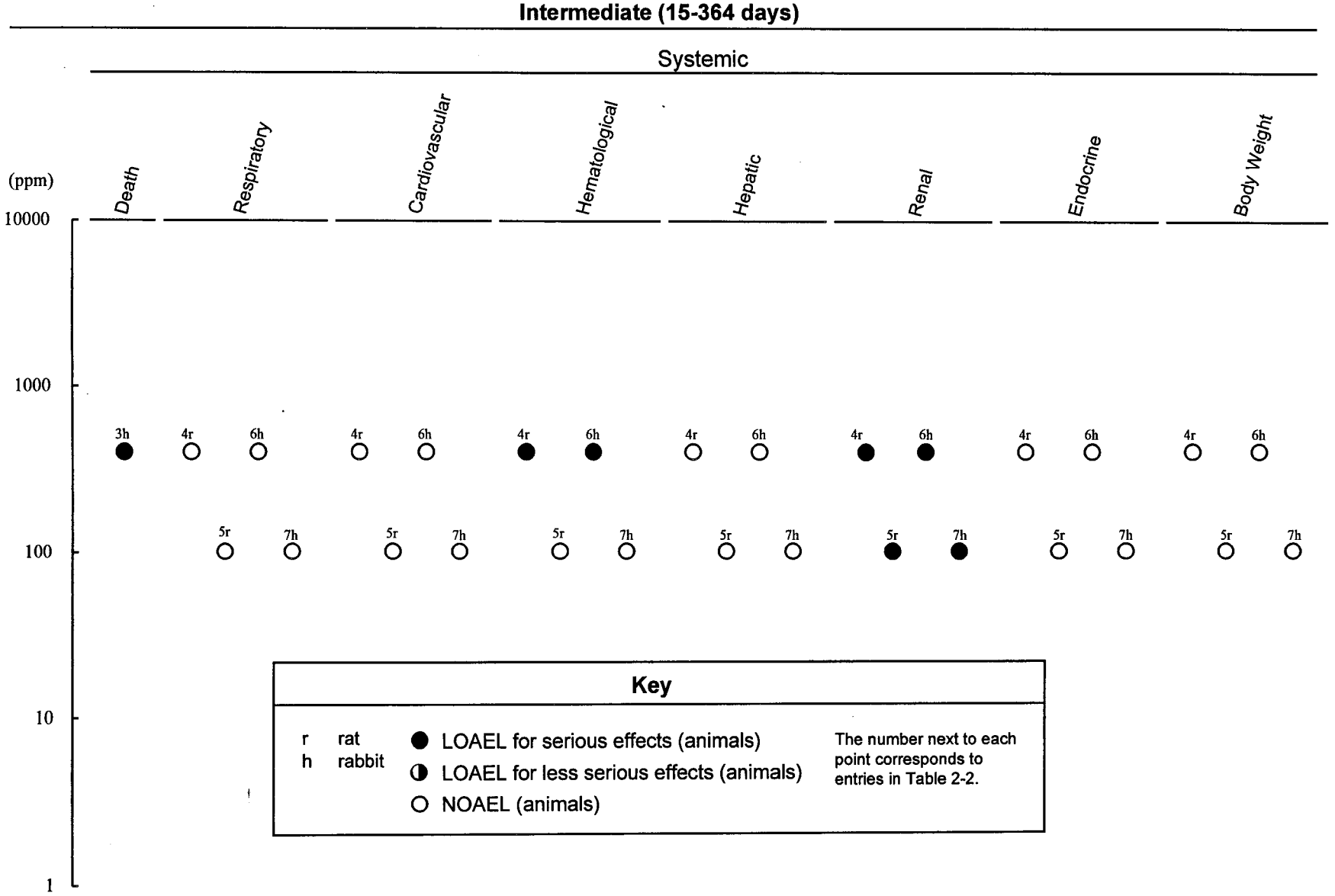
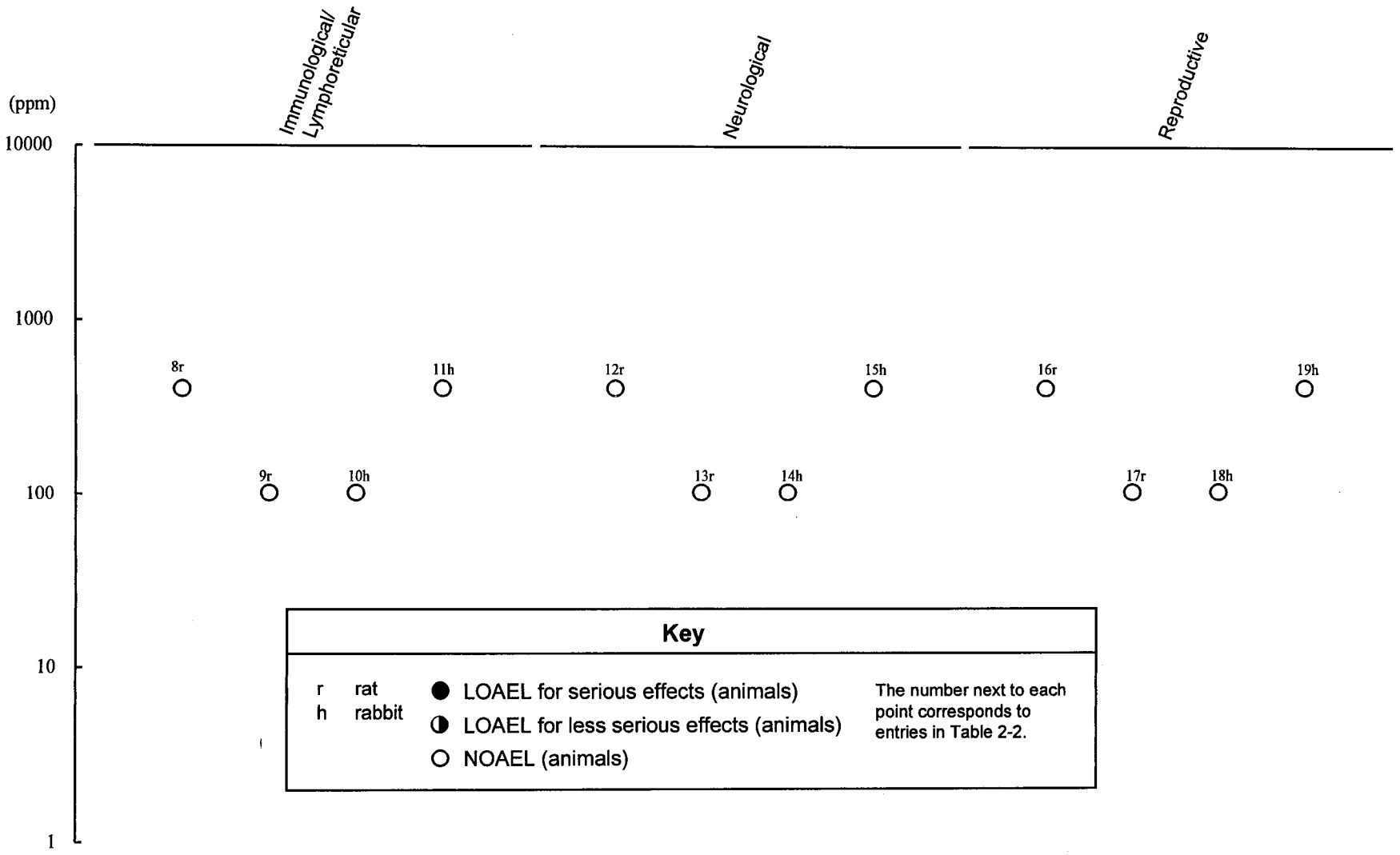


Figure 2-2. Levels of Significant Exposure to 2-Butoxyethanol Acetate - Inhalation (cont.)

Chronic (≥365 days)



2.2.1.2 Systemic Effects

No studies were located regarding musculoskeletal, endocrine, dermal, or body weight effects in humans after inhalation exposure to 2-butoxyethanol. No studies were located regarding any systemic effects in humans, or regarding gastrointestinal, musculoskeletal, dermal, or ocular effects in animals after inhalation exposure to 2-butoxyethanol acetate. Available data pertaining to systemic effects of each compound are presented below.

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

Respiratory Effects. Volunteers were exposed to 98, 113, or 195 ppm 2-butoxyethanol for 4-8 hours (Carpenter et al. 1956). The recorded responses of those exposed to 195 ppm included immediate irritation of the nose and throat. The subjects exposed to 113 ppm also experienced nasal irritation and a slight increase in nasal mucus discharge. In another study, no effects on pulmonary ventilation or respiratory frequency were found in seven male volunteers exposed to 2-butoxyethanol for 2 hours at the Swedish occupational exposure limit (20 ppm) during light physical exercise on a bicycle ergometer (Johanson et al. 1986a).

Respiratory effects have been observed in animals exposed to 2-butoxyethanol by inhalation for acute durations. In Fischer 344 rats exposed for 4 hours to 0, 202, 523, or 867 ppm 2-butoxyethanol, rapid and shallow breathing, followed by death, occurred at 867 ppm (Dodd et al. 1983). No respiratory effects were observed at ≤523 ppm. In the same study, Fischer 344 rats exposed for 9 days, 6 hours per day, 5 days per week to 2-butoxyethanol at 0, 20, 86, or 245 ppm exhibited audible respiration and nasal discharge in a few male animals only at 245 ppm (Dodd et al. 1983). Perinatal encrustation was noted in timed-pregnant female Fischer 344 rats exposed to 2-butoxyethanol concentrations of 100 or 200 ppm for 6 hours per day on gestational days 6-15 (Tyl et al. 1984). Male Swiss Webster mice exposed to a range of concentrations of 153-1,666 ppm 2-butoxyethanol for 10-15 minutes exhibited a 20% decrease in respiratory rate at the lowest concentration and a 40% decrease at the highest concentration (Kane et al. 1980). The decrease in respiration rate occurred during the first few seconds of exposure and is an indicator of respiratory irritation. Male albino rabbits exposed to approximately 400 ppm 2-butoxyethanol for l-2 days at 7 hours per day exhibited nasal discharge and congestion of the lungs and nasal turbinates upon necropsy (Dow 1986).

Wetness around the nose was observed in pregnant New Zealand white rabbits exposed to 200 ppm 2-butoxyethanol for 6 hours per day on gestational days 6-l 8 (Tyl et al. 1984). One female dog exposed repeatedly to 617 ppm 2-butoxyethanol for a total of 13.5 hours over 2 days exhibited moderate congestion of the lungs upon necropsy (Carpenter et al. 1956). Similarly, a female dog exposed to 385 ppm 2-butoxy-ethanol for 8 days died and was found to have severely congested and hemorrhagic lungs at necropsy.
Clinical signs and gross necropsy observations did not reveal any adverse respiratory effects in guinea pigs exposed to 691 ppm (females) or 633 ppm (males) 2-butoxyethanol for 1 hour followed by a 14-day observation period (Nachreiner 1994).

In intermediate-duration studies of 2-butoxyethanol, Fischer 344 rats exposed intermittently to 77 ppm for 13 weeks showed no effects on lung weight and no histopathological lesions in the lungs (Dodd et al. 1983), while Sherman rats exposed to 2-butoxyethanol for 7 hours per day, 5 days per week, for a total 30 days exhibited congested and hemorrhagic lungs at necropsy at 432 ppm (males) and 314 ppm (females) (Carpenter et al. 1956). Guinea pigs also exhibited lung congestion at necropsy after intermittent exposure to ≥ 376 ppm 2-butoxyethanol for 30 days (Carpenter et al. 1956). Severely congested and hemorrhagic lungs were found in one dog exposed to 385 ppm for 28 days, and slight capillary engorgement or breakdown in the lungs was observed upon gross necropsy (not confirmed by histological examination) of two dogs exposed to 200 ppm for 31 days (Carpenter et al. 1956). Slightly increased nasal secretions were observed in two dogs exposed to 415 ppm 2-butoxyethano17 hours per day, 5 days per week, for 12 weeks (Werner et al. 1943b). One rhesus monkey exposed to 210 ppm 2-butoxyethanol for up to 30 days and two rhesus monkeys exposed to 100 ppm for 90 days exhibited no respiratory effects (Carpenter et al. 1956).

In a study of 2-butoxyethanol acetate, Wistar rats and New Zealand white rabbits were exposed to a saturated vapor-air mixture of approximately 400 ppm for 4 hours, and then observed for 14 days after exposure (Truhaut et al. 1979). No clinical signs of respiratory distress were noted for either species. When the animals were sacrificed at the end of 2 weeks, no gross pathological lesions were observed in respiratory tissues of either rats or rabbits. No histological examinations were performed. In the intermediate-duration experiments in this study, in which both gross and histological examinations were performed, no respiratory effects were noted in either rats or rabbits after intermittent exposure (5 days per week, 4 hours per day) to 400 ppm 2-butoxyethanol acetate for 1 month or to 100 ppm 2-butoxyethanol acetate for 10 months (Truhaut et al. 1979).

Cardiovascular Effects. No adverse cardiovascular effects were observed in male volunteers exposed to 20 ppm 2-butoxyethanol for 2 hours during light physical exercise on a bicycle ergometer (Johanson et al. 1986a). Specifically, no effects of exposure were noted for heart rate, and no effects were noted in the electrocardiograms. In an earlier study, male and female volunteers exposed to 98, 113, or 195 ppm 2-butoxyethanol for 4-8 hours showed no adverse effects on blood pressure or pulse rate (Carpenter et al. 1956).

Gross necropsy did not reveal any changes in the hearts of guinea pigs exposed to 2-butoxyethanol for 1 hour followed by a 14-day observation period at 691 ppm for females and 633 ppm for males (Nachreiner 1994). No gross or histological lesions in the heart were observed in male or female Fischer 344 rats exposed to 77 ppm 2-butoxyethanol for 6 hours per day, 7 days per week, for 13 weeks (Dodd et al. 1983) or in two dogs exposed to 415 ppm 7 hours per day, 5 days per week, for 12 weeks (Werner et al. 1943b)

In a study of 2-butoxyethanol acetate, Wistar rats and New Zealand white rabbits were exposed to a saturated vapor-air mixture of approximately 400 ppm 2-butoxyethanol acetate for 4 hours and were then observed for 14 days after exposure (Truhaut et al. 1979). When the animals were sacrificed at the end of 2 weeks, no gross pathological lesions were observed in the hearts of either rats or rabbits. No histological examinations were performed. In the intermediate-duration experiments in this study, in which both gross and histological examinations were performed, no lesions in the heart were noted in either rats or rabbits after intermittent exposure (5 days per week, 4 hours per day) to 400 ppm 2-butoxyethanol acetate for 1 month or to 100 ppm 2-butoxyethanol acetate for 10 months (Truhaut et al. 1979).

Gastrointestinal Effects. Two male volunteers exposed to 113 ppm 2-butoxyethanol for 4 hours experienced occasional belching (eructation), while a female volunteer exposed to 98 ppm for 8 hours experienced emesis after 7 hours of exposure and several times on the following day, although other male and female volunteers exposed to the same or higher concentrations did not react similarly (Carpenter et al. 1956). The subject who experienced emesis, however, believed that the emesis was due to the relatively high chamber temperature based on her personal past experience that high temperature often caused emesis.

One female dog, exposed repeatedly to 617 ppm 2-butoxyethanol for a total of 13.5 hours during 2 days, experienced emesis on both days of exposure (Carpenter et al. 1956). Similarly, a female dog exposed to 385 ppm 2-butoxyethanol for 8 days experienced emesis several times during the first 4 days of exposure and died on the 8th day (Carpenter et al. 1956). Gross examination of the gastrointestinal tract did not reveal

adverse effects in guinea pigs exposed to 2-butoxyetbanol at 691 ppm (females) or 633 ppm (males) for 1 hour followed by a 14-day observation period (Nachreiner 1994). Male albino rabbits exposed to approximately 400 ppm 2-butoxyethanol for 1-2 days at 7 hours per day exhibited hemorrhagic gastric ulcers upon necropsy (Dow 1986). One rhesus monkey exposed to 210 ppm 2-butoxyethanol for up to 30 days exhibited emesis four times during the latter part of the exposure period (Carpenter et al. 1956). Sherman rats exposed to 2-butoxyethanol for 7 hours per day, 5 days per week, for a total 30 days exhibited congestion of the abdominal viscera at necropsy at 432 ppm for males and 314 ppm for females (Carpenter et al. 1956). Emesis and histopathological changes in the small and large intestines were not observed in two dogs exposed to 2-butoxyethanol at 415 ppm for 7 hours per day, 5 days per week, for 12 weeks (Werner et al. 1943b).

Hematological Effects. Although hematological effects of 2-butoxyethanol exposure in animals include hemolysis, hemoglobinuria, and hemoglobin in the urine resulting from lysed red blood cells (Jones and Hunt 1983), the only information regarding hematological effects in humans after inhalation exposure to 2-butoxyethanol is negative. Male and female volunteers exposed to 98, 113, or 195 ppm 2-butoxyethanol for 4-8 hours showed no adverse effect on erythrocyte osmotic fragility (Carpenter et al. 1956).

Red blood cell counts, hemoglobin, hematocrit, mean corpuscular volume, mean corpuscular hemoglobin, mean corpuscular hemoglobin concentration, heptoglobin, reticulocytes, and osmotic resistance were measured in 31 male workers exposed to 2-butoxyethanol for l-6 years (Haufroid et al. 1997). Twenty of the workers were exposed to an average concentration of 0.75 ppm, and 11 workers were exposed to an average concentration of 0.46 ppm. The geometric mean exposure concentration for the 31 workers was 0.6 ppm. The workers were also exposed to unspecified concentrations of methyl ethyl ketone. Studies in animals indicate that methyl ethyl ketone does not produce hematologic effects (ATSDR 1992). Twenty-one unexposed workers from the same plant matched for sex, age and smoking habits served as controls; however, 2-butoxyethanol concentrations in the air the controls breathed was not assayed. Urine was collected before the shift and at the end of the shift and assayed for free 2-butoxyacetic acid (thought to be the toxic metabolite), retinol binding protein, and creatinine. Rettenmeier et al. (1993) and Sakai et al. (1994) found that some of the 2-butoxyacetic acid (BAA) in the urine of humans is excreted as amino acid conjugates. Thus, free BAA in urine does not represent all internal dosing. Blood was collected and assayed for red blood cell counts, hemoglobin, hematocrit, mean corpuscular volume, mean corpuscular hemoglobin, mean corpuscular hemoglobin concentration, haptoglobin, reticulocytes, and osmotic resistance. Serum alanine aminotransferase and aspartate aminotransferase were also measured. The exposed workers wore

gloves, thus minimizing dermal exposure. In addition, two exposure groups were combined together for the analysis of measured parameters. A significant correlation ($r = 0.55$, $p = 0.0012$) was observed between end shift free urinary 2-butoxyacetic acid concentrations and 2-butoxyethanol in air. There was no effect on red blood cell numeration, hemoglobin, mean corpuscular volume, mean corpuscular hemoglobin, heptoglobin, reticulocyte (aspartate aminotransferase, alanine aminotransferase) and renal (plasma creatinine, urinary retinol binding protein) parameters. Two small but statistically significant differences in hematology values were observed: a significant decrease ($p=0.03$) in hematocrit values (exposed 43.9%±2.1, range: 39.9-50.7; controls: 45.5%±2.7, range: 40.6-50.4) and a significant ($p=0.02$) increase in mean corpuscular hemoglobin concentration (33.6 g/dL±0.9, range: 31.8-35.6; 32.9 g/dL±l.1, range: 31.1-35.6). The differences may be consistent with hemolysis observed in animal studies, and may be early indicators of potential adverse effects in humans; however, because the changes in both hematocrit and mean corpuscular hemoglobin concentration were in the range of normal clinical values, the effect was considered a NOAEL. Normal clinical values of hematocrit are reported for males as 40-54%, with a level of 30% or less considered to indicate moderate to severe anemia (Fischbach 1992). Normal clinical values of mean corpuscular hemoglobin concentration are reported as 31-37 g hemoglobin/dL (Fischbach 1992). Increased mean corpuscular hemoglobin concentrations often indicate spherocytosis, while decreased mean corpuscular hemoglobin concentrations may indicate macrocytic anemia, chronic blood loss anemia, or pyridoxine-responsive anemia (Fischbach 1992). The study author stated that the usefulness of hematocrit and mean corpuscular hemoglobin concentration as early markers of red blood cell toxicity in workers exposed to low concentrations of 2-butoxyethanol requires further study. Based on the NOAEL of 0.6 ppm for decreased hematocrit and increased mean corpuscular hemoglobin concentration, a chronic-duration inhalation MRL of 0.2 ppm was calculated as described in the footnote to Table 2-l and in Appendix A.

Hematological effects of 2-butoxyethanol have been observed in animals after acute and intermediate inhalation exposure. One male monkey and one female monkey exposed to 200 ppm 2-butoxyethanol for 7 hours did not show any alteration of erythrocyte fragility or evidence of hemoglobinuria (Carpenter et al. 1956). However, one rhesus monkey exposed to 210 ppm 2-butoxyethanol for up to 30 days exhibited elevated osmotic fragility after the fourth exposure, increased fibrinogen after the 14th day, and reduced erythrocyte count and hemoglobin values after 30 exposures (Carpenter et al. 1956). One male monkey and one female monkey exposed to 100 ppm for 90 days had transiently increased erythrocyte osmotic fragility and decreased numbers of erythrocytes .

Female rats exposed to 432 ppm 2-butoxyethanol for 2-8 hours exhibited hemolysis at 2 hours, hemoglobinuria at 3 hours, and hemin crystalluria at 4 hours during exposure, whereas rats exposed to 200 ppm for 7 hours per day for 9 days exhibited a decrease in red blood cell count and hemoglobin level (Carpenter et al. 1956). Elevation of erythrocyte fragility was seen at doses as low as 62 ppm administered to rats for 4 hours, or 54 ppm administered for up to 30 days (Carpenter et al. 1956). In addition, Carpenter et al. (1956) reported hemoglobinuria in rats after inhalation exposure to 203 ppm 2-butoxyethanol for 7 hours; in rats exposed to 250-800 ppm for 2-9 hours per day over l-6 days; in rats exposed to 203 ppm for 7 hours, 5 days per week for 30 days; and in mice after inhalation exposure to 200 ppm 2-butoxyethanol for 7 hours. Hemoglobinuria was noted in male Fischer 344 rats after 6 hours of exposure to 438 ppm (Sabourin et al. 1992a).

In a study in which mice were exposed to 390-1210 ppm 2-butoxyethanol for 7 hours hemoglobinuria was reported (Werner et al. 1943a). The concentrations at which hemoglobinuria was observed were not stated. Mice exhibited increased osmotic fragility of the red blood cells after 7 hours of exposure to 100 ppm 2-butoxyethanol, as did rabbits after exposure to 125 ppm. However, guinea pigs showed no effect of treatment on erythrocyte fragility after exposure to 665 ppm for 8 hours or to ≤c494 ppm for 7 hours per day 5 days per week for 30 days, and dogs showed no effect after exposure to 665 ppm for 7 hours per day for 2 days (Carpenter et al. 1956). C3H mice exhibited increased erythrocyte fragility after exposure to ≥ 100 ppm for 30-90 days, as did one female dog exposed to 385 ppm for 8 days and one male dog exposed to 385 ppm for 28 days (Carpenter et al. 1956). Although the increase in erythrocyte fragility in the dogs was transient, increases in leucocyte count in the male and female dogs and fibrinogen levels in the male dog were also seen (Carpenter et al. 1956). In other experiments, increased osmotic fragility and leucocyte count were seen in one male dog and one female dog after exposure to 200 ppm for 31 days, and increased leucocyte counts were seen after 45 days of exposure to 100 ppm; decreased hematocrit was noted in the male dog throughout the exposure period of 90 days (Carpenter et al. 1956). Decreased hemoglobin and hematocrit, hypochromia, polychromatophilia, and microcytosis were observed in two dogs exposed to 415 ppm 2-butoxyethanol 7 hours per day, 5 days per week, for 12 weeks (Werner et al. 1943b).

Fischer 344 rats were exposed for 9 days, 6 hours per day, 5 days per week to 2-butoxyethanol at 0, 20, 86, or 245 ppm (Dodd et al. 1983). The rats were observed for signs of toxicity during exposure and for 14 days postexposure. There was no effect on the hematologic parameters in rats exposed to 20 ppm 2-butoxyethanol. At the 86-ppm exposure, both sexes exbibited a significant effect on erythroid parameters, including decreased hemoglobin and increased mean corpuscular volume. In addition, mean corpuscular hemoglobin

concentration was decreased in females at 86 ppm. At the 245-ppm exposure, both male and female rats showed significantly depressed red blood cell count, hemoglobin, and mean corpuscular hemoglobin concentration and significant increases in mean corpuscular volume, nucleated red blood cell count, and reticulocytes. A 14-day postexposure recovery period showed substantial recovery (Dodd et al. 1983). Timed-pregnant Fischer 344 rats exposed to 2-butoxyethanol vapors by inhalation on gestational days 6-15 at concentrations of 0, 25, 50, 100, or 200 ppm for 6 hours per day exhibited hemoglobinuria, reduced red blood cell count, reduced mean corpuscular hemoglobin concentration, increased mean corpuscular hemoglobin, and increased mean corpuscular volume at 100 ppm, but not at ≤ 50 ppm (Tyl et al. 1984). Hemoglobin concentration and hematocrit were increased at 200 ppm. In the same study, pregnant New Zealand white rabbits exposed on gestational days 6-l 8 to the same concentrations exhibited increased hemoglobin and hematocrit at 100 ppm but not at 200 ppm, suggesting to the authors a biphasic response of the hematological system in rabbits (Tyl et al. 1984). Based on the NOAEL of 50 ppm for hematological effects in rats in the study by Tyl et al. (1984), an acute inhalation MRL of 6 ppm for 2-butoxyethanol was calculated as described in the footnote to Table 2-l and in Appendix A.

When rats were exposed to 5, 25, or 77 ppm 2-butoxyethanol 5 days per week, 6 hours per day, for 13 weeks, decreased red blood cell count was observed at 77 ppm in males and females; females also exhibited decreased hemoglobin and mean corpuscular hemoglobin concentration at 77 ppm (Dodd et al. 1983). Thus, 25 ppm is the NOAEL for hematological effects in rats after intermediate-duration inhalation exposure to 2-butoxyethanol. Based on this value, an intermediate inhalation MRL of 3.0 ppm for 2-butoxyethanol was calculated as described in the footnote to Table 2-l and in Appendix A.

In a study of 2-butoxyethanol acetate, Wistar rats and New Zealand white rabbits were exposed to a saturated vapor-air mixture of approximately 400 ppm 2-butoxyethanol acetate for 4 hours and then observed for 14 days after exposure (Truhaut et al. 1979). No persistent hematological effects were noted for either species, although rabbits exhibited slight, transient hemoglobinuria. When the animals were sacrificed at the end of 2 weeks, no adverse changes in hematology were observed in either rats or rabbits. In the same study, hemoglobinuria was noted in rats beginning 2 weeks after initiation of intermittent exposure (5 days per week, 4 hours per day) to 400 ppm 2-butoxyethanol acetate for 1 month, but no effect was noted in rats or rabbits similarly exposed to 100 ppm for 10 months (Truhaut et al. 1979). Rabbits exposed to 400 ppm 2-butoxyethanol acetate for 1 month exhibited decreased red blood cell counts, decreased hemoglobin concentration, and pronounced hemoglobinuria (Truhaut et al. 1979).

The hemolytic effects of 2-butoxyethanol and 2-butoxyethanol acetate are reflected in changes in the spleen. These effects are discussed further in Section 2.2.1.3, Immunological and Lymphoreticular Effects.

Musculoskeletal Effects. Gross lesions in bone (femur, vertebrae, joint, sternum) or skeletal muscle were not observed in guinea pigs exposed to 2-butoxyethanol for 1 hour followed by a 14-day observation period at 661 ppm for females and 633 ppm for males (Nachreiner 1994). In Fischer 344 rats exposed to 0, 5, 25, or 77 ppm 2-butoxyethanol for 6 hours per day, 5 days per week for 13 weeks, no gross of histopathological lesions were found in the gastrocnemius muscle or in the sternum (Dodd et al. 1983).

Hepatic Effects. No effects on serum alanine aminotransferase or aspartate aminotransferase were observed when compared to controls in 31 male workers exposed to 2-butoxyethanol for l-6 years (Haufroid et al. 1997). Twenty of the workers were exposed to an average concentration of 0.75 ppm, and 11 workers were exposed to an average concentration of 0.46 ppm. The workers were also exposed to unspecified concentrations of methyl ethyl ketone. Twenty-one unexposed workers from the same plant matched for sex, age and smoking habits served as controls.

Fischer 344 rats exposed for 9 days, 6 hours per day, 5 days per week to 2-butoxyethanol at 0, 20, 86, or 245 ppm showed increased liver weight at ≥ 86 ppm in females and at 245 ppm in males (Dodd et al. 1983). Timed-pregnant Fischer 344 rats exposed to 2-butoxyethanol vapors by inhalation on gestational days 6-15 and New Zealand white rabbits exposed on gestational days 6-18 to concentrations of 0, 25, 50, 100, or 200 ppm for 6 hours per day exhibited no effect on liver weight (Tyl et al. 1984). Gross lesions in the liver were not observed in guinea pigs exposed to 2-butoxyethanol (at 661 ppm for females and 633 ppm for males) for 1 hour followed by a 14-day observation period (Nachreiner 1994). In male albino rabbits exposed to 400-411 ppm 7 hours per day for 1 or 2 days, mottled livers were observed upon necropsy (Dow 1986). Congestion of the liver was observed at necropsy in a female dog that died following exposure to 385 ppm 2-butoxyethanol for 8 days and in a male dog exposed to the same concentration for 28 days (Carpenter et al. 1956). Only one female and one male dog (both exposed to 385 ppm) were used in this study.

In intermediate-duration studies, a statistically significant increase in liver weight was observed in Sherman rats exposed to ≥107 ppm, 7 hours per day, 5 days per week for 30 days (Carpenter et al. 1956). Cloudy swelling of the liver was observed in the rats exposed to 314 ppm (female) and 432 ppm (male and female). In C3H mice exposed for 7 hours per day, a statistically significant increase in liver weight was observed at

200 ppm after 60 exposure days (but not after 30 or 90 days) and at 400 ppm after 30, 60, and 90 days (Carpenter et al. 1956). However, no effects on liver weight and no gross or histologically observed liver lesions were found in guinea pigs exposed to <495 ppm for 7 hours per day, 5 days per week for 30 days. Gross or histopathological changes in the liver were not observed in two dogs exposed to 415 ppm 2-butoxyethanol 7 hours per day, 5 days per week for 12 weeks (Werner et al. 1943b). No effects on clinical blood parameters and no gross or histopathological lesions were found in the livers of Fischer 344 rats exposed to ≤77 ppm 2-butoxyethanol for 6 hours per day, 5 days per week, for 13 weeks (Dodd et al. 1983).

In a study of 2-butoxyethanol acetate, Wistar rats and New Zealand white rabbits exposed to a saturated vapor-air mixture of approximately 400 ppm 2-butoxyethanol acetate for 4 hours, and then observed for 14 days after exposure, had no gross hepatic pathological lesions when the animals were sacrificed at the end of 2 weeks (Truhaut et al. 1979). In the same study, no gross or histologically observed hepatic lesions were noted in either rats or rabbits after intermittent exposure (5 days per week, 4 hours per day) to 400 ppm for 1 month or 100 ppm for 10 months.

Renal Effects. No effects on serum creatinine or urinary retinol binding protein were observed in 31 male workers exposed to 2-butoxyethanol for l-6 years (Haufroid et al. 1997). Twenty of the workers were exposed to an average concentration of 0.75 ppm, and 11 workers were exposed to an average concentration of 0.46 ppm. The workers were also exposed to unspecified concentrations of methyl ethyl ketone. Twentyone unexposed workers from the same plant matched for sex, age and smoking habits served as controls.

A common renal effect of 2-butoxyethanol exposure is hematuria, the detection of red blood cells in the urine (Jones and Hunt 1983). Section 2.4.2 Mechanisms of Toxicity discusses the mechanism by which this effect may occur. When Fischer 344 rats were exposed for 4 hours to 0, 202, 523, or 867 ppm 2-butoxyethanol, red discharge around the urogenital area was detected at concentrations of 523 and 857 ppm (Dodd et al. 1983). Additionally, rats in the high- and mid-dose groups that died and were necropsied after death had red-stained urine in their bladders (presumably hematuria) and enlarged kidneys. This effect was not observed in the surviving rats in the 523-ppm group necropsied 14 days after exposure ceased. It is likelythat the red staining observed in the urogenital area occurred because of hematuria. In the same study, Fischer 344 rats were exposed for 9 days, 6 hours per day, 5 days per week to 2-butoxyethanol at 0, 20, 86, or 245 ppm (Dodd et al. 1983). Animals at the high concentration also exhibited red-stained urine (hematuria) after one or two treatments, which was not observed in the postexposure period of 14 days (Dodd et al. 1983). Hematuria was noted in male Wistar-derived (Alpk/Ap) rats exposed to 800 ppm for 3 hours (Doe 1984), in

nonpregnant female Sprague-Dawley rats exposed to 250 ppm 2-butoxyethanol for up to 7 hours, and on the first exposure day in pregnant Sprague-Dawley rats exposed to 150 and 200 ppm for 7 hours per day on gestational days 7-15 (Nelson et al. 1984). Timed-pregnant Fischer 344 rats exposed by inhalation to 2-butoxyethanol vapor at concentrations of 0, 25, 50, 100, or 200 ppm for 6 hours per day on gestational days 6-15 exhibited hematuria at 100 and 200 ppm (Tyl et al. 1984). In the same study, red fluid on the tray paper, possibly evidence of hematuria, was observed in timed-pregnant New Zealand rabbits exposed to 100 and 200 ppm 6 hours per day during gestation. Male albino rabbits exposed to approximately 400 ppm 2-butoxyethanol for l-2 days at 7 hours per day exhibited darkened kidneys and hematuria (Dow 1986).

Cloudy swelling of the convoluted tubules was observed in female rats exposed to 432 ppm 2-butoxyethanol for 2 hours (Carpenter et al. 1956). No effect on urine output was seen in male rats exposed to 20 or 100 ppm 2-butoxyethanol for 12 days (Johanson 1994), or on kidney weight in pregnant rabbits exposed to doses up to 200 ppm during gestation (Tyl et al. 1984). In a study in which Swiss mice were exposed to 390-1,210 ppm 2-butoxyethanol for 7 hours, interstitial nephritis was reported (Werner et al. 1943a). The concentrations at which nephritis was observed were not stated. Gross lesions in the kidneys were not observed in guinea pigs exposed to 2-butoxyethanol for 1 hour followed by a 14-day observation period at 661 ppm for females and 633 ppm for males (Nacbreiner 1994). Gne female dog, exposed repeatedly to 617 ppm 2-butoxyethanol for a total of 13.5 hours over 2 days exhibited congestion of the kidneys upon necropsy (Carpenter et al. 1956). Similar resuhs were obtained in a female dog that died after exposure to 385 ppm 2-butoxyethanol for 8 days (Carpenter et al. 1956). The effect was not observed in the male dog that died after exposure to the same concentration for 28 days. This study is limited in that only one female dog was exposed to concentrations of 385 or 617 ppm, and only one male was used in the 385 ppm exposure concentration group.

Renal effects have also been observed in animals exposed to 2-butoxyethanol in intermediate-duration studies. Statistically significant increases in kidney weight were observed in Sherman rats exposed to ≥ 107 ppm and in guinea pigs exposed to ≥203 ppm for 7 hours per day, 5 days per week for 30 days (Carpenter et al. 1956). Cloudy swelling of the convoluted and loop tubules was observed upon necropsy of the guinea pigs that died at 376 and 494 ppm. No effect on kidney weight and no gross renal lesions were found in mice exposed to ≤400 ppm for 7 hours per day for 30-90 days. No blood or urinalysis parameters were affected, and no gross or histologically observed lesions in the kidneys or urinary bladder were found in Fischer 344 rats exposed to ≤77 ppm for 7 hours per day, 5 days per week for 13 weeks (Dodd et al. 1983).

Gross or histopathological changes in the kidneys were not observed in two dogs exposed to 415 ppm 2-butoxyethano17 hours per day, 5 days per week for 12 weeks (Werner et al. 1943b).

In a study of 2-butoxyethanol acetate, Wistar rats and New Zealand white rabbits were exposed to a saturated vapor-air mixture of approximately 400 ppm 2-butoxyethanol acetate for 4 hours, and then observed for 14 days after exposure (Truhaut et al. 1979). No blood was found in the urine of rats, although rabbits exhibited transient and slight hematuria. When the animals were sacrificed at the end of 2 weeks, no gross pathological lesions were observed in the renaI system of either rats or rabbits. In the same study, renal effects were noted in both rats and rabbits after intermittent exposure (5 days per week, 4 hours per day) to 400 ppm 2-butoxyethanol acetate for 1 month (Truhaut et al. 1979). Hematuria and tubular nephrosis, ranging from cellular cloudy swelling to hemorrhagic necrosis, were noted in female rats sacrificed at the end of the exposure period, but not in those sacrificed 1 week after exposure ceased. Rabbits exposed for 1 month exhibited hypertrophic kidneys filled with blood and blood-filled bladders (hematuria) at death during week 4. These rabbits, and those surviving to the end of the exposure period, exhibited necrotizing tubular nephrosis, atrophic tubular dilation, and luminar granular deposits. Effects observed in rabbits exposed to 100 ppm 2-butoxyethanol acetate for 10 months, 5 days per week, 4 hours per day included tubular nephritis with tubular enlargement, cortical atrophy, inflammatory fibrosis, and tubular Henle's loop dilation. Effects in rats exposed to the same concentration for 10 months were described as similar to those observed in rabbits, but the lesions were more discrete and inconstant (i.e., not always present) and were characterized (only in males) by tubular enlargement and atrophy, nephritis, inflammatory fibrosis, dilatation of Henle's loop and the distal convoluted tubules, and hyaline casts (Truhaut et al. 1979).

Endocrine Effects. Severely hemorrhaged adrenal glands were observed at necropsy in a female dog that died after exposure to 385 ppm 2-butoxyethanol for 8 days (Carpenter et al. 1956). No gross or histologically observed lesions were found in the adrenal, parathyroid, thyroid, or pituitary in Fischer 344 rats exposed to ≤77 ppm, 6 hours per day, 5 days per week for 13 weeks (Dodd et al. 1983). Gross lesions in the pituitary, thyroid gland, pancreas, parathyroid gland, and adrenal glands were not observed in guinea pigs exposed to 2-butoxyethanol for 1 hour followed by a 14-day observation period at 661 ppm for females and 633 ppm for males (Nachreiner 1994).

In a study of 2-butoxyethanol acetate, Wistar rats and New Zealand white rabbits were exposed to 400 ppm 2-butoxyethanol acetate for 4 hours, followed by a 14-day observation period (Truhaut et al. 1979). When the animals were sacrificed at the end of 2 weeks, no gross pathological lesions were observed in the adrenal

gland or pancreas of either rats or rabbits. Similarly, no gross or histologically observed lesions were noted in the pancreas or adrenal glands of rats and rabbits exposed for 4 hours per day, 5 days per week to 400 ppm of 2-butoxyethanol acetate for 1 month or to 100 ppm for 10 months (Truhaut et al. 1979).

Dermal Effects. Nonpregnant female rats exposed to 2-butoxyethanol at concentrations of 250-500 ppm for up to 7 hours exhibited necrosis of the tail tip 1 week after exposure (Nelson et al. 1984). In addition, pregnant Fischer 344 rats exposed to 200 ppm 2-butoxyethanol for 6 hours per day during gestational days 6-15 exhibited necrosis of the tail tip (Tyl et al. 1984). The cause of necrosis of the tail tip was not clearly stated, but possibly it is associated with the effects of 2-butoxyethanol on the red blood cells, and subsequent vascular response. Stained fur was noted in both pregnant Fischer 344 rats dosed during gestational days 6-15 and pregnant New Zealand white rabbits exposed on gestational days 6-18 to 200 ppm 2-butoxyethanol for 6 hours per day (Tyl et al. 1984). The exact cause of the stained fur was not noted.

Ocular Effects. Male and female volunteers exposed to 98, 113, or 195 ppm 2-butoxyethanol for 4-8 hours experienced ocular irritation during exposure to 113 and 195 ppm (Carpenter et al. 1956).

Pregnant Fischer 344 rats exposed to 25-200 ppm during gestational days 6-15 exhibited periocular wetness at all concentrations, whereas New Zealand white rabbits exposed to 100 and 200 ppm, but not 25 ppm, 2-butoxyethanol during gestation exhibited the same effect (Tyl et al. 1984). The ocular irritation in the human subjects and the periocular wetness in the animals were probably due to direct contact of the eyes with the 2-butoxyethanol vapor; these effects are also discussed in Section 2.2.3.2 for Ocular Effects.

Ocular infections were observed in a female dog that died after 8 days and in a male dog that died after 28 days following exposure to 385 ppm 2-butoxyethanol (Carpenter et al. 1956). It is not clear whether the ocular infections were due to 2-butoxyethanol exposure since no controls were included. In another experiment by Carpenter et al. (1956), a male and a female dog exposed to 200 ppm for 31 days showed no effects on the cornea after staining with fluorescein. A slight increase in ocular secretions was observed in 2 dogs exposed to 415 ppm 2-butoxyethano17 hours per day, 5 days per week for 12 weeks (Werner et al. 1943b). In male albino rabbits exposed to 400-411 ppm for 7 hours per day for l-2 days, a reddish ocular discharge accompanied by yellow discoloration of the sclera was observed upon necropsy (Dow 1986). These effects may have been secondary to the hemolytic property of 2-butoxyethanol.

Ophthalmological examination of the eyes revealed no ocular effects in Fischer 344 rats exposed 5 days per week, 6 hours per day to ≤245 ppm 2-butoxyethanol for 9 days or to ≤77 ppm for 13 weeks (Dodd et al. 1983).

Body Weight Effects. Fischer 344 rats exposed for 4 hours to concentrations of 0, 202, 523, and 867 ppm 2-butoxyethanol exhibited significant weight loss on the first day after exposure at concentrations ≥ 523 ppm 2-butoxyethanol, but weight gain resumed 4 days postexposure; body weight returned to preexposure values between days 7-14, and continued to increase for the duration of the 14-day observation period (Dodd et al. 1983). Similarly, decreased weight gain was observed when male rats were exposed to 245 ppm or female rats were exposed to 86 ppm for 9 days (Dodd et al. 1983), although no effect on body weight was observed in male Sprague-Dawley rats exposed to 57-58 ppm for 7 hours per day for 4 days (Dow 1972) or to 100 ppm 2-butoxyethanol for 12 days, 24 hours per day (Johanson 1994). However, after timed-pregnant Fischer 344 rats were exposed to 2-butoxyethanol vapors by inhalation on gestational days 6-l 5 and New Zealand white rabbits were exposed on gestational days 6-l 8 to concentrations of 0, 25, 50, 100, or 200 ppm for 6 hours per day, reduced final body weight or reduced weight gain were observed at 100 and 200 ppm in rats and at 200 ppm in rabbits (Tyl et al. 1984). It should be noted that the pregnant rats, but not the pregnant rabbits, had reduced food consumption at 100 ppm and reduced water consumption at 200 ppm, as discussed in Other Systemic Effects below. An unspecified weight loss was found in a female dog that died after exposure for 8 days and in a male dog that died after exposure for 28 days to 385 ppm; anorexia was observed in the female dog (Carpenter et al. 1956). However, no effects on body weight were found in two male dogs exposed to 400-411 ppm, 7 hours per day for l-5 days (Dow 1986).

No effect on body weight was observed in male and female guinea pigs exposed to 633 ppm or 691 ppm 2-butoxyethanol, respectively, for 1 hour (Nachreiner 1944). No effect on body weight was observed in male and female Sherman rats exposed to ≤203 ppm 2-butoxyethanol or in guinea pigs exposed to ≤495 ppm for 7 hours per day, 5 days per week, for 30 days (Carpenter et al. 1956). However, in mice exposed for 30, 60, or 90 days, a significant but unspecified decrease in body weight was noted after the 60-day exposure at 400 ppm. Two dogs exposed to 415 ppm 2-butoxyethano17 hours per day, 5 days per week, for 12 weeks gradually lost 6-9% of their body weight (Werner et al. 1943b). No control body weight data were provided, although the study authors indicated that the other dogs in the study gained weight or remained constant throughout the study period. In an intermediate-duration study in male and female Fischer 344 rats, female rats exposed to 77 ppm 2-butoxyethanol for 13 weeks, 5 days per week, 6 hours per day showed a transient

unspecified decrease in body weight gain during exposure weeks 24 that was not evident at necropsy (Dodd et al. 1983).

No effects on body weight were noted in Wistar rats or New Zealand white rabbits exposed intermittently (5 days per week, 4 hours per day) to 400 ppm 2-butoxyethanol acetate for 1 month or to 100 ppm 2-butoxyethanol acetate for 10 months (Truhaut et al. 1979).

Other Systemic Effects. No effect on food or water intake was observed for male Sprague-Dawley rats exposed to 20 or 100 ppm 2-butoxyethanol for 24 hours per day for 12 days (Johanson 1994). Timed-pregnant Fischer 344 rats exposed to 2-butoxyethanol vapors by inhalation on gestational days 6-15 at concentrations of 0, 25, 50, 100, or 200 ppm for 6 hours per day exhibited reduced food consumption at 100 ppm and reduced water consumption at 200 ppm (Tyl et al. 1984). No effect on food or water consumption was observed for rabbits exposed by the same regimen on gestational days 6-l 8 (Tyl et al. 1984). Several investigators note red staining or red fluid on cage papers or in the cages but do not further identify this observation (Tyl et al. 1984). Based on the observations of hematuria and hemoglobinuria by other investigators, it seems likely that observations of red stains or fluid represent hematuria or hemoglobinuria. No adverse changes were noted with respect to general appearance and appetite after exposure of guinea pigs and dogs to concentrations ≤58 ppm 2-butoxyethanol for 4 days, 7 hours per day (Dow 1972) or after exposure of guinea pigs to 400-411 ppm for l-5 days, 7 hours per day (Dow 1986).

2.2.1.3 Immunological and Lymphoreticular Effects

No studies were located regarding immunological and lymphoreticular effects in humans after inhalation exposure to 2-butoxyethanol or 2-butoxyethanol acetate. No studies were located regarding immunological effects in animals after inhalation exposure to 2-butoxyethanol or 2-butoxyethanol acetate. However, studies in animals that examined effects on lymphoreticular organs were located.

In a study in which Swiss mice were exposed to 390-1,210 ppm 2-butoxyethanol for 7 hours,focal necrosis and lymphoid hyperplasia in the spleen were observed (Werner et al. 1943a). The concentrations at which these effects occurred were not stated. Timed-pregnant Fischer 344 rats exposed to 0, 25, 50, 100, or 200 ppm 2-butoxyethanol by inhalation for 6 hours per day on gestational days 6-l 5 exhibited increased spleen weights at 200 ppm; pregnant New Zealand white rabbits exposed to the same concentrations for a comparable period during gestation (gestational days 6-18) showed no effect on spleen weight (Tyl et al.

1984). No histological examination of the spleen was performed. Gross examination did not reveal any effects in the spleen, thymic region, or lymph nodes in guinea pigs exposed to 2-butoxyethanol for 1 hour followed by a 14-day observation period at 633 ppm for males and 691 ppm for females (Nachreiner 1994). Histological examination of the spleen, mediastinal lymph nodes, and thymus revealed no pathological lesions in Fischer 344 rats exposed intermittently to <77 ppm for 13 weeks (Dodd et al. 1983). Histological examination did not reveal any effects in the spleen of 2 dogs exposed to 415 ppm 2-butoxyethano17 hours per day, 5 days per week for 12 weeks (Werner et al. 1943b).

In a study of 2-butoxyethanol acetate, Wistar rats and New Zealand white rabbits were exposed to 400 ppm 2-butoxyethanol acetate for 4 hours and then observed for 14 days after exposure (Truhaut et al. 1979). When the animals were sacrificed at the end of 2 weeks, no gross pathological lesions were observed in the spleen of either rats or rabbits. Similarly, no gross or histologically observed lesions were found in the spleens after rats and rabbits were exposed intermittently to 400 ppm of 2-butoxyethanol acetate for 1 month or to 100 ppm for 10 months (Truhaut et al. 1979).

Spleen weight effects may be due to hemolysis, which is discussed under Hematological Effects. Refer to Section 2.4.2 Mechanisms of Toxicity for information on how this effect may occur.

The highest NOAEL values and all LOAEL values from each reliable study for lymphoreticular effects in each species and duration category are recorded in Tables 2-1 and 2-2 and plotted in Figures 2-l and 2-2.

2.2.1.4 Neurological Effects

No studies were located regarding neurological effects in humans after inhalation exposure to 2-butoxyethanol acetate. Male and female volunteers exposed to 98, 113, or 195 ppm 2-butoxyethanol for 4-8 hours experienced headache at 98 ppm and disturbed taste sensation at 113 and 195 ppm (Carpenter et al. 1956).

Male and female Fischer 344 rats experienced loss of coordination after exposure to 523 and 867 ppm 2-butoxyethanol, but not to 202 ppm, for 4 hours (Dodd et al. 1983). Extreme physical weakness was observed in a female dog exposed to 617 ppm for 13.5 hours over 2 days, and weakness and apathy were observed in other dogs that died after exposure to 385 ppm for 8 days (one female) and 28 days (one male) (Carpenter et al. 1956). An additional study of two male beagles exposed to approximately 400 ppm 2-butoxyethanol for 7 hours per day for up to 5 days showed excess salivation; no adverse effects on

demeanor were observed at this or lower concentrations (Dow 1972, 1986). Male albino rabbits exhibited poor coordination of the extremities and loss of eqluilibrium after exposure to approximately 400 ppm 2-butoxyethanol for 7 hours per day for 1-2 days (Dow 1986). Histological examination of the brain and sciatic nerve revealed no pathological lesions in Fischer 344 rats exposed to ≤77 ppm intermittently for 13 weeks (Dodd et al. 1983).

Wistar rats and New Zealand white rabbits exposed to 400 ppm 2-butoxyethanol acetate for 4 hours and then observed for 14 days after exposure showed no clinical signs of neurological effects (Truhaut et al. 1979). When the animals were sacrificed at the end of 2 weeks, no gross pathological lesions were observed in the brain of either rats or rabbits. Similarly, no clinical neurological effects and no gross or histologically observed lesions in brain were noted when rats and rabbits were exposed intermittently to 400 ppm of 2-butoxyethanol acetate for 1 month or to 100 ppm for 10 months (Truhaut et al. 1979).

The highest NOAEL values and all LOAJZL values from each reliable study for neurological effects in each species and duration category are recorded in Tables 2-l and 2-2 and plotted in Figures 2-l and 2-2.

2.2.1.5 Reproductive Effects

No studies were located regarding reproductive effects in humans after inhalation exposure to 2-butoxyethanol or 2-butoxyethanol acetate.

Male Alpk/Ap (Wistar-derived) rats were exposed to 800 ppm 2-butoxyethanol for 3 hours (Doe 1984). The rats were then observed during exposure and throughout a subsequent 14-day observation period. On the 15th day, animals were killed and gross macroscopic examination was conducted. No effect on testicular weight was observed. Effects on testicular weight were not noted in Fischer 344 rats exposed to ≤867 ppm for 4 hours, to ≤245 ppm intermittently for 9 days, or to ≤77 ppm intermittently for 13 weeks (Dodd et al. 1983); histological examination of the epididymides and testes of male rats in a 13-week study revealed no pathological lesions. Reproductive organs of the female rats in the 13-week study by Dodd etal. (1983) were not examined histologically. Gross changes in male (testes, penis, seminal vesicle, epididymides, prostate) and female (ovaries, uterus, cervix, vagina, vulva) reproductive organs were not observed in guinea pigs exposed to 2-butoxyethanol for 1 hour at 633 ppm for males and 691 ppm for females, followed by a 14-day observation period (Nachreiner 1994).

Pregnant Fischer 344 rats exposed intermittently to 25-200 ppm 2-butoxyethanol during gestational days 6-l5 exhibited reduced maternal gravid uterine weight, a 50% decrease in viable implants and in live fetuses per litter, and an eight-fold increase in non-viable implants at 200 ppm, but not at the lower concentrations (Tyl et al. 1984). New Zealand white rabbits similarly exposed to the same concentrations on gestational days 6-18 also exhibited decreases in total implants and implant viability at 200 ppm (Tyl et al. 1984). The exposure concentrations that resulted in reproductive effects following exposure of rats and rabbits to 2-butoxyethanol during gestation also resulted in maternal toxicity (see Section 2.2.1.2).

Wistar rats and New Zealand white rabbits exposed to 400 ppm 2-butoxyethanol acetate for 4 hours, and then observed for 14 days after exposure, showed no signs of vaginal bleeding or testicular shrinkage (Truhaut et al. 1979). When the animals were sacrificed at the end of 2 weeks, no gross pathological lesions were observed in the testes or ovaries of either rats or rabbits exposed for 4 hours to 400 ppm 2-butoxyethanol acetate. Similarly, no gross or histologically observed lesions in the testes or ovaries were seen when rats and rabbits were exposed intermittently to 400 ppm of 2-butoxyethanol acetate for 1 month or to 100 ppm for 10 months (Truhaut et al. 1979).

The highest NOAEL values and all LOAEL values from each reliable study for reproductive effects in each species and duration category are recorded in Tables 2-1 and 2-2 and plotted in Figures 2-l and 2-2.

2.2.1.6 Developmental Effects

No studies were located regarding developmental effects in humans after inhalation exposure to 2-butoxyethanol or 2-butoxyethanol acetate. No studies were located regarding developmental effects in animals after inhalation exposure to 2-butoxyethanol acetate.

2-Butoxyethanol was vaporized and administered at concentrations of 0, 150, or 200 ppm to pregnant rats for 7 hours per day on gestational days 7-15 (Nelson et al. 1984). On day 20, the females were weighed and euthanized. The entire uterus was removed and the numbers of resorption sites and live fetuses were determined. Fetuses were serially removed, weighed, aud examined for visceral malformations and skeletal defects. No developmental effects were noted. In a similar study, timed-pregnant Fischer 344 rats were exposed to 2-butoxyethanol vapors by inhalation on gestational days 6-15 at concentrations of 0, 25, 50, 100, or 200 ppm for 6 hours per day (Tyl et al. 1984). Fetuses were weighed and sexed, and evaluated for viability, body weight, and morphological development, including external, visceral, and skeletal

malformations. Fetotoxicity, observed as retarded skeletal ossification of vertebral arches or centra, sternebrae, or phalanges, was observed at 100 and 200 ppm, concentrations that resulted in maternal toxicity. For pregnant New Zealand white rabbits similarly exposed to the same concentrations on gestational days 6-18, fetal skeletal ossification of sternebrae and rudimentary rib was also delayed at 200 ppm but not at ≤ 100 ppm (Tyl et al. 1984). Gravid uterine weight was reduced and maternal toxicity was observed at 200 ppm in rabbits (Tyl et al. 1984).

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

2.2.1.7 Genotoxic Effects

No increases in micronuclei or sister chromatid exchanges were observed in varnish production workers exposed to both 2-ethoxyethanol and 2-butoxyethanol (Sohnllein et al. 1993). The concentration of 2-ethoxyethanol in the air of the workroom ranged from <0. 1-15.2 ppm, while the range for 2-butoxyethanol was <0.1-1.4 ppm. Postshift biological monitoring for the acetic acids in urine indicated that urinary levels of 2-ethoxyacetic acid (53.8 mg/L) were higher than urinary levels of 2-butoxyacetic acid (16.4 mg/L). No studies were located regarding genotoxic effects in animals after inhalation exposure to 2-butoxyethanol or 2-butoxyethanol acetate. *In vitro* genotoxicity studies are discussed in Section 2.5.

2.2.1.8 Cancer

No studies were located regarding carcinogenic effects in humans or animals after inhalation exposure to 2-butoxyethanol or 2-butoxyethanol acetate.

2.2.2 Oral Exposure

Oral exposure to 2-butoxyethanol or 2-butoxyethanol acetate may occur through accidental ingestion of contaminated food, water, or commercial products, or from intentional ingestion of commercial products containing these chemicals. Studies describing human oral exposure, in addition to experimental animal studies, are discussed in this section (Section 2.2.2) and have been summarized in several publications (Browning and Curry 1994; EPA 1984; NIOSH 11990; Tyler 1984).

2.2.2.1 Death

No studies were located regarding death in humans after oral exposure to 2-butoxyethanol acetate, or death in humans or animals after oral exposure to the major metabolite, 2-butoxyacetic acid.

An 87-year-old woman died after ingesting an unknown amount of disinfectant cleaner containing 6.5% 2-butoxyethanol (Litovitz et al. 199 1). The other ingredients in the cleaner were not stated. Upon admission to the hospital the blood ethylene glycol level was 100 mg/dL. Complications included prolonged metabolic acidosis, hypotension, ventricular arrhythmias, hepatic and renal failure, and disseminated intravascular coagulation. Although the blood ethylene glycol level decreased to 10 mg/dL, she had a cardiac arrest and died 3 days after admission.

Acute oral LD_{50} (lethal dose, 50% kill) values of 2-butoxyethanol for rats ranged from 530 to 3,000 mg/kg (Carpenter et al. 1956; Eastman Kodak 1988; Nelson et al. 1984; Olin 1976; Smyth et al. 1941; Union Carbide 1980b). In rats of an unspecified strain given 2-butoxyethanol by gavage in water, death occurred in three of five rats at 500 mg/kg and two of two rats at 1,000 mg/kg (Dow 1959). No deaths occurred at 252 mg/kg. In female Fischer 344 rats, deaths occurred in two of three rats at 2,000 mg/kg, while no deaths occurred at ≤ 1,000 mg/kg (Dow 1981). In contrast, when male Fischer 344 rats were given 50, 100, 200, or 400 mg/kg/day for 2 days, one of six rats died and another was moribund at 200 mg/kg/day; six of six rats died at 400 mg/kg/day; no rats died at ≤ 100 mg/kg/day (Smialowicz et al. 1992). When female Sprague-Dawley female rats were treated with a single gavage dose of 1,500 mg/kg 2-butoxyethanol in water, 75% died (Sivarao and Mehendale 1995). When rats were treated with a non-lethal dose (500 mg/kg) followed 7 days later by a dose of 1,500 mg/kg, only 13% died. No male Fischer 344 rats given 126 mg/kg 2-butoxyethanol in water by gavage died (Corley et al. 1994). Pregnant Fischer 344 rats dosed on gestational days 9-l1 or 1l-l3 with doses of 150, 300, or 600 mg/kg/day in a preliminary dose range-finding study exhibited 11% (1 of 9) and 17% (1 of 6) mortality within the first 24 hours at doses of 300 and 150 mg/kg/day, respectively (NTP 1989). Two of 6 of the pregnant rats treated on gestational days 9-l1 and 1 of 7 pregnant rats treated on gestational days 1l-l3 died at 600 mg/kg/day dose level. However, no mortaliiy was observed in the definitive study with doses ≤ 200 mg/kg/day on gestational days 9-l1 and ≤ 300 mg/kg/day on gestational days 11-13 (NTP 1989). Similarly, no deaths occurred in male or female Fischer 344/N rats dosed with ≤346 mg/kg/day or ≤265 mg/kg/day, respectively, in drinking water for 2 weeks (NTP 1993). However, in male COBS CD (SD)BR rats given doses of 222, 443, or 885 mg/kg/day for 5 days per week for

23 days, death occurred in 1 of 10 on day 13 at 443 mg/kg/day, and in 1 of 10 on day 13, and 1 of 9 on day 23 at 885 mg/kg/day (Eastman Kodak 1983; Krasavage 1986).

Acute oral LD_{50} values for mice were reported to be 1,230 mg/kg (Carpenter et al. 1956) and 1,519 mg/kg (Eastman Kodak 1988). When time-mated CD-l mice were treated by gavage on gestational days 6-l 3 with 1,180 mg/kg/day 2-butoxyethanol, death occurred in 4 of 35 pregnant mice and 6 of 15 nonpregnant mice (Hardin et al. 1987; Schuler et al. 1984). In a similar study in mated CD-l mice given 0, 350, 650, 1,000, 1,500, or 2,000 mg/kg 2-butoxyethanol by gavage on gestational days 8-14, three of six died at 1,500 mg/kg/day and six of six died at 2,000 mg/kg/day (Wier et al. 1987). In Swiss CD-l mice given 2-butoxyethanol in drinking water at doses ≤ 12,750 mg/kg/day for 2 weeks, deaths were observed in two of eight males and five of eight females at 12,750 mg/kg/day (Heindel et al. 1990). None of the mice died at doses ≤6,375 mg/kg/day. No deaths were observed in $B6C3F_1$ mice dosed with ≤627 mg/kg/day for males or ≤ 1,364 mg/kg/day for females in drinking water for 2 weeks (NTP 1993).

Acute oral LD_{50} values for other species were reported to be 1,200 (Carpenter et al. 1956; Smyth et al. 1941) and 1,414 mg/kg (Shepard 1994b) for guinea pigs and 320-370 mg/kg for rabbits (Carpenter et al. 1956).

In intermediate-duration studies of 2-butoxyethanol, no deaths occurred in male or female Sherman rats dosed with ≤ 1540 mg/kg/day in the diet for 90 days (Carpenter et al. 1956), in Fischer 344/N rats dosed with ≤452 mg/kg/day for males and ≤470 mg/kg/day for females in drinking water for 13 weeks (NTP 1993), in male Fischer 344/N rats dosed with ≤443 mg/kg/day in drinking water for 60 days (NTP 1993), or in an unspecified strain of rats dosed with ≤976 mg/kg/day in the diet for 91-93 days (Weil and Carpenter 1963). Similarly, no deaths occurred in $B6C3F_1$ mice dosed with ≤694 mg/kg/day (males) or ≤ 1,306 mg/kg/day (females) for 13 weeks (NTP 1993). However, in Swiss CD-l mice given 0, 700, 1,300, or 2,000 mg/kg/day in drinking water for 21 weeks, deaths occurred in 1 of 20 females at 700 mg/kg/day, 6 of 20 females at 1,300 mg/kg/day, and 13 of 20 females at 2,000 mg/kg/day (Heindel et al. 1990). None of the male mice died. In addition, deaths occurred in five of five male JCL-ICR mice dosed with 2,000 mg/kg/day by gavage 5 days per week for 5 weeks (Nagano et al. 1979, 1984). No deaths occurred in the mice given lower doses (≤ 1,000 mg/kg/day).

Mortality data for 2-butoxyethanol acetate are limited to LD_{50} values. In Wistar rats given an oral dose of 2-butoxyethanol acetate in olive oil by gastric intubation, the LD_{50} values were determined to be 3,000 mg/kg for males and 2,400 mg/kg for females (Truhaut et al. 1979).

The LD_{50} values and all LOAEL values from each reliable study for death in each species and duration category are recorded in Tables 2-3 and 2-4 and plotted in Figures 2-3 and 2-4.

2.2.2.2 Systemic Effects

No studies were located regarding gastrointestinal, musculoskeletal, endocrine, dermal, or body weight effects in humans, or metabolic effects in animals following oral exposure to 2-butoxyethanol. No reliable studies were located describing systemic effects in humans or animals following oral exposure to 2-butoxyethanol acetate. One study, Truhaut et al. (1979), gives some data for oral exposure regarding respiratory, cardiovascular, hematological, hepatic, renal, and endocrine effects, but does not specify the doses given. The data from that study have been included in this section for reference. Only one study (Foster et al. 1987) described systemic effects of 2-butoxyacetic acid after oral exposure in animals, consisting of hepatic, renal, and body weight effects. Available data pertaining to systemic effects of all three compounds are presented below.

The highest NOAEL values and all LOAEL values from each reliable study for systemic effects in each species and duration category are recorded in Tables 2-3 and 2-5 and plotted in Figures 2-3 and 2-5.

Respiratory Effects. A male patient who was an abuser of alcohol and had a history of trichloroethylene ingestion was admitted to the hospital after ingestion of 500 mL of a household cleaning fluid, Glassex (Bauer et al. 1992). The concentration of 2-butoxyethanol as determined by gas-chromatography was 9.1% (45.5 g), equivalent to a dose of 650 mg/kg. Diffuse pulmonary edema was noted. In another case report, a woman was admitted to the hospital after ingesting 250-500 mL window cleaner containing 12% 2-butoxyethanol (Rambourg-Schepens et al. 1988). The estimated dose was 467-933 mg/kg. The woman was ventilating poorly and required a respirator for 5 days after admittance. Similarly, a woman who ingested 25-30 g (391-469 mg/kg) 2-butoxyethanol contained in a window cleaning agent exhibited obstructive respiration and was placed on a respirator upon admission to the hospital (Gijsenbergh et al. 1989). However, two children who accidentally ingested a window cleaning agent containing 3 or 24 mL 2-butoxyethanol(270 or 1,862 mg/kg 2-butoxyethanol) showed no respiratory effects (Dean and Krenzelok 1992). Gastric emptying by administration of syrup of ipecac or by gastric lavage within minutes or hours of ingestion (time not specified) may have prevented any adverse effects.

Table 2-3. Levels of Significant Exposure to 2-Butoxyethanol - Oral

Key to[a] figure	Species/ (Strain)	Exposure/ Duration/ Frequency (Specific Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference
	ACUTE EXPOSURE						
	Death						
1	Rat (Wistar, Sherman, Carworth-Wistar)	once (G)				530-3000 (LD_{50})	Carpenter et al. 1956
2	Rat (NS)	NS (GW)				500 (death in 3/5 animals)	Dow 1959
3	Rat (Fischer- 344)	once (G)				2000 F (2/3 died)	Dow 1981
4	Rat (Crl:COBS CD (SD)BR)	13 d 5 d/wk (G)				443 M (death in 1/10)	Eastman Kodak 1983; Krasavage 1986
5	Rat (CD)	NS (G)				1746 M (LD_{50})	Eastman Kodak 1988
6	Rat (NS)	NS				1480 (LD_{50})	Nelson et al. 1984
7	Rat (Fischer- 344)	Gd 11-13 (GW)				150 F (1/9 died 24 hrs after exposure)	NTP 1989

Table 2-3. Levels of Significant Exposure to 2-Butoxyethanol - Oral (continued)

Key to[a] figure	Species/ (Strain)	Exposure/ Duration/ Frequency (Specific Route)	System	NOAEL (mg/kg/day)	LOAEL			Reference
					Less Serious (mg/kg/day)	Serious (mg/kg/day)		
8	Rat (Wistar)	once (G)				1590 M	(LD_{50})	Olin 1976
9	Rat (Fischer- 344)	2 d (GW)				200 M	(death in 1/6)	Smialowicz et al. 1992
10	Rat (Wistar)	once (GW)				1480 M	(LD_{50})	Smyth et al. 1941
11	Rat (Wistar)	once (G)				2417	(LD_{50})	Union Carbide 1980b
12	Mouse (NS)	once				1230 M	(LD_{50})	Carpenter et al. 1956
13	Mouse (Charles River, COBS, CD-1)	NS (G)				1519 M	(LD_{50})	Eastman Kodak 1988
14	Mouse (CD-1)	Gd 6-13 (GW)				1180 F	(death in 4/35 pregnant mice and 6/15 nonpregnant mice)	Hardin et al. 1987; Schuler et al. 1984
15	Mouse (Swiss CD-1)	2 wk (W)				12750	(death in 2/8 males, 5/8 females)	Heindel et al. 1990

Table 2-3. Levels of Significant Exposure to 2-Butoxyethanol - Oral (continued)

Key to[a] figure	Species/ (Strain)	Exposure/ Duration/ Frequency (Specific Route)	System	NOAEL (mg/kg/day)	LOAEL			Reference
					Less Serious (mg/kg/day)	Serious (mg/kg/day)		
16	Mouse (CD-1)	Gd 8-14 1x/d (G)				1500 F	(death in 3/6)	Wier et al. 1987
17	Gn Pig (NS)	once (G)				1200	(LD_{50})	Carpenter et al. 1956
18	Gn Pig (Hartley)	once (GW)				1414	(LD_{50})	Shepard 1994b
19	Rabbit (NS)	once				320-370 M	(LD_{50})	Carpenter et al. 1956
	Systemic							
20	Human	once (IN)	Resp			650 M	(diffuse pulmonary edema)	Bauer et al. 1992
			Cardio			650 M	(hypotension, tachycardia, sinusal rhythm)	
			Hemato			650 M	(low prothrombin time, nonhemolytic anemia, thrombopenia)	
			Hepatic			650 M	(abnormal liver function)	
			Renal		650M	(slight albuminuria)		
			Metab			650 M	(metabolic acidosis and hypoxemia with lactic acidosis)	

Table 2-3. Levels of Significant Exposure to 2-Butoxyethanol - Oral (continued)

Key to[a] figure	Species/ (Strain)	Exposure/ Duration/ Frequency (Specific Route)	System	NOAEL (mg/kg/day)	LOAEL		Reference
					Less Serious (mg/kg/day)	Serious (mg/kg/day)	
21	Human	once (IN)	Resp			391 F (obstructive respiration)	Gijsenbergh et al. 1989
			Cardio			391 F (low blood pressure)	
			Hemato			391 F (decreased hemoglobin from 11.9 to 8.9 g/dL)	
			Renal			391 F (hematuria)	
			Ocular			391 F (isocoric light reactive mydriasis)	
			Metab			391 F (marked metabolic acidosis)	
22	Human	2x	Hepatic		1006M (increased serum ALT, AST, bilirubin; only after first exposure)		Gualtieri et al. 1995
			Metab			1006 M (significant acid-base disturbance)	
23	Human	once (IN)	Resp			467 F (poor ventilation)	Rambourg-Schepens et al. 1988
			Cardio	467 F			
			Hemato			467 F (hemoglobinuria, progressive erythropenia)	
			Hepatic	467 F			
			Renal			467 F (increased serum creatinine, oxaluria)	
			Metab			467 F (metabolic acidosis, hypokalaemia)	

Table 2-3. Levels of Significant Exposure to 2-Butoxyethanol - Oral (continued)

Key to[a] figure	Species/ (Strain)	Exposure/ Duration/ Frequency (Specific Route)	System	NOAEL (mg/kg/day)	LOAEL				Reference
					Less Serious (mg/kg/day)		Serious (mg/kg/day)		
24	Rat (Wistar, Sherman, Carworth-Wistar)	once (G)	Resp				530	(congested or hemorrhagic lungs)	Carpenter et al. 1956
			Hemato				3000 M 1500 F	(hemoglobinuria)	
			Hepatic				530	(mottled livers)	
			Renal				530	(severely congested kidneys)	
			Dermal		530	(rough coat)			
25	Rat (Fischer- 344)	once (GW)	Hemato	8.6 M			126 M	(hemolysis, hemoglobinuria)	Corley et al. 1994
26	Rat (NS)	NS (GW)	Renal	1000 F	2000 F	(rapid, shallow breathing)			Dow 1959
			Resp	252			500	(hematuria)	
27	Rat (Fischer- 344)	once (G)	Dermal	500 F	1000 F	(rough hair coats; necrosis of tail)			Dow 1981
			Ocular	1000 F	2000 F	(palpebral closure)			
			Bd Wt	2000 F					
			Other		130 F	(staining in perineal region)			
28	Rat (Fischer- 344)	once (GW)	Hemato				250 M	(hemolysis)	Ghanayem and Sullivan 1993

Table 2-3. Levels of Significant Exposure to 2-Butoxyethanol - Oral (continued)

Key to[a] figure	Species/ (Strain)	Exposure/ Duration/ Frequency (Specific Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference
29	Rat (Fischer- 344)	once (GW)	Hemato		32 [b] M (hemoglobinuria)		Ghanayem et al. 1987a
			Hepatic	125 M		250 M (focal coagulative necrosis of hepatocytes, 1/6)	
			Renal			125 M (hemoglobin casts in proximal tubules)	
30	Rat (Fischer- 344)	once (GW)	Hemato			500 M (increase in free hemoglobin in the plasma, hemoglobinuria, hemolysis)	Ghanayem et al. 1987b
			Hepatic			500 M (coagulative necrosis and hemosiderin deposition in hepatocytes and Kupffer cells)	
			Renal			500 M (intracytoplasmic hemoglobin and hemoglobin casts in the proximal tubules)	
31	Rat (Fischer- 344)	once (GW)	Hemato			125 M (increase in free hemoglobin in the plasma, hemoglobinuria, hemolysis)	Ghanayem et al. 1987b
32	Rat (Fischer- 344)	once (GW)	Hemato			125 M (increased HCT, PCV, and MCV followed by decline with hemolysis)	Ghanayem et al. 1990b

Table 2-3. Levels of Significant Exposure to 2-Butoxyethanol - Oral (continued)

Key to[a] figure	Species/ (Strain)	Exposure/ Duration/ Frequency (Specific Route)	System	NOAEL (mg/kg/day)	LOAEL		Reference
					Less Serious (mg/kg/day)	Serious (mg/kg/day)	
33	Rat (Fischer- 344)	1-12 d 1x/d (GW)	Hemato			125 M (hemolytic anemia)	Ghanayem et al. 1992
			Hepatic		125M (time-dependent changes in liver weight: declined 10% on days 3 and 6, increased 5% on day 12)		
34	Rat (Fischer- 344)	4 d (GW)	Hemato			500 M (reduction of 23% in RBC, 11% in HGB, increase of 24% in MCV, 600% in reticulocyte counts and 16% in MCH; marrow hyperplasia)	Grant et al. 1985
			Hepatic		500M (15.8% increase in relative liver weights after day 1 recovery)		
			Renal	500 M	1000M (12.4% increase in relative kidney weight after day 1 recovery)		
			Bd Wt	500 M		1000 M (13.4% reduction in body weight gain after day 1 recovery and 26% reduction in body weight gain at day 4 recovery)	

Table 2-3. Levels of Significant Exposure to 2-Butoxyethanol - Oral (continued)

Key to[a] figure	Species/ (Strain)	Exposure/ Duration/ Frequency (Specific Route)	System	NOAEL (mg/kg/day)	LOAEL		Reference
					Less Serious (mg/kg/day)	Serious (mg/kg/day)	
35	Rat (Fischer- 344)	Gd 9-11 (GW)	Resp	300 F	600 F (dyspnea)		NTP 1989
			Hemato			150 F (increased reticulocytes, MCV, MCH and platelet count, decreased RBC, HGB, HCT, and MCHC)	
			Hepatic	150 F	300 F (27.1% decreased absolute liver weight [Gd 12])		
			Renal	300 F		600 F (urethral bleeding [hematuria])	
			Dermal	300 F	600 F (pale coloration, not further described)		
			Ocular	300 F	600 F (chromodacryorrhea)		
			Bd Wt			150 F (gestational weight gain decreased 34.7% [Gd 12])	
			Other		150 F (reduced food and water intake)		
				300 F	600 F (dehydration, cold to touch)		

Table 2-3. Levels of Significant Exposure to 2-Butoxyethanol - Oral (continued)

Key to[a] figure	Species/ (Strain)	Exposure/ Duration/ Frequency (Specific Route)	System	NOAEL (mg/kg/day)	LOAEL		Reference
					Less Serious (mg/kg/day)	Serious (mg/kg/day)	
36	Rat (Fischer- 344)	Gd 11-13 (GW)	Resp	300 F	600 F (dyspnea)		NTP 1989
			Hemato			150 F (increased reticulocytes, MCV, MCH and platelet count, decreased RBC, HGB, HCT and MCHC)	
			Hepatic	150 F	300 F (11.5% decreased absolute liver weight [Gd 14])		
			Renal	300 F		600 F (urethral bleeding [hematuria])	
			Dermal	300 F	600 F (pale coloration, not further described)		
			Ocular	300 F	600 F (chromodacryorrhea)		
			Bd Wt			150 F (gestational weight gain decreased 28.9% [Gd 14])	
			Other		150 F (reduced food and water intake)		
				300 F	600 F (dehydration, cold to touch)		

Table 2-3. Levels of Significant Exposure to 2-Butoxyethanol - Oral (continued)

Key to[a] figure	Species/ (Strain)	Exposure/ Duration/ Frequency (Specific Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference
37	Rat (Fischer- 344)	Gd 9-11 (GW)	Hemato	30 F		100 F (reduced RBC, HCT and HGB, increased reticulocytes, WBC, platelet count, MCV and MCH)	NTP 1989
			Hepatic	100 F	200 F (decreased absolute maternal liver weights: 11.1% [Gd 20], 15.5% [Gd 12])		
			Renal	200 F			
			Bd Wt	100 F		200 F (gestational weight gain decreased 35.3% [Gd 20])	
			Other	100 F	200 F (decreased food and water intake)		
38	Rat (Fischer- 344)	Gd 11-13 (GW)	Hemato	30 F		100 F (reduced RBC, HCT and HGB, increased reticulocytes, WBC, platelet count, MCV and MCH)	NTP 1989
			Hepatic	100 F	300 F (decreased 11.8% [Gd 14] absolute maternal liver weights)		
			Renal	300 F			
			Bd Wt	100 F		300 F (gestational weight gain decreased 20.4 % [Gd 20])	
			Other	100 F	300 F (decreased food and water consumption)		

Table 2-3. Levels of Significant Exposure to 2-Butoxyethanol - Oral (continued)

Key to[a] figure	Species/ (Strain)	Exposure/ Duration/ Frequency (Specific Route)	System	NOAEL (mg/kg/day)	LOAEL		Reference
					Less Serious (mg/kg/day)	Serious (mg/kg/day)	
39	Rat (F344/N)	2 wk (W)	Resp	346 M 265 F			NTP 1993
			Cardio	346 M 265 F			
			Hepatic	346 M 265 F			
			Renal	346 M 265 F			
			Bd Wt	346 M 203 F		265 F (11% decreased final body weight, 32% decrease in body weight gain)	
			Other	174 M 102 F	242 M (14.1% decreased water consumption) 152 F (16.3% decreased water consumption)		
40	Rat (Wistar)	once (G)	Gastro	1310 M	2560 M (very red small intestine)		Olin 1976
			Musc/skel	1310 M	2560 M (flaccid)		
			Hepatic	670 M	1310 M (very dark liver)		
			Renal	670 M	1310 M (dark kidneys in 3/10, enlarged in 4/10)	2560 M (blood in bladder [hematuria])	
			Dermal	670 M	1310 M (piloerection)		

Table 2-3. Levels of Significant Exposure to 2-Butoxyethanol - Oral (continued)

Key to[a] figure	Species/ (Strain)	Exposure/ Duration/ Frequency (Specific Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference
41	Rat (Wistar)	once (G)	Gastro	1127		2255 (distended stomach, liquid & gas-filled; blood in intestines)	Union Carbide 1980b
			Hepatic	1127	2255 (dark liver)		
			Renal	1127	2255 (red kidneys)		
			Endocr	1127	2255 (red adrenals)		
			Other		1127 (bloody saliva in 1 animal)		
42	Mouse (CD-1)	Gd 6-13 1x/d (GW)	Bd Wt			1180 F (80% decrease in body weight gain)	Hardin et al. 1987; Schuler et al. 1984
43	Mouse (Swiss CD-1)	2 wk (W)	Bd Wt	6375		12750 M (31% weight loss)	Heindel et al. 1990
			Other	637	1275 (unspecified decrease in fluid intake)		
44	Mouse (B6C3F1)	2 wk (W)	Resp	627 M 1364 F			NTP 1993
			Cardio	627 M 1364 F			
			Hepatic	627 M 1367 F			
			Renal	627 M 1364 F			
			Bd Wt	627 M 1364 F			
			Other	210 M	370 M (dehydration in 3/5) 150 F (26.5% decreased water consumption)		

Table 2-3. Levels of Significant Exposure to 2-Butoxyethanol - Oral (continued)

Key to[a] figure	Species/ (Strain)	Exposure/ Duration/ Frequency (Specific Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference
45	Mouse (CD-1)	Gd 8-14 (G)	Resp	1000 F	1500 F (abnormal breathing)		Wier et al. 1987
			Bd Wt	1500 F	2000 F (unspecified decrease in maternal weight gain)		
46	Mouse (CD-1)	Gd 8-14 1x/d (G)	Bd Wt	650 F	1000 F (unspecified decrease in maternal weight gain)		Wier et al. 1987
47	Gn Pig (Hartley)	once (GW)	Hemato	250 M			Ghanayem and Sullivan 1993
48	Gn Pig (Hartley)	once (GW)	Gastro	500	1000 (moderate to mild necrosis and hemorrhage of the gastric mucosa in 1/5 males and 1/5 females)		Shepard 1994b
			Bd Wt	1000			
	Immunological/Lymphoreticular						
49	Rat (Fischer- 344)	once (GW)		63 M	125M (significant increase (110-150%) in relative spleen weight)		Ghanayem et al. 1987a
50	Rat (Fischer- 344)	once (GW)			500M (>220% increase in relative spleen weight due to trapped RBCs)		Ghanayem et al. 1987b

Table 2-3. Levels of Significant Exposure to 2-Butoxyethanol - Oral (continued)

Key to[a] figure	Species/ (Strain)	Exposure/ Duration/ Frequency (Specific Route)	System	NOAEL (mg/kg/day)	LOAEL			Reference
					Less Serious (mg/kg/day)		Serious (mg/kg/day)	
51	Rat (Fischer- 344)	once (GW)			125M	(relative spleen weight 145-170% of control)		Ghanayem et al. 1987b
52	Rat (Fischer- 344)	1-12 d 1x/d (GW)			125M	(increase of about 62% in spleen weight after 6 days, decline of about 45% during days 6-12)		Ghanayem et al. 1992
53	Rat (Fischer- 344)	4 d (GW)			500M	(87% increase in relative spleen weight on day 1, extramedullary hematopoiesis)		Grant et al. 1985
54	Rat (Fischer- 344)	Gd 9-11 (GW)			150 F	(increased absolute spleen weight [13.5% Gd 20; 54.8% Gd 12])		NTP 1989
55	Rat (Fischer- 344)	Gd 11-13 (GW)			150 F	(increased absolute spleen weights [27.1% Gd 20; 67.7% Gd 14])		NTP 1989
56	Rat (Fischer- 344)	Gd 9-11 (GW)		30 F	100 F	(increased absolute spleen weight [11.8% Gd 20; 43.9% Gd 12])		NTP 1989
57	Rat (Fischer- 344)	Gd 11-13 (GW)		30 F	100 F	(increased absolute spleen weight [17.5% Gd 20; 44.4% Gd 14])		NTP 1989

Table 2-3. Levels of Significant Exposure to 2-Butoxyethanol - Oral (continued)

Key to[a] figure	Species/ (Strain)	Exposure/ Duration/ Frequency (Specific Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference
58	Mouse (B6C3F1)	2 wk (W)		210 M 1364 F	370M (decreased 38.3% absolute thymus weight, 38.9% relative thymus weight)		NTP 1993
	Neurological						
59	Human	once (IN)				391 F (coma)	Gijsenbergh et al. 1989
60	Human	once (IN)				467 F (coma)	Rambourg-Schepens et al. 1988
61	Rat (Wistar, Sherman, Carworth-Wistar)	once				530 (sluggishness, prostration, narcosis)	Carpenter et al. 1956
62	Rat (NS)	NS (GW)			252 (drowsiness)		Dow 1959
63	Rat (Fischer- 344)	once (G)		1000 F	2000 F (lethargy)		Dow 1981
64	Rat (Crl:COBS CD (SD)BR)	1-3 d (G)		222 M	443M (lethargy after 1st dose)		Eastman Kodak 1983; Krasavage 1986

Table 2-3. Levels of Significant Exposure to 2-Butoxyethanol - Oral (continued)

Key to[a] figure	Species/ (Strain)	Exposure/ Duration/ Frequency (Specific Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference
65	Rat (Fischer- 344)	Gd 9-11 (GW)		300 F	600 F (lethargy)		NTP 1989
66	Rat (Fischer- 344)	Gd 11-13 (GW)		300 F	600 F (lethargy)		NTP 1989
67	Rat (Wistar)	once (G)		670 M	1310 M (lethargy, piloerection)		Olin 1976
68	Rat (Sprague-Dawley)	once (GW)				500 F (ataxia, piloerection)	Sivarao and Mehendale 1995
69	Rat (Wistar)	once (G)		1127	2255 (sluggish, unsteady gait)		Union Carbide 1980b
70	Mouse (CD-1)	Gd 8-14 1x/d (G)		1000 F		1500 F (lethargy, failure to right)	Wier et al. 1987
71	Gn Pig (Hartley)	once (GW)			500 (slight weakness directly after dosing)	1000 (moderate to severe weakness and prostration directly after dosing)	Shepard 1994b

Table 2-3. Levels of Significant Exposure to 2-Butoxyethanol - Oral (continued)

Key to[a] figure	Species/ (Strain)	Exposure/ Duration/ Frequency (Specific Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference
	Reproductive						
72	Rat (Fischer- 344)	once (GW)		500 M			Ghanayem et al. 1987a
73	Rat (Fischer- 344)	4 d (GW)		1000 M			Grant et al. 1985
74	Rat (Fischer- 344)	Gd 11-13 (GW)		300 F		600 F (vaginal bleeding)	NTP 1989
75	Rat (Fischer- 344)	Gd 9-11 (GW)		100 F		200 F (increased resorptions, implantation loss, vaginal bleeding)	NTP 1989
76	Rat (Fischer- 344)	Gd 11-13 (GW)		100 F		300 F (increased resorptions, implantation loss, vaginal bleeding)	NTP 1989
77	Rat (F344/N)	2 wk (W)		346 M			NTP 1993
78	Mouse (CD-1)	Gd 6-13 1x/d (GW)				1180 F (19% decrease in incidence of viable litters)	Hardin et al. 1987; Schuler et al. 1984

Table 2-3. Levels of Significant Exposure to 2-Butoxyethanol - Oral (continued)

Key to[a] figure	Species/ (Strain)	Exposure/ Duration/ Frequency (Specific Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference
79	Mouse (CD-1)	Gd 8-14 (G)		650 F		1000 F (increased incidence of resorptions)	Wier et al. 1987
	Developmental						
80	Rat (Fischer- 344)	Gd 9-11 (GW)		150 F	300 F (decreased fetal weight)		NTP 1989
81	Rat (Fischer- 344)	Gd 11-13 (GW)		300 F	600 F (decreased fetal body weight, decreased gravid uterine weight)		NTP 1989
82	Rat (Fischer- 344)	Gd 9-11 (GW)		200			NTP 1989
83	Rat (Fischer- 344)	Gd 11-13 (GW)		300			NTP 1989
84	Mouse (CD-1)	Gd 8-14 1x/d (G)		650		1000 (cleft palate in 1/5 litters)	Wier et al. 1987

Table 2-3. Levels of Significant Exposure to 2-Butoxyethanol - Oral (continued)

Key to[a] figure	Species/ (Strain)	Exposure/ Duration/ Frequency (Specific Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference
	INTERMEDIATE EXPOSURE						
	Death						
85	Rat (Crl:COBS CD (SD)BR)	23 d 5 d/wk (G)				885 M (death in 1/9)	Eastman Kodak 1983; Krasavage 1986
86	Mouse (Swiss CD-1)	21 wk (W)				1300 F (death in 6/20)	Heindel et al. 1990
87	Mouse (JCL-ICR)	5 wk 5 d/wk (GO or GW)				2000 M (5/5 dead)	Nagano et al. 1979, 1984
	Systemic						
88	Rat (Sherman)	90 d (F)	Resp	1540			Carpenter et al. 1956
			Hemato	1540			
			Hepatic	76	310 (unspecified increase in relative liver weight)		
			Renal	310	1540 (unspecified increase in relative kidney weight)		
			Bd Wt	310	1540 (unspecified decrease in body weight gain)		

Table 2-3. Levels of Significant Exposure to 2-Butoxyethanol - Oral (continued)

Key to[a] figure	Species/ (Strain)	Exposure/ Duration/ Frequency (Specific Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference
89	Rat (Crl:COBS CD (SD)BR)	6 wk 5 d/wk (G)	Resp	885 M			Eastman Kodak 1983; Krasavage 1986
			Cardio	885 M			
			Gastro		222M (mild hyperkeratosis and acanthosis in stomach)		
			Hemato			222 M (12% decreased RBC count, 7% decreased HGB, 6% increased MCH, hemoglobinuria)	
			Hepatic	222 M	443M (focal hemosiderin deposition, 30% increase in serum alkaline phosphatase activity)		
			Renal	222 M	443M (focal hemosiderin deposition in the proximal convoluted tubules)		
			Endocr	885 M			
			Ocular	885 M			
			Bd Wt	443 M	885M (7-12% decreased body weight gain in the presence of reduced feed consumption)		
			Other	443 M	885M (12-31% reduced feed consumption during days 0-20)		

Table 2-3. Levels of Significant Exposure to 2-Butoxyethanol - Oral (continued)

Key to[a] figure	Species/ (Strain)	Exposure/ Duration/ Frequency (Specific Route)	System	NOAEL (mg/kg/day)	LOAEL		Reference
					Less Serious (mg/kg/day)	Serious (mg/kg/day)	
90	Rat (Sprague-Dawley)	21 d (W)	Hepatic	506 M 444 F			Exon et al. 1991
			Renal	506 M 444 F			
			Bd Wt	506 M 444 F			
			Other	180 M	506 M (12% decreased water consumption)		
				204 F	444 F (31% decreased water consumption)		

Table 2-3. Levels of Significant Exposure to 2-Butoxyethanol - Oral (continued)

Key to[a] figure	Species/ (Strain)	Exposure/ Duration/ Frequency (Specific Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference
91	Rat (F344/N)	13 wk (W)	Resp	452 M 470 F			NTP 1993
			Cardio	452 M 470 F			
			Gastro	367 M 363 F	452 M (diarrhea) 470 F (diarrhea)		
			Hemato	129 M		281 M (decreased RBC, mild anemia) 82 F (decreased RBC, HCT, HGB)	
			Musc/skel	452 M 470 F			
			Hepatic		69[c] M (hepatocellular alteration-cells that stained eosinophilic and lacked cytoplasmic granularity) 82 F		
			Renal		69 M (moderate increase in blood urea nitrogen) 82 F (decreased urine volune, possibly due to dehydration)		
			Endocr	452 M 470 F			
			Dermal	452 M 470 F			
			Ocular	452 M 470 F			
			Bd Wt	281 M 304 F	367 M (mean body weight gains decreased 12.5%)	452 M (24% decrease in body weight gain) 363 F (12% decreased final body weight and 32.5% decrease in body weight gain)	
			Other	69 M 82 F	129 M (water consumption decreased 12.1%) 151 F (water consumption decreased 17.6%)		

Table 2-3. Levels of Significant Exposure to 2-Butoxyethanol - Oral (continued)

Key to[a] figure	Species/ (Strain)	Exposure/ Duration/ Frequency (Specific Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference
92	Rat (F344/N)	60 d (W)	Bd Wt	234 M	443 M (13% decrease in mean body weight gain)		NTP 1993
93	Rat (DW albino)	91-93 d (F)	Resp	919 M 976 F			Weil 1963
			Cardio	919 M 976 F			
			Gastro	919 M 976 F			
			Hepatic	188 M	919 M (25% increase in relative liver weight)		
				222 F	976 F (27% increase in relative liver weight)		
			Renal	188 M	919 M (18% increase in relative kidney weight)		
				222 F	976 F (23% increase in relative kidney weight)		
			Endocr	919 M 976 F			
			Bd Wt	28 M	188 M (body weight 91-7% less than controls over the course of the study)	919 M (body weight gain 53% lower than controls)	
				222 F		976 F (body weight gain 45% lower than controls)	
			Other	28 M	188 M (food intake 18% lower than controls)		
				222 F	976 F (food intake 23% lower than controls)		

Table 2-3. Levels of Significant Exposure to 2-Butoxyethanol - Oral (continued)

Key to[a] figure	Species/ (Strain)	Exposure/ Duration/ Frequency (Specific Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference
94	Mouse (Swiss CD-1)	25 wk (W)	Hepatic	1300			Heindel et al. 1990
			Renal		1300 (13-22% increase in kidney weight)		
			Bd Wt	1300 M			
					1300 F (10% decrease in terminal body weight)		
			Other		700 (unspecified low water consumption)		
95	Mouse (Swiss CD-1)	14 wk (W)	Hepatic		700 (6-9% increase in absolute liver weight)		Heindel et al. 1990
			Renal	700 M			
					700 F (22% increase in absolute kidney/adrenal weight)		
			Bd Wt	700			
96	Mouse (JCL-ICR)	5 wk 5 d/wk (G)	Hemato			500 M (decrease in RBC count)	Nagano et al. 1979, 1984

Table 2-3. Levels of Significant Exposure to 2-Butoxyethanol - Oral (continued)

Key to[a] figure	Species/ (Strain)	Exposure/ Duration/ Frequency (Specific Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference
97	Mouse (B6C3F1)	13 wk (W)	Resp	694 M 1306 F			NTP 1993
			Cardio	694 M 1306 F			
			Gastro	694 M 1306 F			
			Musc/skel	694 M 1306 F			
			Hepatic	694 M 1306 F			
			Renal	694 M 1306 F			
			Endocr	694 M 1306 F			
			Dermal	694 M 1306 F			
			Ocular	694 M 1306 F			
			Bd Wt	223 M 370 F	553M (18.5% decreased body weight gain)	676 F (26.1% decreased mean body weight gain; 10% decreased mean final body weight)	
	Immunological/Lymphoreticular						
98	Rat (Crl:COBS CD (SD)BR)	6 wk 5 d/wk (G)		222 M	443M (enlarged, dark spleen in 3/9, 57% increase in spleen weight)		Eastman Kodak 1983; Krasavage 1986

Table 2-3. Levels of Significant Exposure to 2-Butoxyethanol - Oral (continued)

Key to[a] figure	Species/ (Strain)	Exposure/ Duration/ Frequency (Specific Route)	System	NOAEL (mg/kg/day)	LOAEL		Reference
					Less Serious (mg/kg/day)	Serious (mg/kg/day)	
99	Rat (Sprague-Dawley)	21 d (W)		506 M 444 F			Exon et al. 1991
100	Rat (F344/N)	13 wk (W)		69 M 82 F	129 M (increased hemosiderin 151 F pigmentation in spleen)		NTP 1993
101	Mouse (B6C3F1)	13 wk (W)		694 M 1306 F			NTP 1993
	Neurological						
102	Rat (Crl:COBS CD (SD)BR)	6 wk 5 d/wk (G)		222 M			Eastman Kodak 1983; Krasavage 1986
103	Rat (F344/N)	13 wk (W)		452 M 470 F			NTP 1993
104	Mouse (B6C3F1)	13 wk (W)		694 M 1306 F			NTP1993
	Reproductive						
105	Rat (Crl:COBS CD (SD)BR)	6 wk 5 d/wk (G)		885 M			Eastman Kodak 1983; Krasavage 1986

Table 2-3. Levels of Significant Exposure to 2-Butoxyethanol - Oral (continued)

Key to[a] figure	Species/ (Strain)	Exposure/ Duration/ Frequency (Specific Route)	System	NOAEL (mg/kg/day)	LOAEL				Reference
					Less Serious (mg/kg/day)		Serious (mg/kg/day)		
106	Rat (Sprague-Dawley)	21 d (W)		506 M					Exon et al. 1991
107	Rat (F344/N)	13 wk (W)		129 M 304 F	281 M 363 F	(11.3% decreased sperm concentration) (altered estrous cycle)			NTP 1993
108	Rat (F344/N)	60 d (W)		443 M					NTP 1993
109	Mouse (Swiss CD-1)	21 wk (W)		700			1300	(21% decrease in litters/pair, 51% decrease in pups/litter)	Heindel et al. 1990
110	Mouse (Swiss CD-1)	25 wk (W)					1300 F	(58% decrease in fertility, 66% decrease in live pups per litter; altered estrous cycle)	Heindel et al. 1990
111	Mouse (Swiss CD-1)	14 wk (W)		700					Heindel et al. 1990
112	Mouse (JCL-ICR)	5 wk 5 d/wk (G)		1000 M					Nagano et al. 1979, 1984

Table 2-3. Levels of Significant Exposure to 2-Butoxyethanol - Oral (continued)

Key to[a] figure	Species/ (Strain)	Exposure/ Duration/ Frequency (Specific Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference
113	Mouse (B6C3F1)	13 wk (W)		694 M 1306 F			NTP 1993
	Developmental						
114	Mouse (Swiss CD-1)	21 wk (W)			700 (decrease in live pup weight)		Heindel et al. 1990
115	Mouse (Swiss CD-1)	14 wk (W)		700			Heindel et al. 1990

[a]The number corresponds to entries in Figure 2-3. Differences in levels of health effects and cancer effects between males and females are not indicated in Figure 2-3. Where such differences exist, only the levels of effect for the most sensitive gender are presented.

[b]Used to derive an acute oral minimal risk level (MRL) of 0.4 mg/kg/day; dose divided by an uncertainty factor of 90 (10 for use of a LOAEL, 3 for extrapolation from animals to humans, and 3 for human variability). For further details, see MRL worksheets in Appendix A.

[c]Used to derive an intermediate oral MRL of 0.07 mg/kg/day; dose divided by an uncertainty factor of 1,000 (10 for use of a LOAEL, 10 for extrapolation from animals to humans, and 10 for human variability). For further details, see MRL worksheets in Appendix A.

Bd Wt = body weight; Cardio = cardiovascular; d = day(s); Endocr = endocrine; F = female; (G) = gavage; Gastro = gastrointestinal; Gd = gestational day; Gn Pig = guinea pig; (GO) = gavage in oil; (GW) = gavage in water; HCT = hematocrit; Hemato = hematological; HGB = hemoglobin; (IN) = ingestion; LD_{50} = lethal dose, 50% kill; LOAEL = lowest-observable-adverse-effect level; M = male; MCH = mean corpuscular hemoglobin; MCHC = mean corpuscular hemoglobin concentration; MCV = mean corpuscular volume; Metab = metabolic; Musc/skel = musculoskeletal; NOAEL = no-observable-adverse-effect level; NS = not specified; PCV = packed cell volume; RBC = red blood cell; Resp = respiratory; (W) = water; WBC = white blood cell; wk = week(s); x = times.

Table 2-4. Levels of Significant Exposure to 2-Butoxyethanol Acetate - Oral

Key to[a] figure	Species/ (Strain)	Exposure/ Duration/ Frequency (Specific Route)	System	NOAEL (mg/kg/day)	LOAEL		Reference
					Less Serious (mg/kg/day)	Serious (mg/kg/day)	
	ACUTE EXPOSURE						
	Death						
1	Rat (Wistar)	once (GO)				3000 M (LD_{50}) 2400 F (LD_{50})	Truhaut et al. 1979

[a] The number corresponds to entries in Figure 2-4. Differences in levels of health effects and cancer effects between males and females are not indicated in Figure 2-4. Where such differences exist, only the levels of effect for the most sensitive gender are presented.

F = female; (GO) = gavage with oil; LD_{50} = lethal dose, 50% kill; LOAEL = lowest-observable-adverse-effect level; M = male; NOAEL = no-observable-adverse-effect level

Figure 2-3. Levels of Significant Exposure to 2-Butoxyethanol - Oral

Acute (≤14 days)

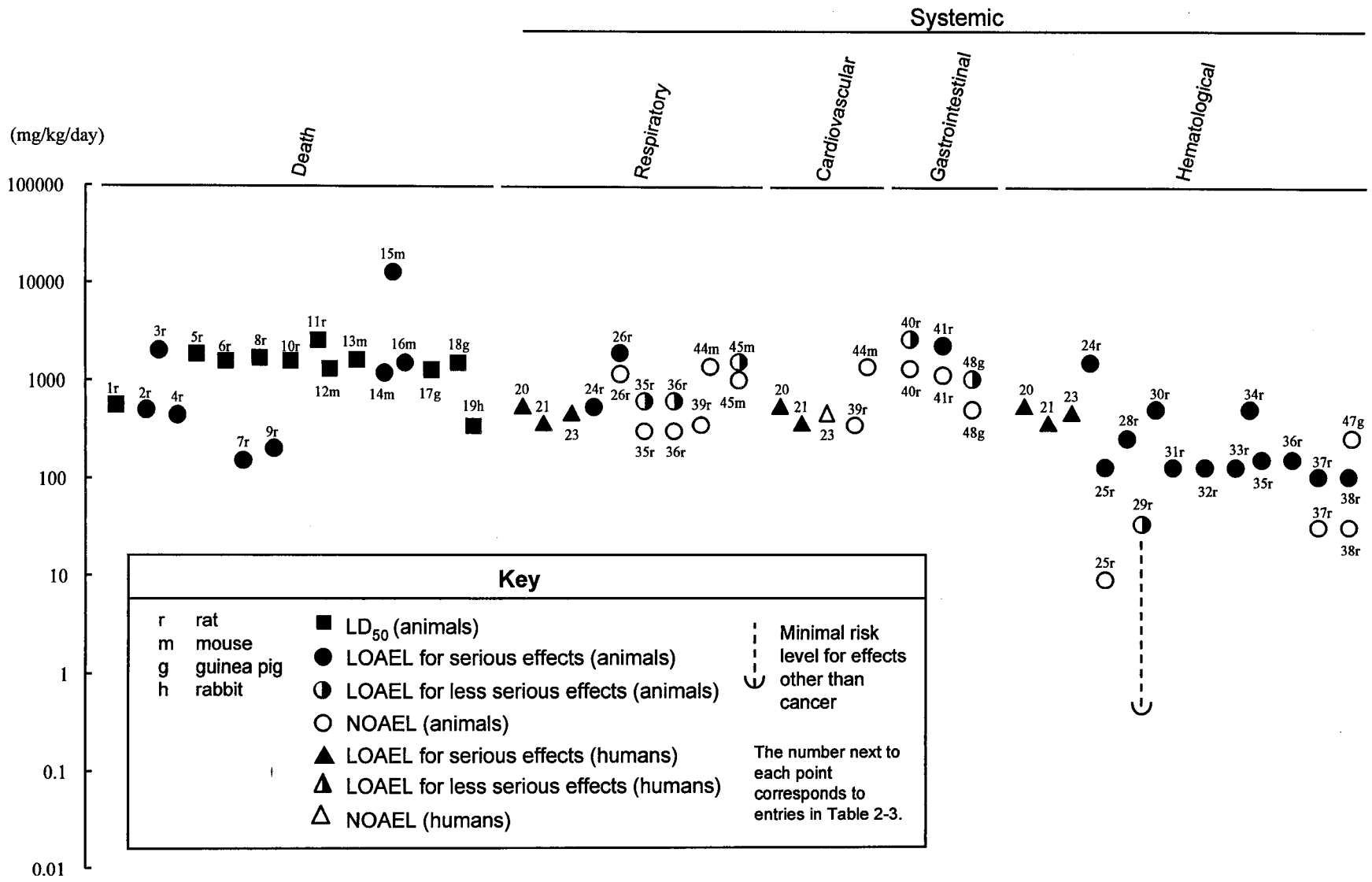


Figure 2-3. Levels of Significant Exposure to 2-Butoxyethanol - Oral (cont.)

Acute (≤14 days)

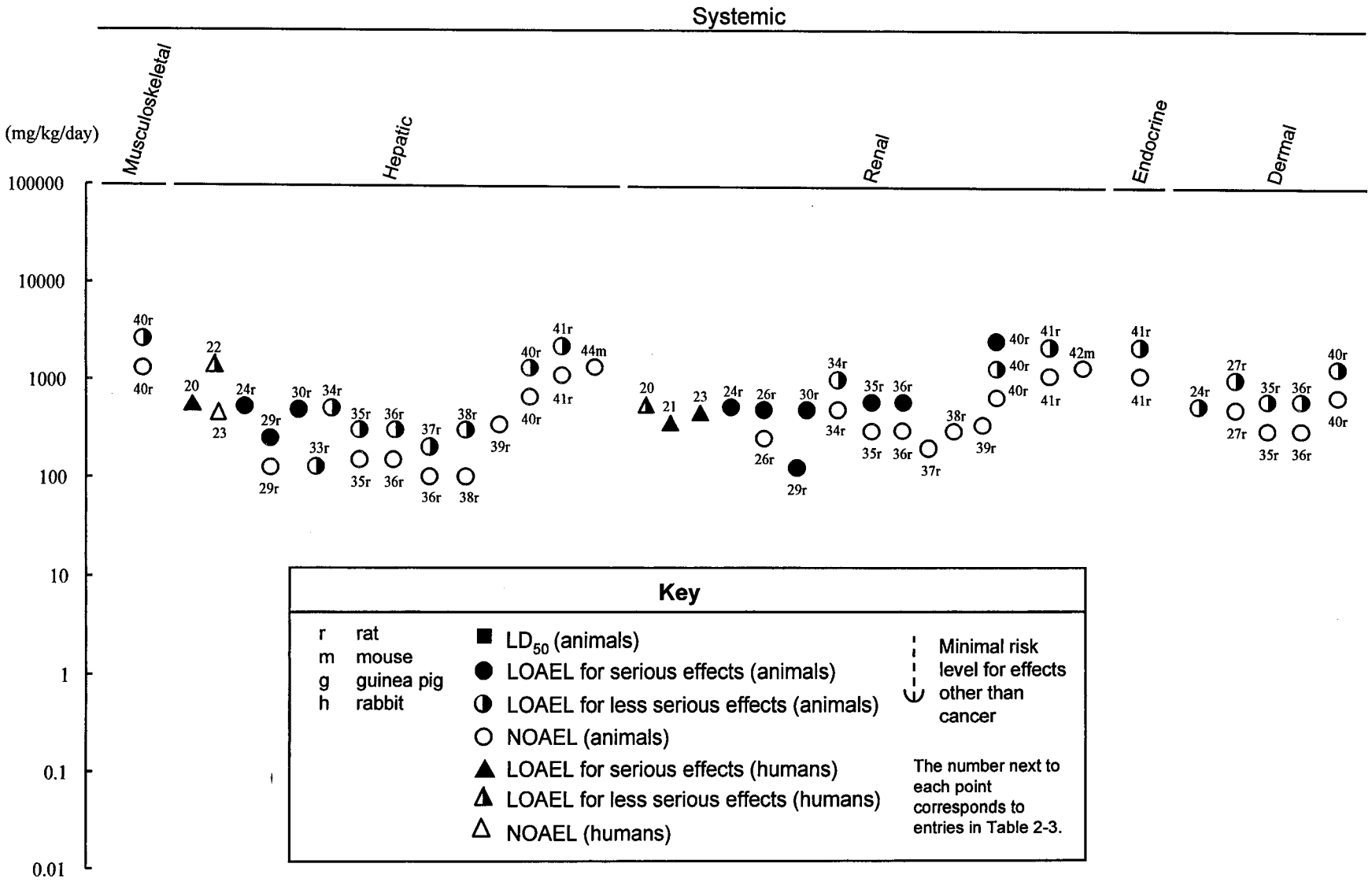


Figure 2-3. Levels of Significant Exposure to 2-Butoxyethanol - Oral (cont.)

Acute (≤14 days)

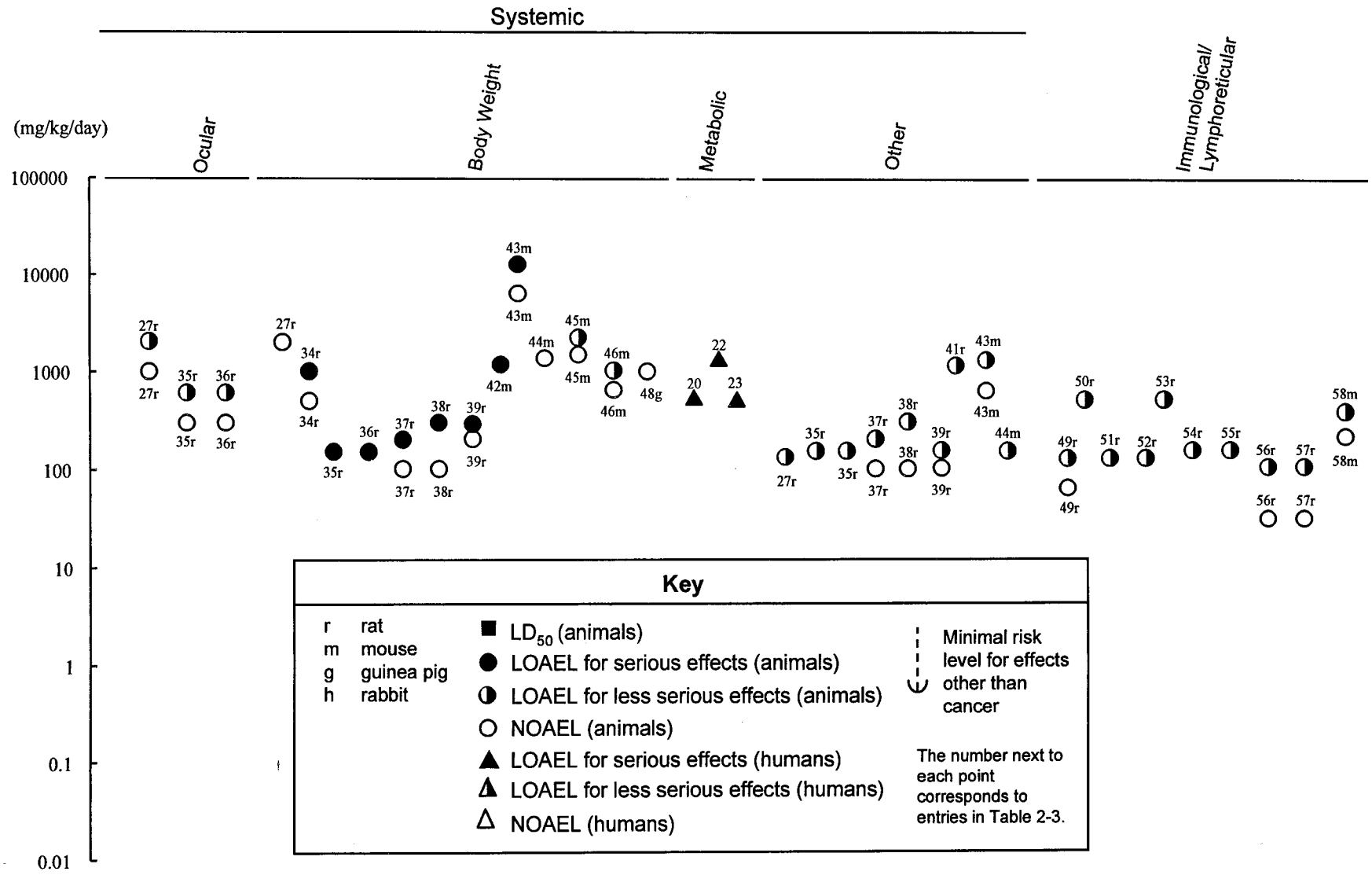


Figure 2-3. Levels of Significant Exposure to 2-Butoxyethanol - Oral (cont.)

Acute (≤14 days)

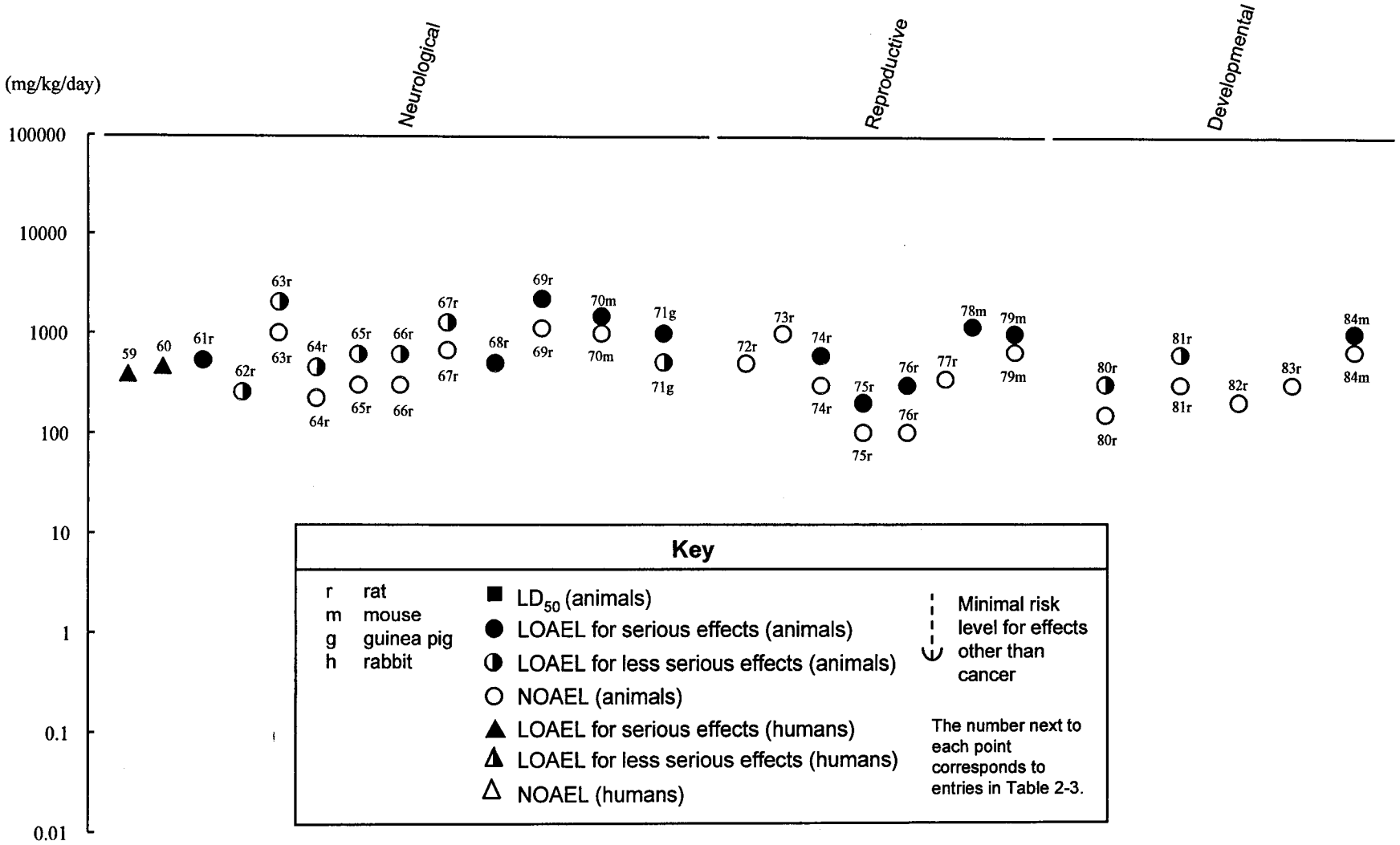


Figure 2-3. Levels of Significant Exposure to 2-Butoxyethanol - Oral (cont.)

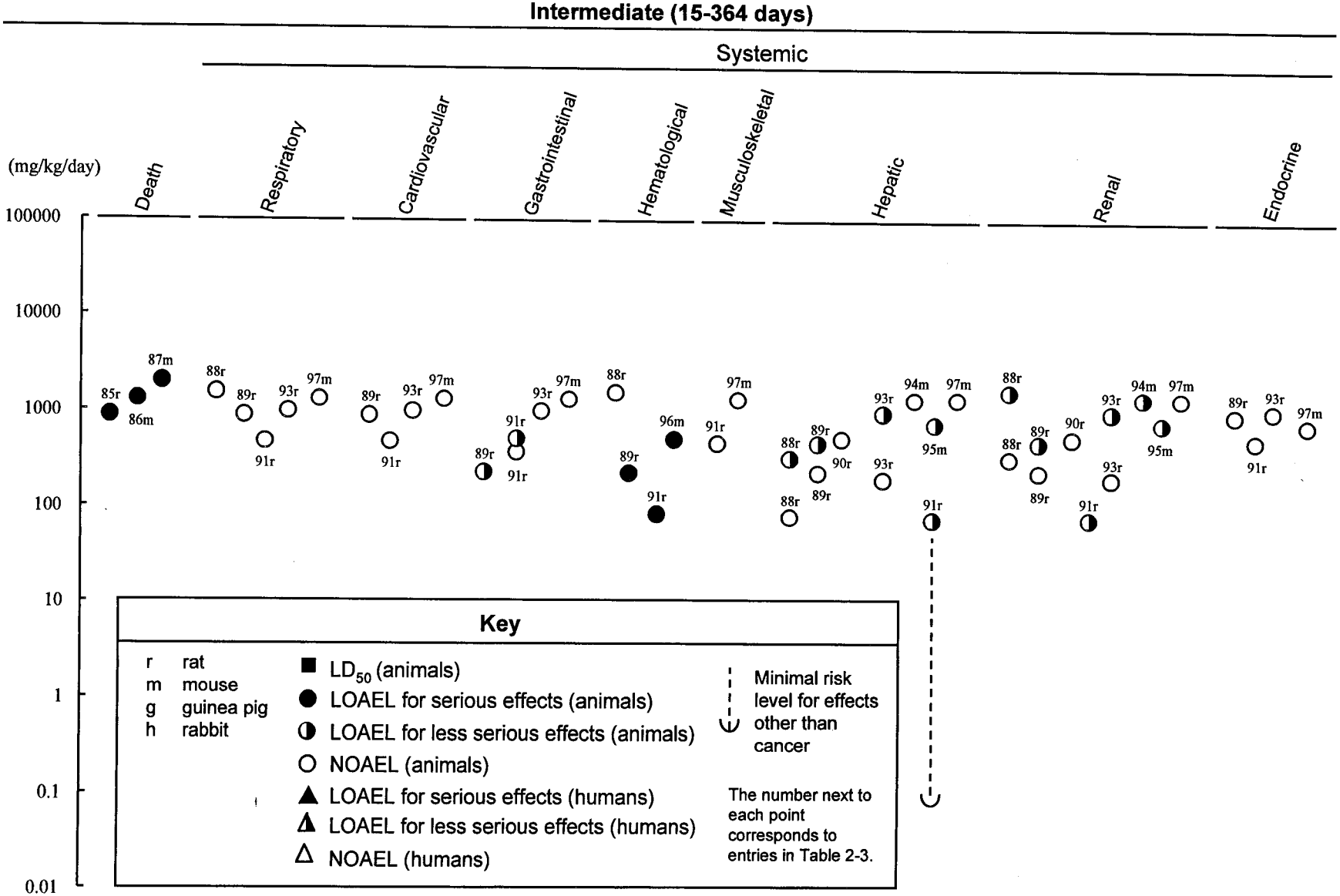


Figure 2-3. Levels of Significant Exposure to 2-Butoxyethanol - Oral (cont.)

Intermediate (15-364 days)

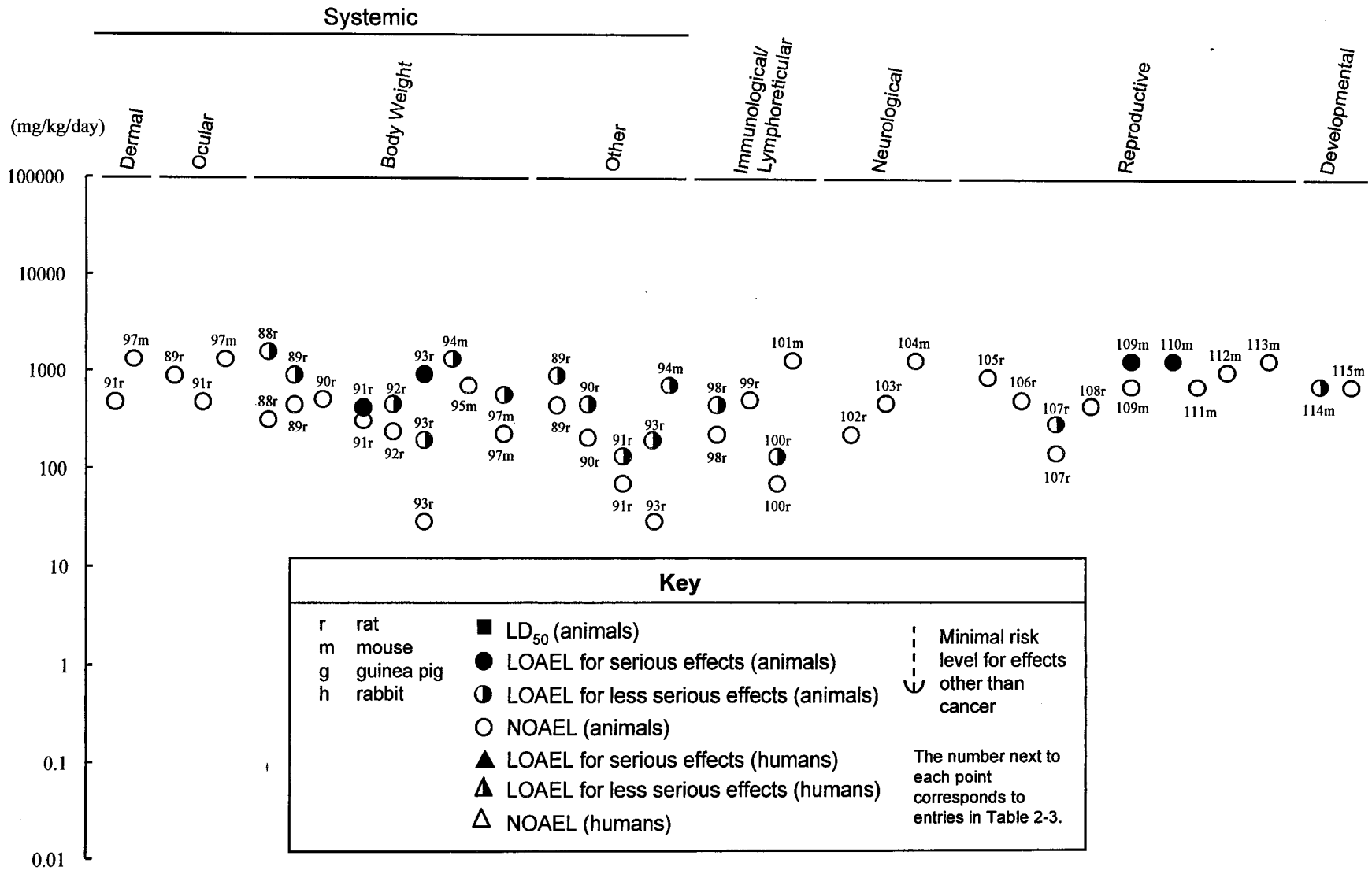


Figure 2-4. Levels of Significant Exposure to 2-Butoxyethanol Acetate - Oral

Acute (≤14 days)

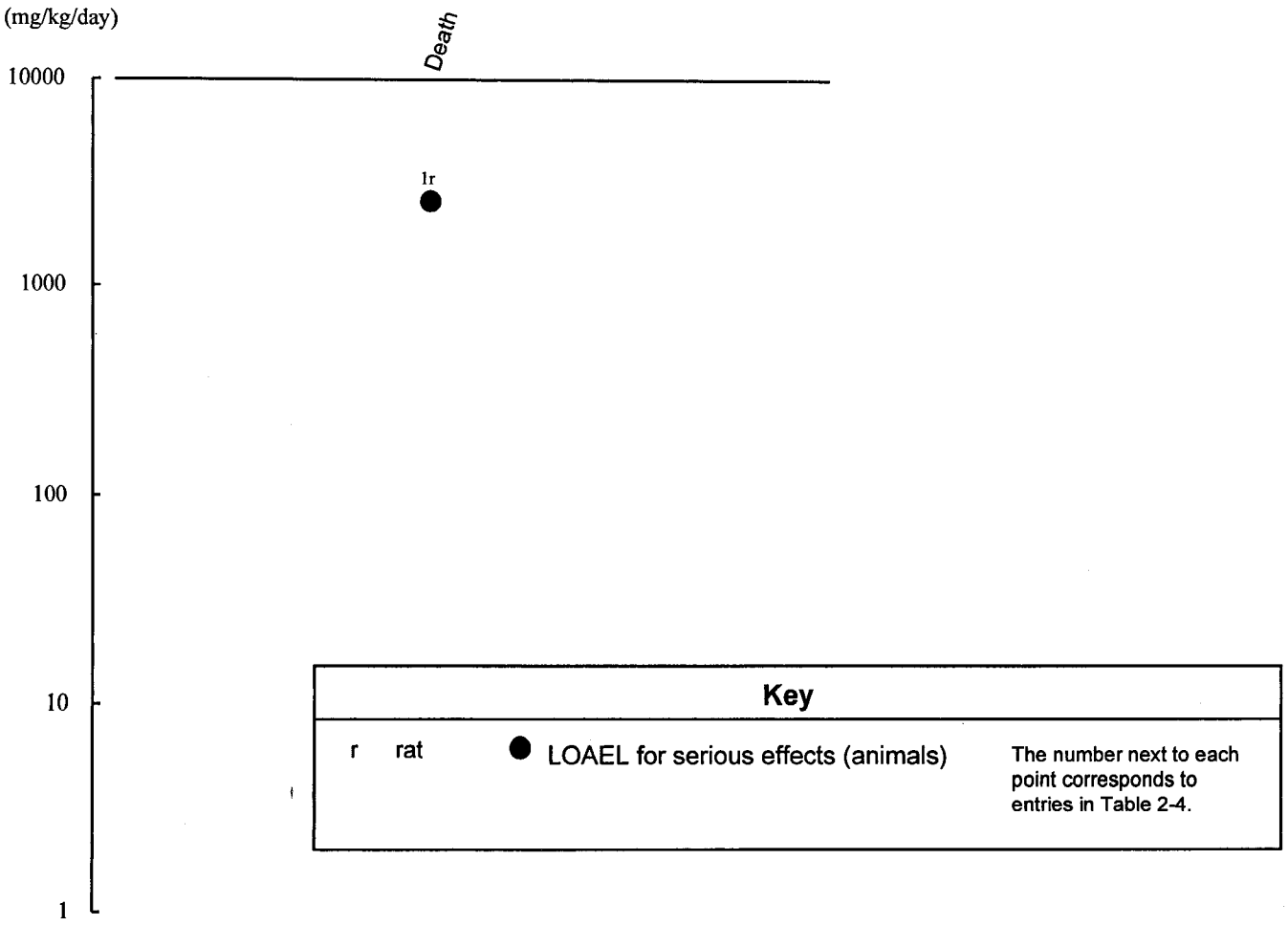


Table 2-5. Levels of Significant Exposure to Butoxyacetic Acid - Oral

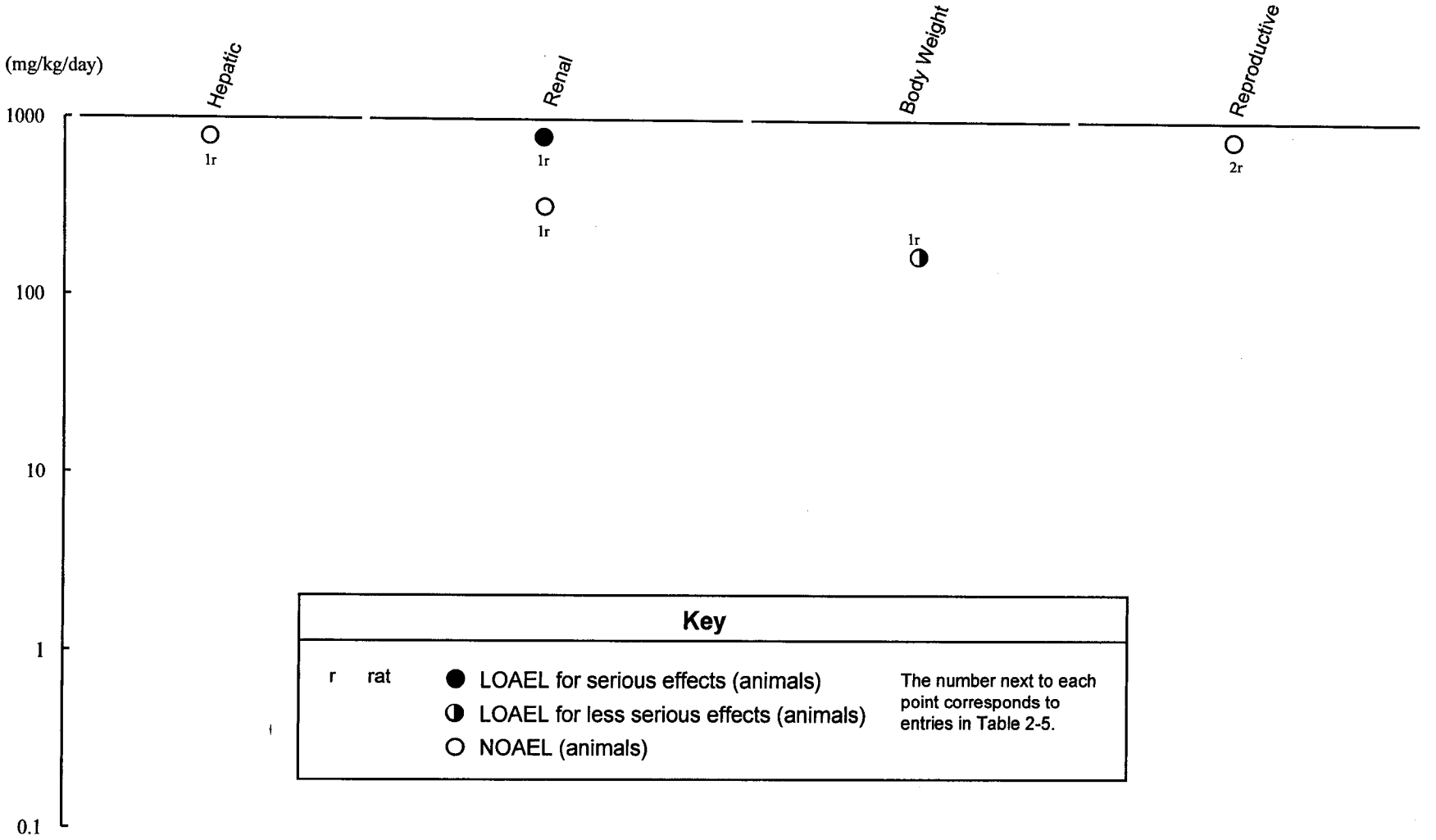
Key to[a] figure	Species/ (Strain)	Exposure/ Duration/ Frequency (Specific Route)	System	NOAEL (mg/kg/day)	LOAEL		Reference
					Less Serious (mg/kg/day)	Serious (mg/kg/day)	
	ACUTE EXPOSURE						
	Systemic						
1	Rat (Alpk/AP)	once (GW)	Hepatic	868 M			Foster et al. 1987
			Renal	434 M		868 M (hematuria)	
			Bd Wt		174M (unspecified decrease in body weight gain)		
	Reproductive						
2	Rat (Alpk/AP)	once (GW)		868 M			Foster et al. 1987

[a] The number corresponds to entries in Figure 2-5.

Bd Wt = body weight; (GW) = gavage in water; LOAEL = lowest-observable-adverse-effect level; M = male; NOAEL = no-observable-adverse-effect level

Figure 2-5. Levels of Significant Exposure to 2-Butoxyethanol Acid - Oral

Acute (≤14 days)



Animals also exhibit abnormal breathing after ingestion of high doses of 2-butoxyethanol. Female Fischer 344 rats had rapid, shallow, or difficult breathing after a single gavage dose of 2,000 mg/kg 2-butoxyethanol (Dow 1981) or a single gavage dose of 4,510 or 9,019 mg/kg (Union Carbide 1980b). The 2,000-mg/kg dose resulted in the death of two of three of the rats treated (Dow 198 l), while all rats (five in each group) treated with 4,510 mg/kg or 9,019 mg/kg died (Union Carbide 1980b). Rats (Wistar, Sherman, and Carworth-Wistar) also exhibited congested or hemorrhaged lungs at death after a gavage dose of 530 mg/kg (Carpenter et al. 1956). However, no overt respiratory effects were seen in Fischer 344 rats or B6C3F$_1$ mice after ingestion of 2-butoxyethanol at doses of ≤346 mg/kg/day (males) or ≤265 mg/kg/day (females) for rats and ≤ 627 mg/kg/day (males) or ≤ 1,364 mg/kg/day (females) for mice in drinking water for 2 weeks (NTP 1993). Pregnant Fischer 344 rats exhibited dyspnea after oral dosing by gavage in a preliminary teratology study with 600 mg/kg/day 2-butoxyethanol on gestational days 9-l1 or 1l-13 (NTP 1989). In the definitive study, no respiratory effects were noted at doses ≤200 mg/kg/day on gestational days 9-l 1 or ≤ 300 mg/kg/day on gestational days 11-13 (NTP 1989). However, in mated CD-1 mice given 0, 350, 650, 1,000, 1,500, or 2,000 mg/kg/day 2-butoxyethanol by gavage on gestational days 8-14, abnormal breathing was observed at doses of 1,500 and 2,000 mg/kg/day (Wier et al. 1987). These doses resulted in the deaths of three of six mice at 1,500 mg/kg/day and six of six mice at 2,000 mg/kg.

In intermediate-duration studies, no overt signs of respiratory effects were found in male and female Sherman rats after ingestion of ≤ 1,540 mg/kg/day 2-butoxyethanol in the feed for 90 days (Carpenter et al. 1956). No overt signs of respiratory effects and no gross or histologically observed lung or nasal cavity lesions were observed in male COBS CD (SD)BR rats receiving 885 mg/kg/day by gavage for 6 weeks (Eastman Kodak 1983; Krasavage 1986) or in Fischer 344 rats receiving doses ≤452 mg/kg/day (males) and ≤470 mg/kg/day (females) or B6C3F$_1$ mice receiving ≤694 mg/kg/day (males) and ≤ 1,306 mg/kg/day (females) in drinking water for 13 weeks (NTP 1993). In addition, no histopathological lesions were found in the nasal turbinates of rats or mice in the NTP (1993) 13-week drinking water study. Histopathological changes in the lungs were not observed in DW Albino rats treated with 2-butoxyethanol at doses up to 919 mg/kg/day for males and 976 mg/kg/day for females (Weil and Carpenter 1963).

Rats given unspecified acute oral doses of 2-butoxyethanol acetate in olive oil as part of a lethality study exhibited no adverse respiratory clinical signs or gross or histopathological changes in the lungs (Truhaut et al. 1979).

Cardiovascular Effects. An 87-year-old woman died from cardiac arrest 3 days after she ingested an unknown amount of a cleaner containing 6.5% 2-butoxyethanol (Litovitz et al. 1991). Before death she also experienced hypotension and ventricular tachycardia and arrhythmias. Ingestion of 2-butoxyethanol has been associated with cardiovascular effects including tachycardia, and low blood pressure in a patient who ingested 650 mg/kg (Bauer et al. 1992) and low blood pressure in a patient who ingested 391 mg/kg (Gijsenbergh et al. 1989). However, in another case report of a woman who ingested 467-933 mg/kg, no noticeable cardiovascular effects were observed (Rambourg-Schepens et al. 1988). In addition, two children who accidentally ingested 290 or 1,862 mg/kg 2-butoxyethanol did not exhibit adverse cardiovascular effects (Dean and Krenzelok 1992). Gastric emptying by administration of syrup of ipecac or by gastric lavage within minutes or hours of ingestion (time not specified) may have prevented any adverse effects.

2-Butoxyethanol does not appear to cause cardiovascular effects in animals after oral exposure. In Fischer 344 rats receiving ≤ 346 mg/kg/day (males) and ≤ 265 mg/kg/day (females) and in B6C3F_1 mice receiving ≤ 627 mg/kg/day (males) and ≤ 1,364 mg/kg/day (females) in the drinking water for 2 weeks, no changes in the heart weight and no gross pathological lesions were observed in hearts (NTP 1993). Similarly, in Fischer 344 rats receiving ≤ 452 mg/kg/day (males) and ≤ 470 mg/kg/day (females) and in B6C3F_1 mice receiving ≤ 694 mg/kg/day (males) and ≤ 1,306 mg/kg/day (females) in the drinking water for 13 weeks, no changes in heart weight and no gross or histologically observed lesions were observed in the heart or the aorta (NTP 1993). No effects on heart weight and no histopathological lesions in the heart were observed in adult male COBS CD (SD)BR rats given undiluted 2-butoxyethanol by gavage at doses of 0, 222, 443, or 885 mg/kg/day, 5 days per week, over a 6-week period (Eastman Kodak 1983; Krasavage 1986). Cardiovascular effects were not observed in DW Albino rats provided with 2-butoxyethanol in the drinking water for 91-93 days at doses of 919 mg/kg/day for males and 976 mg/kg/day for females (Weil and Carpenter 1963).

Male and female rats given acute oral doses of 2-butoxyethanol acetate in olive oil as part of a lethality study exhibited no adverse cardiovascular clinical signs or gross or histopathological lesions in the heart (Truhaut et al. 1979).

Gastrointestinal Effects. Very little information is available to describe gastrointestinal effects of 2-butoxyethanol by oral exposure. Wistar rats exhibited distended stomachs filled with liquid and gas, and bloody intestines at necropsy after single gavage doses of 2,255-9,019 mg/kg 2-butoxyethanol (Union Carbide 1980b). At 2,244 mg/kg, two of five rats died, while five of five rats died at each of the two highest

doses (4,510 and 9,019 mg/kg). Very red small intestines were noted 1 of 10 male Wistar rats after a single gavage dose of 2,560 mg/kg 2-butoxyethanol, a dose that resulted in the death of 9 of 10 treated animals (Olin 1976). Moderate-to-mild necrosis and hemorrhage of the gastric mucosa were observed in one of five male guinea pigs and one of five female guinea pigs treated with a single gavage dose of 1,000 mg/kg 2-butoxyethanol in water, a dose that resulted in the death of one of each sex; the contribution of the gavage procedure to these gastric effects is unclear, since there were no controls in this experiment (Shepard 1994b). Male COBS CD (SD)BR rats showed mild hyperkeratosis and acanthosis of the stomach after gavage doses of 222-885 mg/kg/day 2-butoxyethanol, 5 days per week, for 6 weeks (Eastman Kodak 1983; Krasavage 1986). No gross pathological or histopathological lesions were found in the esophagus, cecum, colon, duodenum, jejunum, or ileum. No gross or histopathological changes were observed in the esophagus, stomach, duodenum, or colon of DW Albino rats treated with 2-butoxyethanol in the diet for 3 months at doses up to 919 mg/kg/day for males and 976 mg/kg/day for females (Weil and Carpenter 1963). Diarrhea was noted in Fischer 344 rats receiving 452 mg/kg/day (males) and 470 mg/kg/day (females) 2-butoxyethanol in the drinking water for 13 weeks, although this effect was not observed at lower doses (≤ 367 mg/kg/day) (NTP 1993). In the same study, B6C3F_1 mice ingesting ≤694 mg/kg/day (males) and ≤ 1,306 mg/kg/day (females) 2-butoxyethanol in the drinking water for 13 weeks did not develop diarrhea. In addition, gross and histological examination of the esophagus, cecum, colon, rectum, duodenum, jejunum, ileum, and stomach of the rats and the mice in the 13-week drinking water study revealed no lesions.

Hematological Effects. A male patient who was an abuser of alcohol and had a history of trichloroethylene ingestion was admitted to the hospital after ingesting 650 mg/kg 2-butoxyethanol. He exhibited nonhemolytic anemia, low prothrombin time (longer time to clotting), and thrombopenia (Bauer et al. 1992). Progressive erythropenia and hemoglobinuria were noted after a woman ingested 467-933 mg/kg 2-butoxyethanol (Rambourg-Schepens et al. 1988), and a decrease in blood hemoglobin was noted in a woman who ingested 391-469 mg/kg 2-butoxyethanol (Gijsenbergh et al. 1989). Hemodialysis, used in two of these cases (Bauer et al. 1992; Gijsenbergh et al. 1989), may have contributed to hematological effects. Intravascular coagulation was observed in an 87-year-old women who died of cardiac arrest 3 days after ingesting an unknown amount of cleaner containing 6.5% 2-butoxyethanol (Litovitz et al. 1991). Two children who accidentally ingested 290 or 1,862 mg/kg 2-butoxyethanol exhibited no adverse hematological effects (Dean and Krenzelok 1992). Gastric emptying by administration of syrup of ipecac or by gastric lavage within minutes or hours of ingestion (time not specified) may have prevented any adverse effects. Hematologic effects were also not observed in a man who ingested two doses of a concentrated glass cleaner 12 days apart (Gualtieri et al. 1995). On the first occasion he ingested 1,006-l,341 mg/kg; on the second

occasion he ingested 1,341 mg/kg. After both doses, this case report indicates that the man was treated with hemodialysis and ethanol. The treatment occurred within 8 hours of the second ingestion, but it is unclear from this report how much time elapsed between the first ingestion and treatment and whether there might have been hematological consequences had these treatments not taken place.

Hemolysis and hemoglobinuria are common observations in animals treated orally with 2-butoxyethanol for acute durations (Carpenter et al. 1956; Corley et al. 1994; Dow 1959; Ghanayem and Sullivan 1993; Ghanayem et al. 1987a, 1987b, 1990b, 1992; Grant et al. 1985; NTP 1989). After Wistar, Sherman, and Cat-worth-Wistar rats were given single doses of 2-butoxyethanol by gavage, hemoglobinuria was observed in male rats at 3,000 mg/kg and in female rats at 1,500 mg/kg (Carpenter et al. 1956). These doses are in the range of the LD_{50} given by Carpenter et al. (1956). Male Fischer 344 rats receiving a single dose of 500 mg/kg 2-butoxyethanol by gavage exhibited bemolysis of erythrocytes accompanied by a drastic increase in the concentration of free hemoglobin in the plasma (Ghanayem et al. 1987b); hemoglobinuria was observed in all rats. In the same experiment, rats receiving a single dose of 125 mg/kg 2-butoxyethanol exhibited the same effects, although to a lesser degree. Hematology profiles of male Fischer 344 rats treated with single doses of 125-500 mg/kg 2-butoxyethanol by gavage indicated changes at ≥ 125 mg/kg that are characteristic of hemolysis, including increased hematocrit, packed cell volumes, and mean cell volume, followed by a decrease in these parameters as hemolysis progressed (Ghanayem et al. 1990b). In male Fischer 344 rats given 250 mg/kg 2-butoxyethanol by gavage, hemolysis was noted, characterized by an increase in mean cell volume and hematocrit, followed by a decline in hemoglobin concentration and red blood cell count (Ghanayem and Sullivan 1993). Mean corpuscular hemoglobin and mean corpuscular hemoglobin concentration initially increased and then declined. In the same study, guinea pigs dosed once with 250 mg/kg 2-butoxyethanol had no adverse hematological effects. In a metabolism study, in which male Fischer 344 rats received radiolabeled 2-butoxyethanol at doses of 8.6 and 126 mg/kg by gavage in water, two rats in the high-dose group exhibited hemolysis and hemoglobinuria (Corley et al. 1994). These effects were not observed in rats that received 8.6 mg/kg.

In another study in male Fischer 344 rats that received 125 mg/kg/day 2-butoxyethanol by gavage for 1, 2, 3, 6, or 12 consecutive days, hemolytic anemia was noted after treatment for 1-3 days, but subsided with longer exposure (Ghanayem et al. 1992). In the same study, 125 mg/kg 2-butoxyethanol was administered to rats for 3 days; the animals were then allowed to recover for 7 days. Additional treatment with 125 or 250 mg/kg 2-butoxyethanol caused a decrease in red blood cell count and hemoglobin. In male rats that were first bled and then allowed a 7-day recovery period, increased hematocrit and reticulocytes were observed in these rats

after treatment with 125 mg/kg 2-butoxyethanol. The purpose of the bleeding/recovery experiment was to assess the effect of 2-butoxyethanol on a hematopoietic system that was already responding to a depletion of red blood cells (Ghanayem et al. 1992). That pretreatment with 2-butoxyethanol is protective has also been shown by Sivarao and Mehendale (1995). In this study, blood taken from rats 7 days after treatment with a single gavage dose of 500 mg/kg 2-butoxyethanol in water was less sensitive to the hemolytic effects of the 2-butoxyethanol metabolite, 2-butoxyacetic acid, than blood taken 21 days after treatment.

Male Fischer 344 rats treated by gavage with 500 mg/kg/day 2-butoxyethanol for 4 days showed reduced red blood cell count and hemoglobin and increased mean corpuscular volume, reticulocyte counts, and mean corpuscular hemoglobin, with bone marrow hyperplasia (Grant et al. 1985). Pregnant Fischer 344 rats exhibited increased reticulocyte counts, mean corpuscular volume, mean corpuscular hemoglobin, and platelet count, and decreased red blood cell count, hemoglobin, hematocrit, and mean corpuscular hemoglobin concentration after oral dosing in a preliminary teratology study with 150 mg/kg/day 2-butoxyethanol on gestational days 9-11 or 11-13, or on gestational day 20 (NTP 1989). In the definitive study, similar effects were noted at 100 mg/kg/day on gestational days 9-11 or gestational days 11-13 (NTP 1989).

In a study showing the age-dependent hemolytic effects of 2butoxyethano1, young (4-5 weeks) and adult (9-13 weeks, 5-6 months, and 16 months) F344 rats were dosed with 0, 32, 63, 125, 250, or 500 mg/kg 2-butoxyethanol in water by gavage (Ghanayem et al. 1987a). Hematotoxicity of 2-butoxyethanol was monitored for 48 hours. In younger rats, 125 mg/kg caused no effect on hematological parameters. At 500 mg/kg, red blood cell count, hemoglobin, and hematocrit were decreased, although not as much as in the older rats. In older (9-13 weeks) rats, 2-butoxyethanol at 125 mg/kg caused severe acute hemolytic anemia resulting in significant decreases at 8 and 24 hours after treatment in red blood cell count, hemoglobin, and hematocrit, and increases 8 hours after administration in the concentration of free plasma hemoglobin. At 4 hours, peak increases in free plasma hemoglobin were seen in both younger (4-5 weeks) and older (9-13 weeks) rats dosed with 500 mg/kg. Older rats (9-13 weeks) were more susceptible to the hematotoxic effects of 2-butoxyethanol. Secondary to the hemolytic effects, 2-butoxyethanol also caused hemoglobinuria. The onset of the development of hemoglobinuria paralleled the observed decline in the concentration of free hemoglobin in the plasma. In the 32-, 63-, and 125-mg/kg dose groups, 100% of the 16-month-old rats developed hemoglobinuria; hemoglobinuria was not monitored in 16-month-old rats receiving 250 or 500 mg/kg. At 6 months of age, 63 and 125 mg/kg caused hemoglobinuria; the same effect was not seen in younger rats (4-l 3 weeks) until doses reached 125 mg/kg. Further, both the hemolytic effects and the secondary effects of 2-butoxyethanol were age-dependent, with older (5-16 months) rats being more sensitive

than younger (4-13 weeks) rats. Young rats (4-5 weeks old) and adult rats (9-13 weeks old) were treated with 500 mg/kg 2-butoxyethanol and reticulocytes and leukocytes were counted up to 48 hours after treatment. Adult (9-13 weeks old) rats exhibited neutrophilic leukocytosis and mild lymphopenia within 4 hours. This was not detected at 48 hours. An absolute increase in reticulocyte count occurred at 48 hours after dosing. In young (4-5 weeks old) rats, an absolute increase in total leukocyte, band neutrophils, lymphocytes, and monocytes was observed at 48 hours, but there was no significant effect on reticulocyte count. Based on the LOAEL of 32 mg/kg for hematological effects in the aged rats in the study by Ghanayem et al. (1987a), an acute oral MRL of 0.4 mg/kg,/day was calculated as described in the footnote to Table 2-3 and in Appendix A.

Hematological effects reflecting hemolysis have also been found in animals after intermediate-duration oral exposure to 2-butoxyethanol. In adult male COBS CD (SD)BR rats given undiluted 2-butoxyethanol by gavage at doses of 0, 222, 443, or 885 mg/kg/day, 5 days per week, over a 6-week period, dose-related decrease in red blood cell counts and hemoglobin and dose-related increase in mean corpuscular hemoglobin and hemoglobinuria were observed at all doses (Eastman Kodak 1983; Krasavage 1986). In a similar study, male JCL-ICR mice exhibited a decrease in red blood cell count after oral doses of 500 and 1,000 mg/kg/day of 2-butoxyethanol 5 days per week for 5 weeks (Nagano et al. 1979,1984). However, while no evidence of blood or hemoglobin pigments were found in the urine of male and female Sherman rats after ingestion of ≤ 1,540 mg/kg/day 2-butoxyethanol in the feed for 90 days (Carpenter et al. 1956), methods were poorly discussed, and data were not shown. Decreased red blood cell count and mild anemia were noted in male Fischer 344 rats receiving ≥ 281 mg/kg/day 2-butoxyethanol, but not ≤129 mg/kg/day, and red blood cell count, hematocrit, and hemoglobin were decreased in female rats receiving ≤82 mg/kg/day (lowest dose in females) in the drinking water for 13 weeks (NTP 1993). No hematological effects were observed in male rats receiving 69 mg/kg/day (lowest dose in males). Hematological evaluations were not performed in the $B6C3F_1$ mice ingesting ≤ 694 mg/kg/day (males) and ≤ 1,306 mg/kg/day (females) 2-butoxyethanol in the drinking water for 13 weeks (NTP 1993).

Male and female Wistar rats given unspecified acute oral doses of 2-butoxyethanol acetate in olive oil as part of a lethality study exhibited severe hemoglobinuria (Truhaut et al. 1979).

The hemolytic effects of 2-butoxyethanol and 2-butoxyethanol acetate in animals have been associated with changes in the spleen, including increased weight, engorgement, and increased hematopoiesis. These effects are discussed further in Section 2.2.2.3, Immunological and Lymphoreticular Effects.

Musculoskeletal Effects. Male Wistar rats given single oral doses of 2,560, or 5,000 mg/kg exhibited muscle flaccidity at 2,560 and 5,000 mg/kg (Glin 1976), doses which resulted in the death of 9 of 10 and 10 of 10 rats, respectively. Histological examination of thigh muscles and bones revealed no pathological lesions in Fischer 344 rats receiving doses of ≤452 mg/kg/day (males) and ≤470 mg/kg/day (females) and in $B6C3F_1$ mice receiving doses of ≤ 694 mg/kg/day (males) and < 1,306 mg/kg/day (females) 2-butoxyethanol in drinking water for 13 weeks (NTP 1993).

Hepatic Effects. No adverse hepatic effects were noted in a woman who ingested 467-933 mg/kg 2-butoxyethanol (Rambourg-Schepens et al. 1988). However, abnormal liver function (increase in aspartate aminotransferase) was noted in a man who had ingested 650 mg/kg 2-butoxyethanol (Bauer et al. 1992). The man was known to abuse alcohol and trichloroethylene, which probably contributed strongly to the abnormal liver function. In a man who ingested two doses of a concentrated glass cleaner containing 2-butoxyethanol 12 days apart, increased serum alanine aminotransferase, aspartate aminotransferase, and bilirubin were observed following the first ingestion (1,006-1,341 mg/kg) but not following the second ingestion (1,341 mg/kg) (Gualtieri et al. 1995). After both doses, this case report indicates that the man was treated with hemodialysis and ethanol; the treatment occurred within 8 hours of the second ingestion, but it is unclear how much time elapsed between the first ingestion and treatment and whether there might have been further hepatic effects had this treatment not taken place. Hepatic failure was reported in an 87-year-old woman who died of cardiac arrest 3 days after ingesting an unknown volume of glass cleaner containing 6.5% 2-butoxyethanol (Litovitz et al. 199 1).

Hepatic effects have been observed in animals exposed orally to 2-butoxyethanol. Mottled livers were observed in Wistar, Sherman, and Car-worth-Wistar rats that died after receiving ≥530 mg/kg by gavage (Carpenter et al. 1956). Young (4-5 weeks) and adult (9-13 weeks) Fischer 344 rats were dosed with 0, 32, 63, 125, 250, or 500 mg/kg 2-butoxyethanol by gavage and then killed at 24 and 48 hours after dosing. The livers were weighed, fixed, and examined for histopathological lesions (Ghanayem et al. 1987a). Histopathological results were presented for the control, 125-, 250-, and 500-mg/kg dose groups. Secondary to the hemolytic effects, 2-butoxyethanol also caused histopathologic changes in the liver, consisting of focal coagulative necrosis of hepatocytes in one of six adult rats at 250 mg/kg and in five of six adult rats at 500 mg/kg; no effects were seen in young rats even at 500 mg/kg. This effect of 2-butoxyethanol was age-, dose- and time-dependent, with older rats being more sensitive. The hepatic effects were more severe at 24 hours after treatment and showed some recovery by 48 hours after treatment. Phagocytized hemoglobin was found in the hepatic parenchymal cells and Kupffer cells, which is consistent with the role of these cells

in hemoglobin degradation. However, although phagocytized hemoglobin was frequently present in or next to areas of focal necrosis, the role of phagocytized hemoglobin, if any, in the mechanism of this necrosis is not known. Male Fischer 344 rats receiving 500 mg/kg 2-butoxyethanol by gavage exhibited similar effects on hepatic tissue (Ghanayem et al. 1987b). Dark livers were observed at the necropsy of male Wistar rats that had been given one dose of 1,310 mg/kg (Olin 1976) or 2,255 mg/kg (Union Carbide 1980b) 2-butoxyethanol by gavage. In male Fischer 344 rats that received 125 mg/kg/day 2-butoxyethanol by gavage for 1, 2, 3, 6, or 12 consecutive days, a slight decrease in relative liver weight was noted after dosing for 3 or 6 days, followed by a slight increase after dosing for 12 days (Ghanayem et al. 1992). An increase in liver weight was also noted in male Fischer 344 rats after a gavage dose of 500 mg/kg/day 2-butoxyethanol for 4 days (Grant et al. 1985). However, no effects on liver weight and no gross pathological hepatic effects were seen in Fischer 344 rats receiving ≤346 mg/kg/day (males) and ≤265 mg/kg/day (females) or in $B6C3F_1$ mice receiving ≤ 627 mg/kg/day (males) and ≤ 1,364 mg/kg/day (females) of 2-butoxyethanol in drinking water for 2 weeks (NTP 1993).

Pregnant Fischer 344 rats exhibited decreased absolute liver weight 24 hours after oral dosing in a preliminary teratology study at 300 and 600 mg/kg/day 2-butoxyethanol on gestational days 9-l 1 or 11-13 (NTP 1989). In the definitive study, decreased absolute maternal liver weight was noted at termination on gestational days 12 or 20 after dosing with 200 mg,Q/day on gestational days 9-l 1 and at termination on gestational day 14 after dosing with 300 mg/kg/day on gestational days 11-13 (NTP 1989).

Hepatic effects have also been observed in animals after intermediate oral exposure to 2-butoxyethanol. An unspecified increase in relative liver weight was reported for Sherman rats that received 1,540 mg/kg/day in the diet for 90 days (Carpenter et al. 1956). Absolute, but not relative, liver weight was only slightly increased (5%) in male Sprague-Dawley rats that received 180 mg/kg/day in the drinking water for 21 days (Exon et al. 1991). No increase in liver weight was observed in the female rats at ≤444 mg/kg/day or in the males at 506 mg/kg/day. Furthermore, histological examination of livers revealed no pathological lesions. In DW Albino rats treated with 2-butoxyethanol in the diet for 91-93 days, relative liver weights were increased by 25% in males at 9 19 mg/kg/day and 27% in females at 976 mg/kg/day (Weil and Carpenter 1963). This effect may have been associated with decreased body weight gain and food consumption. No effects on liver weight were noted at lower doses (188 mg/kg/day males, 222 mg/kg/day females), nor were any histopathological changes observed at any dose.

Adult male COBS CD (SD)BR rats given ≤885 mg/kg/day 2-butoxyethanol for 6 weeks showed increased relative, but not absolute, liver weight in all dose groups (≥222 mg/kg/day) (Eastman Kodak 1983;Krasavage 1986). Histological examination of livers revealed hemosiderin deposition in the liver at ≥443 mg/kg/day and hepatocytomegaly at 885 mg/kg/day. Statistically significant increases were found for serum alkaline phosphatase at ≥443 mg/kg/day and for serum alanine aminotransferase at 885 mg/kg/day.

In Fischer 344 rats exposed to 2-butoxyethanol in the drinking water for 13 weeks, no treatment-related changes in liver weight were observed (NTP 1993). Clinical chemistry evaluation revealed significantly increased levels of serum alkaline phosphatase in males (452 mg/kg/day) at 3 weeks and in females (363 and 470 mg/kg/day) at 13 weeks, and significantly increased levels of serum alanine aminotransferase in males (452 mg/kg/day) and in females (363 and 470 mg/kg/day) at 13 weeks . The increase in alkaline phosphatase levels is consistent with mild cholestasis, while increased serum alanine aminotransferase levels are consistent with hepatocellular necrosis. Histological examination of the livers revealed hepatocellular alterations (hepatocytes that stained more eosinophilic and lacked the amphophilic to basophilic granularity of the cytoplasm) at all doses (≥69 mg/kg/day for males and ≥ 82 mg/kg/day in females); centrilobular hepatocellular degeneration in males at ≥281 mg/kg/day and in females at ≥ 304 mg/kg/day; and brown to green granular pigment staining strongly positive for iron in Kupffer cell cytoplasm in males at 452 mg/kg/day and in females at ≥ 151 mg/kg/day. These lesions, particularly in the females, displayed both increased doserelated incidence and increased dose-related severity. In contrast, no hepatic effects or gall bladder lesions were found in $B6C3F_1$ mice ingesting ≤ 694 mg/kg/day (males) and ≤ 1,306 mg/kg/day (females) 2-butoxy-ethanol in the drinking water for 13 weeks (NTP 1993). Furthermore, no adverse effects on liver weight or histopathology in the liver were noted in Swiss CD-1 male and female mice dosed with 1,300 mg/kg/day 2-butoxyethanol in the drinking water for 25 weeks (Heindel et al. 1990). However, the low- and high-dose mice (700 and 2,000 mg/kg/day, respectively) were not necropsied. When the offspring of these mice in the 700-mg/kg/day dose group were continued on the same treatment beginning at weaning and evaluated after 14 weeks, liver weight was significantly increased by 8% above the control value. Based on the LOAEL of 69 mg/kg/day for hepatic effects in male rats in the 13-week drinking study by NTP (1993), an intermediate oral MRL of 0.07 mg/kg/day was calculated as described in the footnote to Table 2-3 and in Appendix A.

Histological examination of the livers revealed no pathological lesions in male and female Wistar rats given unspecified acute oral doses of 2-butoxyethanol acetate in olive oil as part of a lethality study (Truhaut et al. 1979).

No histopathological lesions were found in the livers of male Alpk/AP (Wistar-derived) rats that were dosed once by gavage with 0, 174, 434, or 868 mg/kg 2-butoxyacetic acid (Foster et al. 1987).

Renal Effects. Intentional ingestion of 2-butoxyethanol by humans has been associated with renal toxicity. Renal failure was reported in an 87-year-old woman that died of cardiac arrest 3 days after ingesting an unknown amount of cleaner containing 6.5% 2-butoxyethanol (Litovitz et al. 1991). A male patient who was an abuser of alcohol and had a history of trichloroethylene ingestion was admitted to the hospital after ingesting 650 mg/kg 2-butoxyethanol contained in a household cleaning product; he exhibited slight albuminuria upon admission (Bauer et al. 1992). Similarly, a woman who ingested 467-933 mg/kg 2-butoxyethanol (also in window cleaner) developed oxahuia and a transient rise in serum creatinine (Rambourg-Schepens et al. 1988). Another female patient developed hematuria after ingesting 391-469 mg/kg 2-butoxyethanol, and also exhibited increased urinary excretion of oxalic acid, although within normal limits (Gijsenbergh et al. 1989). All three patients recovered after supportive therapy. In contrast, two children who accidentally ingested 290 or 1,862 mg/kg 2-butoxyethanol showed no adverse renal effects (Dean and Krenzelok 1992). Gastric emptying by administration of syrup of ipecac or by gastric lavage within minutes or hours of ingestion (time not specified) may have prevented any adverse effects. Renal effects were not observed in a man who ingested two doses (first dose, 1,006-1,341 mg/kg; second dose, 1,341 mg/kg) of a concentrated glass cleaner containing 2-butoxyethanol 12 days apart (Gualtieri et al. 1995). The details about how renal function was measured were not provided in this case report. After both doses, the man was treated with hemodialysis and ethanol; the treatment occurred within 8 hours of the second ingestion, but it is unclear from this report how much time elapsed between first ingestion and treatment and whether there might have been renal consequences had this treatment not taken place.

Renal effects, including hematuria, have been observed in animals after acute oral exposure to 2-butoxyethanol. Severely congested kidneys have been observed in rats that died after gavage doses of ≥ 530 mg/kg (Carpenter et al. 1956). Dark and/or enlarged kidneys were observed in male Wistar rats that received one dose of 1,310 mg/kg. Hematuria was observed in Wistar rats that received one dose of 2,560 or 5,000 mg/kg (Olin 1976) or ≥2,255 mg/kg in rats that died (Union Carbide 1980b). Hematuria was also observed in an unspecified strain of rats given 500 or 1,000 mg/kg, but not 250 mg/kg, by gavage (Dow 1959) and in female Sprague-Dawley rats given 500 mg/kg (Sivarao and Mehendale 1995). Young (4-5 weeks) and adult (9-13 weeks) male Fischer 344 rats were dosed with 0, 32, 63, 125, 250, or 500 mg/kg 2-butoxyethanol by gavage; animals were killed at 24 and 48 hours after treatment and the kidneys were weighed, fixed, and examined histologically (Ghanayem et al. 1987a). Histopathological results were presented for the control,

125-,250-, and 500-mg/kg dose groups. Secondary to the hemolytic effects, 2-butoxyethanol also caused histopathologic changes in the kidney, consisting of hemoglobin casts in the proximal tubules in 100% of the mature rats at ≥125 mg/kg; no effects were seen in young (4-5 weeks old) rats even at 500 mg/kg. The effects on the kidney in the mature rats were more severe at 24 hours after treatment and showed some recovery at 48 hours after treatment. Thus, this effect of 2-butoxyethanol was age-, dose-, and timedependent, with older rats being more sensitive. Male Fischer 344 rats receiving 500 mg/kg 2-butoxyethanol by gavage exhibited similar effects in the kidney (Ghanayem et al. 1987b).

Pregnant Fischer 344 rats exhibited urethral bleeding, which may represent hematuria, after oral dosing by gavage in a preliminary teratology study at 600 mg/kg/day (but not at ≤ 300 mg/kg/day) 2-butoxyethanol on gestational days 9-l 1 or 11-13 (NTP 1989). In the definitive study, no kidney effects were noted at doses up to 200 mg/kg/day on gestational days 9-l 1 or 300 mg/kg/day on gestational days 11-13 (NTP 1989).

Increased kidney weight has been observed in animals after acute oral exposure to 2-butoxyethanol. Male rats were given 2-butoxyethanol orally for 4 consecutive days at 500 or 1,000 mg/kg/day (Grant et al. 1985). Animals were killed on days 1, 4, 8, and 22 after the final treatment. Increased kidney weight was observed at the high dose, but histological examination of the kidneys revealed no pathological lesions. No effects on kidney weight and no gross pathological lesions in the kidney were seen in Fischer 344 rats at ≤346 mg/kg/day (males) or ≤265 mg/kg/day (females) or in $B6C3F_1$ mice at ≤627 mg/kg/day (males) or ≤ 1,364 mg/kg/day (females) in drinking water for 2 weeks (NTP 1993).

Renal effects have also been observed in animals after intermediate-duration oral exposure to 2-butoxyethanol. An unspecified, but statistically significant, increase in relative kidney weight was observed in Sherman rats at a dose of 1,540 mg/kg/day in the diet for 90 days (Carpenter et al. 1956). In DW Albino rats treated with 2-butoxyethanol in the diet for 91-93 days, relative kidney weights were increased by 18% in males at 919 mg/kg/day and 23% in females at 976 mg/kg/day (Weil and Carpenter 1963). This effect may have been associated with decreases in body weight gain and food consumption. No effects on kidney weight were noted at lower doses (188 mg/kg/day males, 222 mg/kg/day females), nor were any histopathological changes observed at any dose. No dose-related effects of treatment on kidney weight and no histopathological lesions in the kidney were observed in Sprague-Dawley rats at ≤506 mg/kg/day (males) and ≤444 mg/kg/day (females) 2-butoxyethanol in the drinking water for 21 days (Exon et al. 1991). After a 6-week exposure to 222, 443, or 885 mg/kg/day 2-butoxyethanol by gavage, male COBS CD (SD)BR rats

exhibited focal hemosiderin accumulation in the proximal tubules at 443 and 885 mg/kg/day (Eastman Kodak 1983; Krasavage 1986).

In Fischer 344 rats exposed to 2-butoxyethanol in the drinking water for 13 weeks, no treatment-related effects on kidney weight were observed (NTP 1993). Clinical chemistry and urinalysis revealed increases in blood urea nitrogen in males at ≥69 mg/kg/day at week 3 and at ≥281 mg/kg/day at week 13 and in females at ≥ 304 mg/kg/day at week 3 and ≥ 151 mg/kg/day at week 13; increases in blood creatinine in females at ≥304 mg/kg/day at week 13; decreases in total blood protein in males at ≥281 mg/kg/day at week 13 and in females at ≥151 mg/kg/day at weeks 3 and 13; decreases in blood albumin in males at ≥281 mg/kg/day at week 13 and in females at ≥363 mg/kg/day at week 3 and at ≥304 mg/kg/day at week 13; decreased urine volume in females at ≥82 mg/kg/day at week 13; and increased specific gravity of the urine in males at ≥69 mg/kg/day and in females at ≥151 mg/kg/day at week 13. Histological examination of the kidneys and urinary bladders of the rats revealed no pathological lesions. No renal effects (including no effects on kidney weight or clinical chemistry and urinalysis parameters, and no histopathological lesions in the kidney or urinary bladder) were found in $B6C3F_1$ mice in the same study at ≤694 mg/kg/day (males) and ≤ 1,306 mg/kg/day (females) 2-butoxyethanol in the drinking water for 13 weeks (NTP 1993). However, an increase in kidney weight, although with no histopathological kidney lesions, was observed at 1,300 mg/kg/day in a reproductive toxicity study in which Swiss mice were given 2-butoxyethanol in the drinking water for 25 weeks; animals from the low- and high-dose groups (700 and 2,000 mg/kg/day) were not necropsied (Heindel et al. 1990). When the offspring of these mice in the 700-mg/kg/day dose group were continued on the same treatment beginning at weaning and evaluated after 14 weeks, there was also an increase in kidney weight.

Male and female Wistar rats given unspecified acute oral doses of 2-butoxyethanol acetate in olive oil as part of a lethality study exhibited renal effects including hematuria, tubular nephrosis, cloudy swelling of cells, dilatation of the proximal and distal convoluted tubules, and acute inflammation of the interstitial tissue (Truhaut et al. 1979).

In Alpk/AP (Wistar-derived) rats dosed once by gavage with 0, 174, 434, or 868 mg/kg 2-butoxyacetic acid, hematuria was noted at 868 mgLkg but not at the lower doses (Foster et al. 1987).

Endocrine Effects. Very few data are available to describe endocrine effects of 2-butoxyethanol by oral exposure. Wistar rats exhibited red adrenal glands, which may be related to the hemolytic effects of

2-butoxyethanol, at necropsy after single gavage doses of 2,255-9,019 mg/kg (Union Carbide 1980b). It should be noted that two of five rats treated with 2,255 mg/kg and all rats treated with higher doses died. Male COBS CD (SD)BR rats showed no histopathologic changes in pancreas, adrenal glands, pituitary, thyroid, or parathyroid after oral exposure to doses of ≤885 mg/kg/day 2-butoxyethanol for 6 weeks, 5 days per week (Eastman Kodak 1983; Krasavage 1986). No adverse histological effects were noted in the adrenal glands, pancreas, thyroid, or parathyroid of DW Albino rats fed 2-butoxyethanol in the diet for 91-93 days at doses up to 919 mg/kg/day for males and 976 mg/kg/day for females (Weil and Carpenter 1963). No adverse histopathological effects on endocrine glands (including adrenal, pancreas, parathyroid, pituitary, and thyroid) were noted in Fischer 344 rats at ≤ 452 mg/kg/day (males) and 1470 mg/kg/day (females) or in B6C3F_1 mice at ≤ 694 mg/kg/day (males) and ≤ 1,306 mg/kg/day (females) (NTP 1993).

Male and female Wistar rats given unspecified acute oral doses of 2-butoxyethanol acetate in olive oil as part of a lethality study exhibited no histopathological changes in the pancreas or adrenal glands (Truhaut et al. 1979).

Dermal Effects. Female Fischer 344 rats exhibited rough hair coats and necrosis of the tail after a single dose of 1,000 mg/kg 2-butoxyethanol (Dow 1981). Necrosis of the tail tip may have been a vascular response to the toxic effects of 2-butoxyethanol on red blood cells. Rough coat was also observed in rats that died after receiving ≥530 mg/kg (Carpenter et al. 1956). Male Wistar rats exhibited piloerection after single doses of ≥1,310 mg/kg of 2-butoxyethanol by gavage (Olin 1976). Pregnant Fischer 344 rats exhibited pale coloration after oral dosing in a preliminary teratology study with 600 mg/kg/day 2-butoxyethanol on gestational days 9-11 or 11-13 (NTP 1989). In the definitive study, no dermal effects were noted at ≤200 mg/kg/day on gestational days 9-11 or ≤300 mg/kg/day on gestational days 11-13 (NTP 1989). Histological examination of skin revealed no treatment-related dermal lesions in Fischer 344 rats at ≤452 mg/kg/day (males) and ≤470 mg/kg/day (females) or in B6C3F_1 mice at ≤694 mg/kg/day (males) and ≤ 1,306 mg/kg/day (females) in drinking water for 13 weeks (NTP 1993).

Ocular Effects. A woman who ingested 391-469 mg/kg 2-butoxyethanol exhibited isoco& light reactive mydriasis upon admission to the hospital (Cijsenbergh et al. 1989). This resolved quickly after supportive therapy.

Female Fischer 344 rats exhibited palpebral closure after a single gavage dose of 2,000 mg/kg 2-butoxyethanol, a dose which resulted in the death of 2 of 3 rats (Dow 1981). Pregnant Fischer 344 rats exhibited

chromodacryorrhea after oral dosing in a preliminary teratology study with 600 mg/kg/day 2-butoxyethanol on gestational days 9-l 1 or 11-13 (NTP 1989). In the definitive study, no ocular effects were noted at ≤200 mg/kg/day on gestational days 9-l 1 or ≤300 mg/kg/day on gestational days 11-13 (NTP 1989). Histological examination of the eyes revealed no pathological lesions in male COBS CD (SD)BR rats dosed by gavage with ≤885 mg/kg/day 2-butoxyethanol for 6 weeks (Eastman Kodak 1983; Krasavage 1986), in Fischer 344 rats given 2-butoxyethanol in drinking water at ≤452 mg/kg/day (males) and .≤470 mg/kg/day (females) for 13 weeks, or in B6C3F_1 mice given 2-butoxyethanol in drinking water at ≥694 mg/kg/day (males) and ≤1,306 mg/kg/day (females) for 13 weeks (NTP 1993).

Body Weight Effects. Reduced body weight has been observed in animals after acute oral exposure to 2-butoxyethanol. While female Fischer 344 rats given single gavage doses of 2-butoxyethanol ≤2,000 mg/kg were reported to gain weight during the post-treatment observation period, no controls were used for comparison (Dow 1981). Guinea pigs treated with a single gavage dose of 500, 1,000 or 2,000 mg/kg 2-butoxyethanol were reported to gain weight during a 14-day observation period (Shepard 1994b), but no controls were used in this study. In male Fischer 344 rats, a gavage dose of 1,000 mg/kg/day for 4 days resulted in reduced body weight gain compared with controls (Grant et al. 1985). No effect on body weight was seen in male Fischer 344 rats that received 2-butoxyethanol in drinking water at ≤346 mg/kg/day for 2 weeks, but the female rats displayed a decrease in body weight gain during exposure and decreased final body weight at 265 mg/kg/day but not at ≤ 203 mg/kg/day (NTP 1993). The decrease in body weight gain may have been related to decreased water consumption. Male Swiss mice lost 31% of their initial body weight at 12,750 mg/kg/day 2-butoxyethanol administered in the drinking water for 2 weeks (Heindel et al. 1990). No weight loss was observed in the male mice at ≤2,550 mg/kg/day. The female mice were reported to respond similarly, but the degree of weight loss and the doses associated with the weight loss were not reported. No effect on body weight was observed in B6C3F_1 mice at ≤627 mg/kg/day (males) and ≤ 1,364 mg/kg/day (females) of 2-butoxyethanol in the drinking water for 2 weeks (NTP 1993).

Pregnant Fischer 344 rats exhibited decreased gestational weight gain in a preliminary teratology study at ≥150 mg/kg/day 2-butoxyethanol on gestational days 9-11 or 11-13 (NTP 1989). The decreased gestational weight gain may have been related to decreases in food and water intake. In the definitive study, gestational weight gain was reduced at 200 mg/kg/day in the pregnant rats treated on gestational days 9-11 and at 300 mg/kg/day in the pregnant rats treated on gestational days 11-13 (NTP 1989). The decreased gestational weight gain may have been related to decreases in food and water intake. In a developmental study in which pregnant CD-l mice were given 0, 350, 650, 1,000, 1,500, or 2,000 mg/kg/day 2-butoxy-

ethanol by gavage on gestational days 8-14 to determine prenatal responses, or 0, 650, or 1,000 mg/kg/day on gestational days 8-14 to determine postnatal responses, unspecified decreases in maternal body weight gain were reported at 2,000 mg/kg/day in the prenatal experiment and at 1,000 mg/kg/day in the postnatal experiment (Wier et al. 1987). Similarly, in another developmental study, pregnant CD-1 mice given a dose of 1,180 mg/kg/day on gestational days 6-l 3 exhibited an 80% decrease in body weight gain (Hardin et al. 1987; Schuler et al. 1984).

Effects on body weight have also been observed in animals exposed orally to 2-butoxyethanol for intermediate durations. An unspecified decrease in body weight gain was reported for Sherman rats that received 1,540 mg/kg/day in drinking water for 90 days (Carpenter et al. 1956). In DW Albino rats treated with 2-butoxyethanol in the diet for 91-93 days, body weight gain was decreased by 53% in males at 919 mg/kg/day, and 45% in females at 976 mg/kg/day (Weil and Carpenter 1963). Body weight gain was decreased by as much as 91% at day 1 of the study (on day 1) in males at 188 mg/kg/day, but was only 9% lower than controls by the end of the study. The l88-mg/kg/day dose in males, and the 976-mg/kg/day dose in females, was also associated with about an 18% decrease in food intake. No effects on body weight gain were noted at 28 mg/kg/day in males and 222 mg/kg/day in females. Decreased body weight gain in the presence of reduced food consumption occurred in male COBS CD (SD)BR rats given 885 mg/kg/day by gavage for 6 weeks (Eastman Kodak 1983; Krasavage 1986). While no effect on body weight was observed in Sprague-Dawley rats given ≤506 mg/kg/day (males) and ≤444 mg/kg/day (females) in drinking water for 21 days (Exon et al. 1991), decreased body weight gain was noted in male Fischer 344 rats treated for 60 days at 443 mg/kg/day 2-butoxyethanol and in Fischer 344 rats treated for 13 weeks at ≥367 mg/kg/day (males) and ≥363 mg/kg/day (females) with 2-butoxyethanol in drinking water (NTP 1993). The decreased body weight gain may have been related to decreased water consumption.

Swiss mice that were exposed to 0, 700, 1,300, or 2,100 mg/kg/day 2-butoxyethanol in the drinking water for 25 weeks exhibited decreased terminal body (at the end of the 25 week treatment period) weight at 1,300 mg/kg/day in females but not in males; the low- and high-dose groups were not evaluated (Heindel et al. 1990). The decreased body weight may have been related to decreased water consumption.When the offspring of these mice in the 700-mg/kg/day dose group were continued on the same treatment beginning at weaning and evaluated after 14 weeks, there was no effect on body weight. No effect on body weight was observed in $B6C3F_1$ mice at ≤694 mg/kg/day (males) or ≤ 1,306 mg/kg/day (females) 2-butoxyethanol in drinking water for 13 weeks (NTP 1993).

Groups of male Alpk/AP (Wistar-derived) rats were dosed once by gavage with 0, 174, 434, or 868 mg/kg 2-butoxyacetic acid (Foster et al. 1987). Body weight gain was decreased during the first 2 days after treatment at all doses, but returned to normal by day 14.

Metabolic Effects. Metabolic acidosis was reported in an 87-year-old woman who died of cardiac arrest 3 days after she ingested an unknown amount of a cleaner containing 6.5% 2-butoxyethanol (Litovitz et al. 1991). Ethylene glycol levels were 110 mg/dL in this patient shortly after hospital admission and 10 mg/dL 3 days later. A male patient, who had a history of alcohol and trichloroethylene abuse, was admitted to the hospital after ingestion of 500 mL of a household cleaning fluid containing 2-butoxyethanol, which was equivalent to a dose of 650 mg/kg (Bauer et al. 1992). Metabolic acidosis and hypoxemia with lactic acidosis were observed. In other case reports, a woman admitted to the hospital after ingesting 250-500 mL of a window cleaner containing 12% (467-933 mg/kg) 2-butoxyethanol developed metabolic acidosis and hypokalemia (Rambourg-Schepens et al. 1988) and a woman ingesting 391-469 mg/kg 2-butoxyethanol developed metabolic acidosis (Gijsenbergh et al. 1989). Significant acid-base disturbance, not described further in this case report, was observed in a man following ingestion of 1,006-l ,341 mg/kg 2-butoxyethanol in a concentrated glass cleaner (Gualtieri et al. 1995). A second ingestion of the glass cleaner (1,341 mg/kg 2-butoxyethanol) 12 days later did not result in metabolic disturbances. After both doses, the man was treated with hemodialysis and ethanol; the treatment occurred within 8 hours of the second ingestion, but it is unclear from this report how much time elapsed between the first ingestion and treatment and how the treatment affected the observed outcome. Two children who ingested 290 or 1,862 mg/kg 2-butoxyethanol did not exhibit evidence of metabolic acidosis during a 24-hour observation period following gastric emptying (Dean and Krenzelok 1992). Metabolic acidosis is likely due to the formation of butoxyacetic acid from 2-butoxyethanol (see section 2.3.3 Metabolism).

Other Systemic Effects. Decreased water consumption was seen in Fischer 344 rats ingesting 2-butoxyethanol at ≥ 152 mg/kg/day (females) and ≥242 mg/kg/day (males) in drinking water for 2 weeks (NTP 1993). In B6C3Fr mice treated similarly, decreased water consumption was seen in female mice at ≥ 150 mg/kg/day. In the same study, male mice receiving ≥370 mg/kg/day exhibited signs of dehydration. An unspecified decrease in fluid intake was reported for male and female Swiss mice at ≥ 1,275 mg/kg/day of 2-butoxyethanol administered in drinking water for 2 weeks (Heindel et al. 1990).

Pregnant Fischer 344 rats exhibited decreased food and water intake at ≥150 mg/kg/day and were cold to the touch at 600 mg/kg/day after gavage dosing in a preliminary teratology study with 2-butoxyethanol on

gestational days 9-11 or 11-13 (NTP 1989). In the definitive study, decreased food and water intake were noted at doses of 200 mg/kg/day on gestational days 9-l 1 or 300 mg/kg/day on gestational days 11-13 (NTP 1989).

Sprague-Dawley rats given ≤ 506 mg/kg/day 2-butoxyethanol in drinking water for 21 days exhibited decreased water consumption at the high dose (506 mg/kg/day for males and 444 mg/kg/day for females) (Exon et al. 1991). In male COBS CD (SD)BR rats, 885 mg/kg/day 2-butoxyethanol administered by gavage for 6 weeks caused decreased food consumption (Eastman Kodak 1983; Krasavage 1986). In Fischer 344 rats given 2-butoxyethanol in drinking water for 13 weeks, decreased water consumption was observed at ≥129 mg/kg/day (males) and ≥151 mg/kg/day (females) (NTP 1993). In $B6C3F_1$ mice similarly exposed at ≤694 mg/kg/day (males) and ≥ 1,306 mg/kg/day (females), average water consumption was variable, and no clear treatment-related patterns were evident. However, an unspecified decrease in water consumption was reported for male and female Swiss mice at 700 mg/kg/day 2-butoxyethanol administered in drinking water for up to 25 weeks (Heindel et al. 1990). Decreased food intake was observed DW albino rats treated with 2-butoxyethanol in the diet (Weil and Carpenter 1963). Food intake was about 18% lower than controls in males fed 188 mg/kg/day and 23% lower than controls in females fed 976 mg/kg/day. No effects on food intake were observed at 28 mg/kg/day in males or at 222 mg/kg/day in females.

Staining of the perineal region was observed in female Fischer 344 rats given single gavage doses of ≥130 mg/kg 2-butoxyethanol (Dow 1981). In a developmental study, in which pregnant CD-l mice were given 350, 650, 1,000, 1,500, or 2,000 mg/kg/day on gestational days 8-14, a green-brown or red-brown staining of cage papers was observed at ≥650 mg/kg/day (Wier et al. 1987). At 1,500 and 2,000 mg/kg/day, green or red vaginal discharge was also observed. These effects were most likely the result of hematuria, hemoglobinuria, or vaginal bleeding (see Section 2.2.2.5), although the investigators did not draw this conclusion in their publications.

2.2.2.3 Immunological and Lymphoreticular Effects

No studies were located regarding immunological or lymphoreticular effects in humans after oral exposure to 2-butoxyethanol or 2-butoxyethanol acetate. No studies were located regarding immunological effects in animals after oral exposure to 2-butoxyethanol acetate. No studies were located regarding immunological or lymphoreticular effects in animals or humans after oral exposure to 2-butoxyacetic acid.

Information on immunological effects of 2-butoxyethanol in animals exposed orally to 2-butoxyethanol is limited to one acute and one intermediate study. Male Fischer 344 rats were immunized with trinitrophenyl-lipopolysaccharide (TNP-LPS) and then dosed by gavage (4 and 28 hours after immunization) to 0, 50, 100, 200, or 400 mg/kg/day 2-butoxyethanol in distilled water for 2 consecutive days (Smialowicz et al. 1992). Three days later, the primary plaque-forming cell (PFC) response to TNP-LPS was determined. The PFC response to TNP-LPS was used to evaluate B-lymphocyte antibody production. No toxic effect of 2-butoxyethanol in the immune system was detected. In Sprague-Dawley rats, in which males received doses of 180 or 506 mg/kg/day 2-butoxyethanol and females received doses of 204 or 444 mg/kg/day 2-butoxyethanol in the drinking water for 21 days, serum samples were analyzed for antibody levels to keyhole limpet hemocyanin (KLH), and spleens were used as the cell source to assess natural killer cell cytotoxicity and interleukin 2 and interferon production. In addition, the weights of thymus and spleen were determined and thymuses were examined histologically. For antigen presentation, KLH was injected subcutaneously in the base of the tail 7 days after the beginning of treatment with 2-butoxyethanol and again on day 13 to initiate an IgG antibody response. On day 20, KLH was injected into the right footpad to assess the 24-hour delayed-type hypersensitivity reaction by subtracting the thickness of the saline-injected left footpad from the thickness of the KLH-injected footpad. There was no effect of 2-butoxyethanol treatment on anti-KLH IgG antibody production and on delayed-type hypersensitivity reaction. Splenic natural killer cytotoxic responses, which were assessed by the 4-hour ^{51}Cr release from YAC-1 tumor target cells *in vitro* were significantly ($p \leq 0.05$) increased in both sexes at the low dose only. The increase in natural killer cytotoxic responses is not considered an adverse effect. There were no effects of 2-butoxyethanol treatment on splenocyteproduction of interleukin 2 and interferon, and thymus and spleen weights. No histological lesions in the thymus were observed.

Several acute oral studies have examined the effects of 2-butoxyethanol on lymphoreticular organs. In a preliminary teratology study, pregnant Fischer 344 rats treated by gavage with ≥150 mg/kg/day 2-butoxy-ethanol on gestational days 9-11 or 11-13 exhibited increased absolute spleen weight at all doses compared with controls (NTP 1989). In the definitive study, in which the doses were 30, 100, or 200 mg/kg/day on gestational days 9-11 and 30, 100, and 300 mg/kg/day on gestational days 11-13, increased absolute spleen weight was noted at a dose level of ≥100 mg/kg/day in both experiments (NTP 1989).

Young (4-5 weeks), adult (9-13 weeks) and older (5-16 months) male Fischer 344 rats were dosed with 0, 32, 63, 125, 250, or 500 mg/kg 2-butoxyethanol by gavage (Ghanayem et al. 1987a). Young and adult rats receiving ≥125 mg/kg exhibited a dose-related increase in relative spleen weight; at 125 mg/kg the increase

relative to control was 110% for young rats and 150% for adult rats. Spleen weights were not recorded for the older rats. Ghanayem and coworkers have published other results indicating similar effects in male Fischer 344 rats after a single dose of 125 or 500 mg/kg 2-butoxyethanol by gavage (Ghanayem et al. 1987b). Repeated dosing with 2-butoxyethanol seems to lessen the effect on the spleen weight increase. Male Fischer 344 rats received 0 or 125 mg/kg/day 2-butoxyethanol in water by gavage for 1, 2, 3, 6, or 12 consecutive days (Ghanayem et al. 1992). Treated animals showed a time-dependent increase in spleen weight which reached a maximum of 62% in 6 days and declined to 45% between days 6 and 12. In the same study, using a different study design, male Fischer 344 rats received 125 mg/kg/day 2-butoxyethanol in water by gavage for 3 consecutive days. Controls received 5 mL water/kg body weight daily for 3 consecutive days. Rats in both groups were then allowed to recover without treatment for 7 days. These rats were then subgrouped with control and pretreated animals in each subgroup, and treated with water, 125 mg/kg, or 250 mg/kg 2-butoxyethanol. Twenty-four hours after dosing, the spleen was removed and weighed. Pretreatment with 2-butoxyethanol resulted in a smaller increase in spleen weight than was observed in the rats that were not pretreated. Male Fischer 344 rats given 2-butoxyethanol orally for 4 consecutive days at 500 or 1,000 mg/kg/day exhibited increased spleen weight and extramedullary hematopoiesis (Grant et al. 1985).

No effects on absolute or relative thymus weight were observed in Fischer 344 rats given 2-butoxyethanol at ≤346 mg/kg/day (males) and ≤265 mg/kg/day (females) in the drinking water for 2 weeks (NTP 1993). However, male $B6C3F_1$ mice exhibited a 38% decrease in absolute thymus weight and a 39% decrease in relative thymus weight after treatment with 370 mg/k/day, and a 20% decrease in absolute thymus weight and a 23% decrease in relative thymus weight at 627 mg/kg/day 2-butoxyethanol in the drinking water for 2 weeks, but females dosed at ≤1,364 mg/kg/day showed no effect on thymus weight (NTP 1993).

Effects on lymphoreticular organs have also been examined in intermediate-duration oral studies. Increased spleen weights were found at ≥443 mg/kg/day in male CQBS CD (SD)BR rats treated by gavage with 2-butoxyethanol for 6 weeks (Eastman Kodak 1983; Krasavage 1986). Enlarged dark spleens were observed in three of nine rats exposed at 443 mg/kg/day and in four of eight rats at 885 mg/kg/day. Splenic congestion secondary to the hematological effects was seen at all doses (≥ 222 mg/kg/day). Histological examination of the thymus, mesenteric lymph nodes, and bone matrow revealed no pathological lesions, and no effect on white blood cells was noted. In Fischer 344 rats exposed to 2-butoxyethanol in drinking water for 13 weeks, bone marrow hyperplasia was found in males at ≥281 mg/kg/day and in females at ≥ 363 mg/kg/day; hematopoietic cell proliferation and congestion in the spleen were found in males at ≥ 367 mg/kg/day and in

females at ≥363 mg/kg/day; increased hemosiderin pigmentation in the spleen was found in males at ≥129 mg/kg/day and in females at ≥ 151 mg/kg/day (NTP 1993). The absolute thymus weight was decreased in males at ≥367 mg/kg/day and in females at 470 mg/kg/day. No histopathological lesions were found in the thymus or lymph nodes. In contrast to rats, $B6C3F_1$ mice receiving ≤ 694 mg/kg/day (males) and ≤ 1,306 mg/kg/day (females) for 13 weeks in the drinking water showed no effect on thymus weight and no gross or histopathological lesion in the spleen, thymus, lymph nodes, or bone marrow (NTP 1993). No effect on thymus or spleen weight, and no histopathological lesions in the thymus were observed in rats given <506 mg/kg/day for 21 days (Exon et al. 1991).

In a study on 2-butoxyethanol acetate, Wistar rats given unspecified acute oral doses of 2-butoxyethanol acetate in olive oil as part of a lethality study exhibited no bistopathological changes in the spleen (Truhaut et al. 1979).

Spleen weight effects, spleen congestion, spleen and bone marrow hematopoiesis, and hemosiderin pigmentation in the spleen are probably related to hemolytic effects of 2-butoxyethanol, which are discussed in Section 2.2.2.2 under Hematological Effects

The highest NOAEL values and all LOAEL values from each reliable study for immunological and lymphoreticular effects in each species and duration category are recorded in Table 2-3 and plotted in Figure 2-3.

2.2.2.4 Neurological Effects

No studies were located regarding neurological effects in humans after oral exposure to 2-butoxyethanol acetate or 2-butoxyacetic acid or in animals after oral exposure to 2-butoxyacetic acid.

Intentional poisoning with 2-butoxyethanol has been associated with neurological effects. A woman who died of cardiac arrest 3 days after she ingested an unspecified amount of cleaner containing 6.5% 2-butoxyethanol was comatose upon admission to the hospital (Litovitz et al. 199 1). A male patient, who was a known alcohol and trichloroethylene abuser, was comatose upon admission to the hospital after ingesting a household cleaning fluid that provided a dose of about 650 mg/kg 2-butoxyethanol (Bauer et al. 1992). Similarly, a woman who ingested 467-933 mg/kg 2-butoxyethanol (also in window cleaner) was in a coma with no response to painful stimuli (Rambourg-Schepens et al. 1988), and another woman who ingested

391-469 mg/kg 2-butoxyethanol became comatose (Gijsenbergh et al. 1989). All three patients recovered after supportive therapy. In contrast, no signs of neurological effects were seen in two children who accidentally ingested 270 or 1,862 mg/kg 2-butoxyethanol and who were observed for 24 hours after gastric lavage (Dean and Krenzelok 1992).

Clinical signs of neurotoxicity have also been observed in animals after acute oral exposure to 2-butoxyethanol. Rats that died after a single gavage dose of ≥ 530 mg/kg exhibited sluggishness, prostration, and narcosis (Carpenter et al. 1956). Other investigators have reported drowsiness, sluggishness, lethargy, muscular flaccidity, and/or ataxia after acute exposure of rats to 252-9,000 mg/kg/day (Dow 1959, 198 1; Eastman Kodak 1983; Krasavage 1986; Olin 1976; Sivarao and Mehendale 1995; Union Carbide 1980b). Moderate-to-severe weakness and prostration were observed directly after dosing in guinea pigs given a single gavage dose of 1,000 mg/kg 2-butoxyethanol, a dose that resulted in the death of 1 of 5 males and 1 of 5 females (Shepard 1994b). Slight weakness was observed directly after dosing at 500 mg/kg, a dose at which all treated guinea pigs survived. Pregnant Fischer 344 rats exhibited lethargy after oral dosing in a preliminary teratology study with 600 mg/kg/day 2-butoxyethanol given by gavage on gestational days 9-11 or 11-13 (NTP 1989). Ii-r the definitive study, no adverse neurological clinical signs were noted at ≤200 mg/kg/day on gestational days 9-11 or ≤300 mg/kg/day on gestational days 11-13 (NTP 1989). Pregnant CD-1 mice given doses of 1,500 or 2,000 mg/kg/day 2-butoxyethanol by gavage on gestational days 8-14 exhibited lethargy and failure to right when turned over, but no effect on demeanor was seen at ≤ 1,000 mg/kg/day (Wier et al. 1987).

Adult male COBS CD (SD)BR rats were given undiluted 2-butoxyethanol by gavage in doses of 0, 222, 443, or 885 mg/kg/day, 5 days per week over a 6-week period, and exhibited lethargy at ≥443 mg/kg/day after the first dose (Eastman Kodak 1983; Krasavage 1986). The high-dose rats were slightly weak and inactive after the second and third doses. No effect on absolute brain weight was observed, but relative brain weight was increased at ≥443 mg/kg/day. No histopathological changes were seen in brain tissue at any dose. Fischer 344 rats that received 2-butoxyethanol at ≤452 mg/kg/day (males) and ≤470 mg/kg/day (females) and $B6C3F_1$ mice that received ≤ 694 mg/kg/day (males) and ≤ 1,306 mg/kg/day (females) in the drinking water for 13 weeks showed no adverse effects on demeanor or histopathological changes in brain, spinal cord, or sciatic nerve (NTP 1993).

Wistar rats given unspecified acute oral doses of 2-butoxyethanol acetate in olive oil as part of a lethality study exhibited no adverse neurological clinical signs or histopathological changes in the brain (Truhaut et al. 1979).

The highest NOAEL values and all LOAEL values from each reliable study for neurological effects in each species and duration category are recorded in Table 2-3 and plotted in Figure 2-3.

2.2.2.5 Reproductive Effects

No studies were located regarding reproductive effects in humans after oral exposure to 2-butoxyethanol, 2-butoxyethanol acetate, or 2-butoxyacetic acid.

In acute studies, no effects on testicular weight and no bistopathological effects in the testes were found in male Fischer 344 rats given 2-butoxyethanol orally once at ≤ 500 mg/kg (Ghanayem et al. 1987a) or for 4 consecutive days at 500 or 1,000 mg/kg/day (Grant et al. 1985). Likewise, no effects on testicular weight and no histopathological lesions in the testes or epididymides were found in Fischer 344 rats at ≤ 346 mg/kg/day or in B6C3F_1 mice at ≤ 627 mg/kg/day 2-butoxyethanol in drinking water for 2 weeks (NTP 1993).

In intermediate-duration studies, no effects on testicular weight and no histopathological lesions in the testes were found in Sprague-Dawley rats at ≤ 506 mg/kg/day 2-butoxyethanol for 21 days (Exon et al. 199 1). No effects on testicular weight and no histopathological lesions in the testes, seminal vesicles, epididymides, or prostate were found in COBS CD (SD)BR rats treated by gavage at ≤885 mg/kg/day for 6 weeks (Eastman Kodak 1983; Krasavage 1986). No effects on testicular weight and no histopathological lesions in the testes or epididymides (or seminal vesicles in the 13-week study) were found in Fischer 344 rats given ≤443 mg/kg/day in the drinking water for 60 days or ≤452 mg/kg/day in the drinking water for 13 weeks (NTP 1993). Although spermatozoa concentrations (10^6/g caudal epididymal tissue) were significantly ($p \leq 0.01$) decreased compared to controls at ≥281 mg/kg/day, a dose-related effect was not observed (control, 713.9±16.2; 281 mg/kg/day, 633.0±13.1; 367 mg/kg/day, 656.3±13.3; 452 mg/kg/day, 617.2 ±22.9), and there were no significant effects on spermatid heads (10^7/g tests or 10^7/testis), spermatid counts (mean/10^{-4} mL), or percent mobile spermatozoa. A small, older study (Weil and Carpenter 1963) did find histological evidence of testicular atrophy after 93 days exposure to 188 mg/kg/day in DW albino rats.

No effects on the weights of testes/seminal vesicles and coagulating gland and no histopathological lesions in these tissues were found in JCL-ICR mice treated by gavage at 1,000 mg/kg/day for 5 weeks (Nagano et al. 1979, 1984). No significant effects on the weights of testes, seminal vesicles, or prostate and no effects on sperm parameters were found in Swiss mice in a continuous breeding drinking water study at 1,300 mg/kg/day or in the male offspring at 700 mg/kg/day (Heindel et al. 1990). Slight but statistically significant ($p \leq 0.0l$) decreases in left testis, but not right testis, weights were observed in $B6C3F_1$ mice at ≥210 mg/kg/day in drinking water for 13 weeks (NTP 1993). Because the effect on left testis weight was not dose-related (and there were no effects on right testis weight), it was not considered biologically significant. In addition, no histopathological changes were observed in the seminal vesicles or testes/epididymides, nor were there any consistent treatment-related effects on sperm parameters in mice at ≤627 mg/kg/day in drinking water for 13 weeks (NTP 1993).

Female Fischer 344 rats treated with 2-butoxyethanol in the drinking water for 13 weeks exhibited altered estrous cycles at ≥ 363 mg/kg/day, with more time spent in diestrus than in the other phases (NTP 1993). The total duration of the estrous cycle was not affected. No histopathological lesions were found in the mammary glands, ovaries, or uterus. No effect on estrous cycles and no histopathological lesions in the mammary glands, ovaries, or uterus were found in female $B6C3F_1$ mice similarly treated for 13 weeks at ≤ 1,306 mg/kg/day (NTP 1993).

Pregnant Fischer 344 rats exhibited vaginal bleeding after oral dosing by gavage in a preliminary teratology study with 600 mg/kg/day 2-butoxyethanol on gestational days 9-11 or 11-13 (NTP 1989). In the definitive study, increased resorptions, implantation loss, and vaginal bleeding were noted at the high doses of 200 mg/kg/day on gestational days 9-11 and 300 mg/kg/day on gestational days 11-13 (NTP 1989). These doses were also maternally toxic. A decrease in the incidence of viable litters was found in pregnant CD- 1 mice treated with 1,180 mg/kg/day by gavage on gestational days 6-13, a dose that resulted in decreased maternal body weight gain (Hardin et al. 1987; Schuler et al. 1984). Pregnant female CD-l mice given 350-2,000 mg/kg/day 2-butoxyethanol on gestational days 8-14 exhibited a green or red vaginal discharge during gestation at doses of 1,500 and 2,000 mg/kg/day; surviving animals in the 1,000- and -1,500-mg/kg/day dose groups had an increased incidence of resorptions (Wier et al. 1987).

Mating pairs of Swiss mice exposed to 1,300 or 2,100 mg/kg/day 2-butoxyethanol in the drinking water for 21 weeks exhibited a decrease in the number of litters produced per pair, and in the size of each litter, with decreased pup weight. However, the low dose, 700 mg/kg./day, produced no adverse effect on fertility

(Heindel et al. 1990). When the females that had been dosed with 1,300 mg/kg/day 2-butoxyethanol were mated with control males, and then maintained on treatment throughout gestation to delivery, they produced 58% fewer litters, and the litters were 66% smaller compared to control mating pairs. When the offspring of the low-dose mating pairs (700 mg/kg/day) were mated, there was no adverse effect on fertility. A few females in the treated group (1,300mg/kg/day) had long or unclear estrous cycles, but the significance of this is not clear (Heindel et al. 1990).

Wistar rats given unspecified acute oral doses of 2-butoxyethanol acetate in olive oil as part of a lethality study exhibited no adverse histopathological changes in the testes or ovaries (Truhaut et al. 1979).

Groups of male Alpk/AP (Wistar-derived) rats were dosed once by gavage with 0, 174, 434, or 868 mg/kg 2-butoxyacetic acid (Foster et al. 1987). Occasional significant decreases in the weight of the prostate and seminal vesicles were observed, but the decreases were not time or dose related. Histological examination of testes, epididymides, and prostate revealed no treatment-related lesions.

The highest NOAEL values and all LOAEL values from each reliable study for reproductive effects in each species and duration category are recorded in Tables 2-3 and 2-5 and plotted in Figures 2-3 and 2-5.

2.2.2.6 Developmental Effects

No studies were located regarding developmental effects in humans after oral exposure to 2-butoxyethanol, 2-butoxyethanol acetate, or 2-butoxyacetic acid. No studies were located regarding developmental effects in animals after oral exposure to 2-butoxyethanol acetate or 2-butoxyacetic acid.

The offspring of pregnant Fischer 344 rats exhibited decreased fetal body weight after oral dosing of the dams in a preliminary teratology study with 300 and 600 mg/kg/day 2-butoxyethanol on gestational days 9-11 (NTP 1989). Decreased gravid uterine weight and decreased fetal body weight were found at 600 mg/kg/day after dams were dosed on gestational days 11-13. However, no external or intera1 malformations were observed in the fetuses after the dams were treated with ≤ 600 mg/kg/day on gestational days 9-11 or 11-13. The doses that resulted in fetal effects also caused maternal toxicity. In the definitive study, no adverse developmental effects (fetal weight or external and visceral malformations) were noted at ≤ 200 mg/kg/day on gestational days 9-11 or ≤ 300 mg/kg/day on gestational days 11-13 (NTP 1989). CD-1 mice were treated by gavage with 1 ,180 mg/kg/day 2-butoxyethanol on gestational days 6-l 3 and allowed to

litter (Hardin et al. 1987; Schuler et al. 1984). As noted in Section 2.2.2.5 above, treated females had significantly reduced numbers of viable litters, but no adverse effects were observed in the pups that were born alive. However, the offspring of pregnant female CD-1 mice given 1,000 or 1,500 mg/kg/day 2-butoxyethanol on gestational days 8-14 exhibited evidence of cleft palate when evaluated at term (Wier et al. 1987). The response was not clearly dose related; one of five litters (4/43 fetuses) and one of three litters (l/25 fetuses) were affected at 1,000 and 1,500 mg/kg/day, respectively. The 1,000-mg/kg/day dose was associated with green-brown or red-brown staining of the cage papers, while at 1,500 mg/kg/day three of six mice died. In a postnatal study designed to examine pup survival beyond the neonatal period, cleft palate effect was not evident when mice dosed with 650 or 1,000 mg/kg/day (Wier et al. 1987). In a reproductive toxicity study, male and female Swiss mice were given doses of 700, 1,300, or 2,100 mg/kg/day in drinking water for 21 weeks (Heindel et al. 1990). Decreased pup weight was observed for all treated mating pairs at ≥700 mg/kg/day. A crossover mating trial (performed to assess which sex had been affected by treatment, or which sex is more affected) indicated that the reproductive effects could be attributed primarily to an effect on the female, although no developmental effects were observed in the offspring that were produced (Heindel et al. 1990). The crossover mating trial was performed after the last litter from the continuous breeding phase was weaned to determine the affected sex. The crossover mating trial consisted of 3 groups of 20 pairs each -- control males x control females, control maIes x high dose females, and control females x high dose males. Pairs were matched for 7 days or until a copulatory plug is detected, whichever is first. When the offspring of the 700-mg/kg/day animals were mated while being maintained on treatment from weaning throughout mating, gestation, and delivery (14 weeks), no developmental effects were observed in the pups (Heindel et al. 1990).

The highest NOAEL values and all LOAEL values from each reliable study for developmental effects in each species and duration category are recorded in Table 2-3 and plotted in Figure 2-3.

2.2.2.7 Genotoxic Effects

No studies were located regarding genotoxic effects in humans or animals after oral exposure to 2-butoxyethanol, 2-butoxyethanol acetate, or 2-butoxyacetic acid. *In vitro* genotoxicity studies are discussed in Section 2.5.

2.2.2.8 Cancer

No studies were located regarding carcinogenic effects in humans or animals after oral exposure to 2-butoxyethanol, 2-butoxyethanol acetate, or 2-butoxyacetic acid.

2.2.3 Dermal Exposure

Occupational or environmental exposure to 2butoxyethanol or 2-butoxyethanol acetate usually occurs through the inhalation or dermal route. Both routes of exposure are important in the industrial setting. Occupational exposure to 2-butoxyethanol usually involves co-exposure to other solvents and chemicals. In several NIQSH Health Hazard Evaluations, effects reported by workers included eye, nose, and throat irritation; coughing; runny nose; headache; dizziness; lightheadedness; and nausea (Apoll98 1,1986; Lee 1988). Since personal breathing zone and workplace air samples analyzed for solvents and other chemicals such as, toluene, xylene, methyl ethyl ketone, methyl isobutyl ketone, styrene, along with 2-butoxyethanol, indicated that exposure levels for each chemical were below the NIOSH, ACGIH, and OSHA criteria, NIOSH concluded that the effects were probably due to the additive combination of the solvents. Experimental studies in humans exposed by inhalation are discussed in Section 2.2.1 and have been summarized in several publications (Browning and Curry 1994; EPA 1984; NIOSH 1990; Tyler 1984). Experimental studies in humans and animals exposed dermally to 2-butoxyethanol or 2-butoxyethanol acetate are discussed below. No studies were located describing the effects of dermal exposure to 2-butoxyacetic acid in humans or animals.

2.2.3.1 Death

No studies were located regarding death in humans after dermal exposure to 2-butoxyethanol or 2-butoxyethanol acetate.

A dermal LD_{50} of 2,273 mg/kg has been reported for an unspecified strain and number of rats (Carpenter and Condra 1961). The compound was placed on the clipped skin of the trunk, and the skin was occluded for 4 hours. Several investigators have reported acute dermal LD_{50} values for rabbits, which generally ranged from 406 to 1,804 mg/kg (Carpenter et al. 1956; Eastman Kodak 1988; Olin 1976; Union Carbide 1980a, 1980b). In one determination, female New Zealand rabbits appeared to be more sensitive than male New Zealand rabbits to the lethality of 2-butoxyethanol, with LD_{50} values reported to be 568 mg/kg for females

and 638 mg/kg for males (Union Carbide 1980a). However, one study reported an acute dermal LD_{50} value of 220 mg/kg for an unspecified strain and sex of rabbits, in which four of four rabbits treated with 252 mg/kg for 24 hours died within 2-7 days (Dow 1959). Another study reported an acute dermal LD_{50} (8-hour) as low as 99 mg/kg for female New Zealand rabbits (Duprat and Gradiski 1979). In this study, groups of six female rabbits were exposed dermally (with the use of vials) to several dose levels for 8 hours and observed for 15 days. Mortality incidences were 0 of 6 in controls, 2 of 6 at 72 mg/kg, 2 of 6 at 90 mg/kg, 4 of 6 at 108 mg/kg, 5 of 6 at both 135 and 180 mg/kg, and 6 of 6 at 225 mg/kg. Deaths occurred on days l-8 after treatment; early deaths were attributed to narcosis or failure of respiration or cardiac function, while delayed deaths were observed after weight loss and were probably due to renal impairment.

In acute dermal studies by Carpenter et al. (1956), extreme congestion of the kidney, hemoglobinuria, pale liver, and engorged spleen were noted in the rabbits that died. Deaths occurred at dose levels of 403-l ,804 mg/kg for undiluted 2-butoxyethanol. In New Zealand rabbits of unspecified sex to which 250, 500, 1,000 or 2,000 mg/kg 2-butoxyethanol was applied to clipped, abraded skin and occluded for 24 hours, 1 of 4 died at 500 mg/kg on day 12, and 10 of 10 died at 1,000 and 2,000 mg/kg on days l-2 (Olin 1976). At 24 hours, rabbits exposed to 1,000 mg/kg/day exhibited flaccid muscle tone and hematuria, whereas rabbits exposed to 2,000 mg/kg/day exhibited lacrimation, hematuria, flaccid muscle tone, and anorexia (Olin 1976). No treatment-related deaths occurred in male or female New Zealand rabbits to which doses of ≤ 361 mg/kg/day were applied to the clipped, occluded skin for 6 hours per day for nine applications over 11 days (Union Carbide 1980a).

In a developmental study, application of 2-butoxyethanol four times per day at 0.35 mL per application (1.4 mL/day) to the shaved interscapular skin of pregnant Sprague-Dawley rats on gestational days 7-16 was lethal in 10 of 11 rats on treatment days 3-7 (Hardin et al. 1984). Pregnant rats were subsequently tested at 0.12 mL per treatment (0.48 mL/day), which caused no mortality. No deaths were observed in guinea pigs in which undiluted 2-butoxyethanol(2,000 mg/kg) was placed on the dorsal skin that had been clipped free of hair (Shepard 1994a). The treated area was covered with an occlusive wrap for 24 hours. In guinea pigs exposed epicutaneously to 0.5 or 2.0 mL 2-butoxyethanol and observed for 35 days, 13 of 20 guinea pigs in the high-dose group died within the 1 st week, but none died thereafter (Wahlberg and Boman 1979). No animals died in the low-dose group.

In an intermediate-duration study, no treatment-related deaths were found in male and female New Zealand rabbits exposed dermally to 2-butoxyethanol at 0, 10, 50, or 150 mg/kg/day, 6 hours per day, 5 days per week for 90 days (CMA 1983).

In an acute dermal LD_{50} determination for 2-butoxyethanol acetate, doses of 3,191, 3,957, 4,766, 5,957, or 10,000 mg/kg were applied to the clipped, occluded skin of New Zealand rabbits for 24 hours (Truhaut et al. 1979). The rabbits were kept under observation for 14 days after exposure. The applied doses of 3,191, 2,957, 4,766, 5,957, and 10,000 mg/kg were reported to correspond to absorbed doses of 610, 910, 1,130, 1,830, and 2,200 mg/kg, respectively. The LD_{50} for rabbits was reported to be approximately 1,500 mg/kg of the absorbed dose, which would correspond to an applied dose of between 4,766 and 5,957 mg/kg. The rabbits generally died between 24 and 48 hours after application and no later than 4 days after exposure.

The LD_{50} values for rabbits and all LOAEL values from each reliable study for death in each species duration category are recorded in Tables 2-6 and 2-7.

2.2.3.2 Systemic Effects

No studies were located regarding respiratory, cardiovascular, gastrointestinal, hematological, musculoskeletal, hepatic, renal, endocrine, ocular, or body weight effects in humans. No studies were located describing respiratory, cardiovascular, gastrointestinal, hematological, musculoskeletal, hepatic, renal, ocular, or body weight effects in humans or gastrointestinal, musculoskeletal, or body weight effects in animals following dermal exposure to 2-butoxyethanol acetate. Available data pertaining to systemic effects of both compounds are presented below.

The highest NOAEL values and all LOAEL values from each reliable study for systemic effects in each species and duration category are recorded in Tables 2-6 and 2-7.

Respiratory Effects. Female New Zealand outbred rabbits were epicutaneously exposed to 2-butoxyethanol (Duprat and Gradiski 1979). Several doses of 72-225 mg/kg of the undiluted substance (analytical grade, purity >99.5%) were directly applied over 8 hours onto the clipped back (using vials) of the unrestrained rabbits. Animals were observed daily over a 15-day period. Macroscopic and microscopic examinations were performed on dead animals (death was observed in all dose groups) and those surviving on the 15th day. Respiratory irritation appeared as congestion and thickening of alveolar walls, similar to

Table 2-6. Levels of Significant Exposure to 2-Butoxyethanol - Dermal

Species/ (Strain)	Exposure/ Duration/ Frequency/ (Specific Route)	System	NOAEL	LOAEL			Reference
				Less Serious	Serious		
ACUTE EXPOSURE							
Death							
Rat (NS)	4 hr				2273 mg/kg	F (LD_{50})	Carpenter and Condra 1961
Rat (Sprague-Dawley)	Gd 7-16 4x/d				1.4 mL/d	F (death in 10/11)	Hardin et al. 1984
Gn Pig (NS)	once				2.0 mL	(13/20 died within 1 week)	Wahlberg and Boman 1979
Rabbit (New Zealand)	24 hr				406-1804 mg/kg	M (LD_{50})	Carpenter et al. 1956
Rabbit (NS)	24 h				220 mg/kg	(LD_{50})	Dow 1959
Rabbit (New Zealand)	8 hr				72 mg/kg	F (death in 2/6 on day 5)	Duprat and Gradiski 1979
					99 mg/kg	F (LD_{50})	
Rabbit (NS)	NS				435 mg/kg	(LD_{50})	Eastman Kodak 1988
Rabbit (New Zealand)	24 hr				2000 mg/kg	(10/10 animals died)	Olin 1976
Rabbit (New Zealand)	24 hr				580 mg/kg	(LD_{50})	Olin 1976

Table 2-6. Levels of Significant Exposure to 2-Butoxyethanol - Dermal (continued)

Species/ (Strain)	Exposure/ Duration/ Frequency/ (Specific Route)	System	NOAEL	LOAEL		Reference
				Less Serious	Serious	
Rabbit (New Zealand)	once 6 hr				638 M (LD_{50}) mg/kg	Union Carbide 1980a
					568 F (LD_{50}) mg/kg	
Systemic						
Human	4-8 hr	Ocular	98 ppm	113 ppm (ocular irritation)		Carpenter et al. 1956
Human	24-72 hr	Dermal	0.2 mL 10%			CMA 1992; Greenspan et al. 1995
Human	2 hr	Dermal		100% M (drying of the skin, reduction in skinfold thickness and volume of exposed fingers)		Johanson et al. 1988
Rat (Wistar)	once	Hemato	200 F mg/kg		260 F mg/kg (hemolysis, hemoglobinuria)	Bartnik et al. 1987
Rat (Sprague-Dawley)	Gd 7-16 4x/d	Hemato			1.4 mL/d F (burgundy-colored urine [hemoglobinuria])	Hardin et al. 1984
		Dermal		1.4 mL/d F (necrosis of tail; rough coat)		
Rat (Sprague-Dawley)	Gd 7-16 4x/d	Bd Wt		0.48 F mL/d (16% decrease in body weight gain days 5-12; 13% decrease in body weight gain days 5-17)		Hardin et al. 1984

Table 2-6. Levels of Significant Exposure to 2-Butoxyethanol - Dermal (continued)

Species/ (Strain)	Exposure/ Duration/ Frequency/ (Specific Route)	System	NOAEL	LOAEL		Reference
				Less Serious	Serious	
Rat (Fischer- 344)	Gd 6-15 6 hr/d	Ocular		25 ppm F (periocular wetness)		Tyl et al. 1984
Gn Pig (NS)	once	Bd Wt	0.5 mL			Wahlberg and Boman 1979
Rabbit (New Zealand)	24 hr	Hemato			406 mg/kg M (hemoglobinuria)	Carpenter et al. 1956
		Hepatic			406 mg/kg M (pale liver)	
		Renal			406 mg/kg M (congested kidneys)	
Rabbit (New Zealand)	3 min	Hemato		505 mg/kg M (increased erythrocyte osmotic fragility)		Carpenter et al. 1956
Rabbit (NS)	24 h	Dermal		200 mg/kg (moderate skin irritation)		Dow 1959
		Ocular			100% (moderate conjunctival irritation; corneal injury)	
		Bd Wt		200 mg/kg (slight initial weight loss)		

Table 2-6. Levels of Significant Exposure to 2-Butoxyethanol - Dermal (continued)

Species/ (Strain)	Exposure/ Duration/ Frequency/ (Specific Route)	System	NOAEL	LOAEL				Reference
				Less Serious		Serious		
Rabbit (New Zealand)	8 hr	Resp		72 mg/kg F	(congestion, thickening of alveolar walls)			Duprat and Gradiski 1979
		Hemato				72 mg/kg F	(hemoglobinuria)	
		Hepatic				72 mg/kg F	(congestion in liver, necrosis and steatosis)	
		Renal				72 mg/kg F	(enlarged kidneys with hemoglobinuric nephrosis and interstitial reaction)	
		Dermal		72 mg/kg F	(necrosis of epidermis and dermis)			
		Other				72 mg/kg F	(hypothermia)	
Rabbit (New Zealand)	once	Ocular		0.1 mL of 10%	(mild eye irritation)	0.1 mL of 100%	(severe eye irritation)	Kennah et al. 1989a
Rabbit (New Zealand)	24 hr	Gastro	500 mg/kg			1000 mg/kg	(very dark areas in small intestine)	Olin 1976
		Musc/ske	500 mg/kg			1000 mg/kg	(flaccid muscle tone)	
		Hepatic				250 mg/kg	(discolored liver)	
		Renal	250 mg/kg			500 mg/kg	(blood in urine and bladder [hematuria], discolored kidney)	
		Ocular	250 mg/kg			500 mg/kg	(yellow cornea)	
Rabbit (New Zealand)	once or 4 hr	Dermal		0.5 mL	(moderate skin irritation)			Rohm and Haas 1983
		Ocular				0.1 mL	(severe eye irritation)	

Table 2-6. Levels of Significant Exposure to 2-Butoxyethanol - Dermal (continued)

Species/ (Strain)	Exposure/ Duration/ Frequency/ (Specific Route)	System	NOAEL	LOAEL			Reference
				Less Serious		Serious	
Rabbit (New Zealand)	Gd 6-18 6 hr/d	Ocular	50 ppm F	100 ppm F	(periocular wetness)		Tyl et al. 1984
Rabbit (New Zealand)	11 d 9 x 1 mL/d 6 hr/d	Hemato	180 M mg/kg/d	361 M mg/kg/d	(transient hemoglobinuria)		Union Carbide 1980a
			180 F mg/kg/d	361 F mg/kg/d	(reduced mean erythrocyte counts, hemoglobin, MCHC; increased MCH)		
		Hepatic	361 mg/kg/d				
		Renal	90 F mg/kg/d			180 F mg/kg/d (blood in urine [hematuria])	
		Dermal		18 mg/kg/d	(erythema)		
		Ocular	361 M mg/kg/d				
		Bd Wt	361 M mg/kg/d				
			180 F mg/kg/d			361 F mg/kg/d (121% decrease in weight gain on day 3)	

Table 2-6. Levels of Significant Exposure to 2-Butoxyethanol - Dermal (continued)

Species/ (Strain)	Exposure/ Duration/ Frequency/ (Specific Route)	System	NOAEL	LOAEL				Reference
				Less Serious		Serious		
Rabbit (New Zealand)	once 6 hr	Gastro	451 mg/kg	902 mg/kg	(reddened stomach; females: reddened intestines)			Union Carbide 1980a
		Hemato				451 mg/kg F	(hemoglobinuria)	
		Hepatic	451 mg/kg	902 mg/kg	(mottled with pocked surface)			
		Renal				451 mg/kg	(hematuria; enlarged, dark kidneys; male survivors had pocked surface)	
		Endocr		451 mg/kg	(reddened adrenals)			
		Dermal		451 mg/kg	(erythema; females: slight necrosis at application site)			
		Ocular	451 M mg/kg 902 F mg/kg			902 M mg/kg	(grey iris)	
		Other	451 mg/kg	902 mg/kg	(yellowed peritoneal fat)			
Rabbit (New Zealand)	11 d 9 x 1 mL/d 6 hr/d	Renal		271 mg/kg/d	(tubular vacuolization [4/6] degeneration [6/6] hyperplasia [3/6], glomerular adhesions [4/6], interstitial nephritis [3/6], hemoglobinuric nephrosis)			Union Carbide 1980a
		Dermal		271 mg/kg/d	(necrosis)			

Table 2-6. Levels of Significant Exposure to 2-Butoxyethanol - Dermal (continued)

Species/ (Strain)	Exposure/ Duration/ Frequency/ (Specific Route)	System	NOAEL	LOAEL		Reference
				Less Serious	Serious	
Rabbit (New Zealand)	24 hr	Resp			451 M mg/kg (orange-red lungs)	Union Carbide 1980b
		Gastro			451 M mg/kg (orange peritonea, intestines)	
		Hepatic			451 M mg/kg (orange-red liver)	
		Renal			451 M mg/kg (dark red kidneys, hematuria)	
		Dermal	451 M mg/kg	902 M mg/kg (erythema, necrosis)		
		Ocular	451 M mg/kg		902 M mg/kg (iritis in 2/4)	
Rabbit (NS)	once	Ocular	0.5 mL 5%	0.5 mL 15% (moderate corneal injury)		Union Carbide 1980b
Rabbit (New Zealand)	4 hr	Dermal		0.5 mL (irritant)		Zissu 1995
Rabbit (New Zealand)	24 hr	Dermal			0.5 mL (severe irritant)	Zissu 1995
Immunological/Lymphoreticular						
Human	24-72 hr		0.2 mL 10%			CMA 1992; Greenspan et al. 1995
Rabbit (New Zealand)	24 hr				406 M mg/kg (engorged spleen)	Carpenter et al. 1956

Table 2-6. Levels of Significant Exposure to 2-Butoxyethanol - Dermal (continued)

Species/ (Strain)	Exposure/ Duration/ Frequency/ (Specific Route)	System	NOAEL	LOAEL			Reference
				Less Serious	Serious		
Rabbit (New Zealand)	8 hr				72 mg/kg F	(enlarged spleens filled with erythrocytes, white atrophic pulp)	Duprat and Gradiski 1979
Rabbit (New Zealand)	once 6 hr		902 M mg/kg				Union Carbide 1980a
			451 F mg/kg	902 F mg/kg	(enlarged spleen)		
Rabbit (New Zealand)	once				451 M mg/kg	(dark spleens)	Union Carbide 1980b
Neurological							
Rat (Sprague-Dawley)	Gd 7-16 4x/d				1.4 mL/d F	(ataxia; moderate to marked inactivity)	Hardin et al. 1984
Rabbit (New Zealand)	8 hr				72 mg/kg F	(prostration, narcosis prior to death)	Duprat and Gradiski 1979
Rabbit (New Zealand)	24 hr		1000 mg/kg		2000 mg/kg	(anorexia, no spontaneous movement)	Olin 1976
Rabbit (New Zealand)	once 6 hr		902 M mg/kg				Union Carbide 1980a
			451 F mg/kg		902 F mg/kg	(nystagmus, convulsions)	

Table 2-6. Levels of Significant Exposure to 2-Butoxyethanol - Dermal (continued)

Species/ (Strain)	Exposure/ Duration/ Frequency/ (Specific Route)	System	NOAEL	LOAEL		Reference
				Less Serious	Serious	
Developmental						
Rat (Sprague-Dawley)	Gd 7-16 4x/d		0.48 mL/d			Hardin et al. 1984
INTERMEDIATE EXPOSURE						
Systemic						
Rabbit (New Zealand)	90 d 5 d/wk 6 hr/d	Resp	150 mg/kg			CMA 1983
		Cardio	150 mg/kg			
		Gastro	150 mg/kg			
		Hemato	150 mg/kg			
		Musc/ske	150 mg/kg			
		Hepatic	150 mg/kg			
		Renal	150 mg/kg			
		Endocr	150 mg/kg			
		Dermal		10 mg/kg	(slight to moderate erythema and edema)	
		Ocular	150 mg/kg			
		Bd Wt	150 mg/kg			

Table 2-6. Levels of Significant Exposure to 2-Butoxyethanol - Dermal (continued)

Species/ (Strain)	Exposure/ Duration/ Frequency/ (Specific Route)	System	NOAEL	LOAEL		Reference
				Less Serious	Serious	
Immunological/Lymphoreticular						
Rabbit (New Zealand)	90 d 5 d/wk 6 hr/d		150 mg/kg			CMA 1983
Neurological						
Rabbit (New Zealand)	90 d 5 d/wk 6 hr/d		150 mg/kg			CMA 1983
Reproductive						
Rabbit (New Zealand)	90 d 5 d/wk 6 hr/d		50 mg/kg M 150 F mg/kg	150 M mg/kg	(5.2% increase in relative testes weight)	CMA 1983

Bd Wt = body weight; d = day(s); Endocr = endocrine; F = female; Gastro = gastrointestinal; Gd = gestation day; Gn Pig = guinea pig; Hemato = hematological; hr = hour(s); LD_{50} = lethal dose, 50% kill; LOAEL = lowest-observable-adverse-effect level; M = male; MCH = mean corpuscular hemoglobin; MCHC = mean corpuscular hemoglobin concentration; min = minute(s); Musc/skel = musculoskeletal; NOAEL = no-observable-adverse-effect level; NS = not specified; Resp = respiratory; wk = week(s); x = times

Table 2-7. Levels of Significant Exposure to 2-Butoxyethanol Acetate - Oral

Species/ (Strain)	Exposure/ Duration/ Frequency/ (Specific Route)	System	NOAEL (mg/kg/day)	LOAEL		Reference
				Less Serious (mg/kg/day)	Serious (mg/kg/day)	
ACUTE EXPOSURE						
Death						
Rat (Wistar)	once (GO)				3000 M (LD_{50}) 2400 F (LD_{50})	Truhaut et al. 1979

Cardio = cardiovascular; Endocr = endocrine; HGB = hemoglobin; Hemato =hematological; hr = hour(s); LD_{50} = lethal dose, 50% kill; LOAEL = lowest-observable-adverse-effect level; NOAEL = no-observable-adverse-effect level; RBC = red blood cells; Resp = respiratory

interstitial pneumonitis. In another study, male New Zealand white rabbits given a single dose of 0.5 mL/kg 2-butoxyethanol that remained in contact with intact skin for 24 hours showed orange-red lungs at necropsy (Union Carbide 1980b). Death of one of four animals occurred at this dose. Neither clinical signs of respiratory effects nor gross pathology changes in the trachea and lungs were observed in guinea pigs treated with a single dermal dose of 2,000 mg/kglday 2-butoxyethanol (Shepard 1994a). The treated area was occluded for 24 hours. Histological examination of the lungs revealed no adverse treatment-related lesions of male and female New Zealand rabbits dosed with ≤ 150 mg/kg/day 2-butoxyethano16 hours per day, 5 days per week, for 90 days (CMA 1983).

New Zealand rabbits given 24-hour applications of 2-butoxyethanol acetate to the clipped, occluded skin at doses of 3,191-10,000 mg/kg as part of a lethality study exhibited no adverse respiratory clinical signs or histopathological changes in the lungs (Truhaut et al. 1979).

Cardiovascular Effects. Gross changes in the heart at necropsy were not observed in guinea pigs treated with a single dermal dose of 2,000 mg/kg/day 2-butoxyethanol (Shepard 1994a). The treated area was occluded for 24 hours. No effects on heart weight and no histopathological lesions in the aorta or heart were observed in male or female New Zealand rabbits dosed with ≤ 150 mg/kg/day 2-butoxyethano16 hours per day, 5 days per week, for 90 days (CMA 1983).

New Zealand rabbits given 24-hour applications of 2-butoxyethanol acetate to the clipped, occluded skin at doses of 3,191-10,000 mg/kg as part of a lethality study exhibited no histopathological changes in the heart (Truhaut et al. 1979).

Gastrointestinal Effects. Male and female New Zealand white rabbits were dosed dermally once with 451 or 902 mg/kg 2-butoxyethanol, and occluded exposure was maintained for 6 hours (Union Carbide 1980a). All rabbits in the 902-mg/kg dose group died and exhibited reddened stomachs at necropsy; females also had reddened intestines. Rabbits in the 451-mg/kg dose group had no remarkable gastrointestinal findings, and all except one female survived the experiment. However, in a companion study of male New Zealand rabbits that were similarly exposed dermally for a 24-hour period to 451 or 902 mg/kg, gross necropsy of the rabbits that died at 451 mg/kg (one of four) and 902 mg/kg (four of four) revealed orange peritonea and intestines (Union Carbide 1980b). Very dark intestines were also observed in one rabbit dosed with 1,000 mg/kg 2-butoxyethanol on abraded skin (Olin 1976). All four animals treated at this dose died. Gross examination of the esophagus, stomach, duodenum, jejunum, ileum, cecum, colon, and rectum did not

reveal any adverse effects in guinea pigs treated with a single dermal dose of 2,000 mg/kg/day 2-butoxyethanol (Shepard 1994a). The treated area was occluded for 24 hours. Histological examination of the stomach (pylorus and fundus) and of the intestines (cecum, duodenum, jejunum, ileum, colon) revealed no pathological lesions in male and female New Zealand rabbits dosed with 10, 50, or 150 mg/kg/day 2-butoxyethanol 6 hours per day, 5 days per week, for 90 days (CMA 1983). One male in the 50-mg/kg/day dose group that was found dead on day 15 of the study had a gastric ulcer. Neither the ulcer nor the death appeared to be related to treatment.

Hematological Effects. Dermal exposure of animals to 2-butoxyethanol has also resulted in hemolysis of red blood cells and hemoglobinuria. Doses ranging from 200 to 500 mg/kg 2-butoxyethanol were applied to the shaved dorsal skin of female Wistar rats; hemolysis was observed in two of three animals at 260 mg/kg, and hemoglobinuria was observed in one of three animals (Bartnik et al. 1987). A dose of 500 mg/kg caused increased mean corpuscular volume, decreased erythrocyte count and hemoglobin level, and hemoglobinuria within 6 hours after exposure. In another study, 61, 181, or 299 mg/kg [^{14}C]2-butoxy- ethanol was applied to the clipped backs of male Fischer 344 rats, and nonoccluded percutaneous absorption was measured (Sabourin et al. 1992b, 1993). In the 181-mg/kg dose group, no significant hemolysis was detected; hematology was not evaluated at the other doses. Water or undiluted 2-butoxyethanol(0.35 mL) was applied four times daily (1.4 mL/day) on gestational days 7-16 to the shaved interscapular skin of pregnant Sprague-Dawley rats (Hardin et al. 1984). Burgundy-colored urine, presumably from hemoglobin in the urine, was observed by the end of the 1st day of treatment with 2-butoxyethanol. Ten of 11 treated rats died during gestation days 9-l 3.

Male albino New Zealand rabbits exhibited increased erythrocyte osmotic fragility 1 hour after a 3-minute skin contact period with 505 mg/kg 2-butoxyethanol (Carpenter et al. 1956). Hemoglobinuria was also observed in New Zealand rabbits that died after 24-hour skin contact with ≥406 mg/kg (Carpenter et al. 1956). In male and female New Zealand rabbits to which 451 or 902 mg/kg 2-butoxyethanol was applied to the skin (occluded) for 6 hours, hemoglobinuria was observed in both dose groups (Union Carbide 1980a). Lethality was one of eight at 451 mg/kg and all 8 rabbits at 902 mg/kg. Similarly, hemoglobi&ria was noted prior to death in female New Zealand white rabbits treated dermally with ≥72 mg/kg 2-butoxyethanol over an 8-hour period (Duprat and Gradiski 1979). Clipped male and female New Zealand white rabbits received daily dermal (occluded) applications of 1mL/day of 0%, 5%, 25%, 50%, or 100% concentrations (0, l8, 90, 180, or 361 mg/kg/day, respectively) of 2-butoxyethanol for a total of nine applications over an 1l-day period (Union Carbide 1980a). The rabbits were subjected to covered dermal exposure for 6 hours per day.

Transient hemoglobinuria was observed at the high dose in males. In addition, females exhibited increases in mean corpuscular hemoglobin (MCH) and decreases in red blood cell count, hemoglobin, and mean corpuscular hemoglobin concentrations (MCHC) at 361 mg/kg/day.

No adverse treatment-related hematological effects (on white blood cells, red blood cells, hemoglobin, hematocrit, and mean corpuscular volume) were noted in male or female New Zealand rabbits dosed with ≤ 150 mg/kg/day 2-butoxyethano16 hours per day, 5 days per week, for 90 days (CMA 1983).

New Zealand rabbits were given 24-hour applications of 2-butoxyethanol acetate to the clipped, occluded skin at doses of 3,191-10,000 mg/kg as part of a lethality study (Truhaut et al. 1979). In treated rabbits, a decrease in hemoglobin, a 20-25% decrease in red blood cell count, and marked hemoglobinuria and/or hematuria were observed; for each dose group, some rabbits had almost normal values while others were severely affected.

The hemolytic effect of 2-butoxyethanol and 2-butoxyethanol acetate has been associated in animal studies with changes in the spleen, including engorgement and erythrocytic infiltration. These effects are discussed further in Section 2.2.3.3, Immunological and Lymphoreticular Effects.

Musculoskeletal Effects. Flaccid muscle tone, probably neurological in origin, was observed prior to death in rabbits dosed with 1,000 or 2,000 mg/kg 2-butoxyethanol on abraded, occluded skin for 24 hours (Olin 1976). Histological examination of skeletal muscle and bone (sternum) revealed no lesions in male or female New Zealand rabbits dosed with up to 150 mg/kg/day 2-butoxyethano16 hours per day, 5 days per week, for 90 days (CMA 1983).

Hepatic Effects. Pale livers were observed in New Zealand rabbits that died after 24-hour skin contact with ≥406 mg/kg (Carpenter et al. 1956). Female New Zealand outbred rabbits were epicutaneously exposed to ≥ 72 mg/kg 2-butoxyethanol (Duprat and Gradiski 1979). Histopathological examinations of animals that died prior to the end of the 15-day observation period showed congestion in the liver, with small necrotic foci with mesenchymatous reactions and inconstant (i.e., not always present) steatosis. A single 6-hour occluded dermal exposure of male and female New Zealand white rabbits to 451 and 902 mg/kg 2-butoxyethanol caused mottled livers with pocked surfaces in animals that died (Union Carbide 1980a). In a companion study, in which male rabbits were exposed to the same doses for 24 hours, discolored livers were observed at both doses (Union Carbide 1980b). Discolored or pale liver has also been observed in New Zealand rabbits

dosed with 250-l,000 mg/kg 2-butoxyethanol on abraded skin (Olin 1976). Gross examination of the liver did not reveal any adverse effects in guinea pigs treated with a single dermal dose of 2,000 mg/kg/day 2-butoxyethanol (Shepard 1994a); the treated area was occluded for 24 hours. No effect on liver weight was seen in male or female New Zealand white rabbits exposed dermally to 0, 18, 90, 180, or 361 mg/kg/day of 2-butoxyethanol for a total of nine applications over an 11-day period with a duration of 6 hours of exposure per day (Union Carbide 1980a). Histological examination of livers and gall bladders revealed no lesions in male or female New Zealand rabbits dosed with up to 150 mg/kg/day 2-butoxyethanol 6 hours per day, 5 days per week, for 90 days (CMA 1983). In addition, no effects on liver weight were observed.

New Zealand rabbits given 24-hour applications of 2-butoxyethanol acetate to the clipped, occluded skin at doses of 3,191-10,000 mg/kg as part of a lethality study exhibited no histopathological changes in the liver (Truhaut et al. 1979).

Renal Effects. Congested kidneys were observed in New Zealand rabbits that died after 24-hour skin contact with ≥ 406 mg/kg (Carpenter et al. 1956). Female New Zealand outbred rabbits epicutaneously exposed to 2-butoxyethanol at doses of 0, 72, 90, 1108, 135, 180, and 225 mg/kg of the undiluted substance (directly applied over 8 hours onto the clipped back of the unrestrained rabbits) showed adverse kidney effects (Duprat and Gradski 1979). Enlarged kidneys with extensive hemoglobinuric nephrosis and interstitial reaction were observed in rabbits in each dose group (≥ 72 mg/kg) that died. Rabbits that survived and were sacrificed for macroscopic and microscopic examinations showed no differences from control animals at 72 and 90 mg/kg of exposure. Some rabbits in the groups receiving 108, 135, 180, and 225 mg/kg showed persistent kidney lesions (not specified), but the other previously stated morphological changes were not observed. The study authors concluded that the morphological changes in the kidneys suggested damage due to *in vivo* hemolysis. A single 6-hour occluded dermal exposure of male and female New Zealand white rabbits to 451 or 902 mg/kg 2-butoxyethanol caused hematuria at both doses with enlarged, dark kidneys in animals in the high-dose group and kidneys with pocked surfaces in the males of the 451 -mg/kg/day dose group (Union Carbide 1980a). One of eight rabbits died at 451 mg/kg and all eight rabbits died at 902 mg/kg. In a companion study in which male rabbits were exposed to the same doses for 24 hours, dark red kidneys and bloody urine were observed at both doses (Union Carbide 1980b). Discolored kidneys and bloody urine (hematuria) have also been observed in New Zealand rabbits dosed with 500-2,000 mg/kg 2-butoxyethanol on abraded, occluded skin (Olin 1976). Gross examination of the kidneys did not reveal any adverse effects in guinea pigs treated with a single dermal dose of 2,000 mg/kg/day 2 butoxyethanol; the treated area was occluded for 24 hours (Shepard 1994a).

Increased urinary protein was seen in male and female New Zealand white rabbits exposed dermally for 6 hours per day to 36 1 mg/kg/day of 2-butoxyethanol for a total of nine applications over an 11 -day period; however, rabbits exposed to 271 mg/kg/day 2-butoxyethanol nine times over 11 days had renal effects including dimpling of the renal surface, tubular vacuohzation, tubular degeneration, tubular cell hyperplasia, glomerular adhesions, and interstitial nephritis (Union Carbide 1980a). Hematuria was observed in female rabbits exposed to 180 or 361 mg/kg/day 2-butoxyethanol (Union Carbide 1980a). No effects on kidney weight and no treatment-related histopathological lesions in the kidney or urinary bladder were noted in male and female New Zealand rabbits dosed with ≤ 150 mg/kg/day 2-butoxyethano16 hours per day, 5 days per week, for 90 days (CMA 1983).

New Zealand rabbits exposed dermally to doses of 3,191-10,000 mg/kg 2-butoxyethanol acetate for 24 hours exhibited renal effects (Truhaut et al. 1979). Necropsy revealed bloody kidneys and the presence of large quantities of blood in the bladder (hematuria). The severity of the histopathological renal lesions (necrotizing, hemorrhagic, and atrophic acute tubular nephrosis with occasional glomemlar lesions) was reported to be dose-related, but the specific doses at which they occurred were not reported. Histopathologic examination of a section from the kidney of a rabbit treated with 10,000 mg/kg 2-butoxyethanol acetate showed atrophic tubular dilation, tubular fatty degeneration, vacuolar degeneration, and hemosiderin deposits in the glomerulus and the tubular cells, with glomerular retraction and luminar hyaline deposits, luminar granular deposits, foci of cellular lysis, occasional interstitial fibro-inflammatory organization, and parietal atrophy. Presumably, some of these lesions were also observed in rabbits that died. When the surviving animals were sacrificed after the 2-week observation period, no gross pathologic or histopathological lesions in the kidney were noted.

Endocrine Effects. A single 6-hour occluded dermal exposure of male and female New Zealand white rabbits to 451 or 902 mg/kg 2-butoxyethanol caused reddened adrenal glands in both dose groups (Union Carbide 1980a). One of eight rabbits died at 451 mg/kg and all eight rabbits died at 902 mg/kg. Gross examination of the pituitary and adrenal glands did not reveal any adverse effects in guinea pigs treated with a single dermal dose of 2,000 mg/kg/day 2-butoxyethanol (Shepard 1994a); the treated area was occluded for 24 hours. Histological examination of adrenals, pancreas, pituitary, thyroid, and parathyroid glands revealed no pathological lesions in male or female New Zealand rabbits dosed with ≤ 150 mg/kg/day 2-butoxyethanol 6 hours per day, 5 days per week, for 90 days (CMA 1983).

New Zealand rabbits given 24-hour dermal exposure to 2-butoxyethanol acetate at doses of 3,191-10,000 mg/kg as part of a lethality study exhibited no histopathological changes in the pancreas or adrenal gland (Truhaut et al. 1979).

Dermal Effects. Percutaneous absorption of 2-butoxyethanol was investigated in 12 exposure experiments with five men (Johanson et al. 1988). None had been previously exposed occupationally to industrial solvents. All had participated in a previous inhalation study of 2-butoxyethanol (Johanson et al. 1986a). The subjects kept two or four fingers immersed in neat 2-butoxyethanol for 2 hours. None of the subjects exhibited adverse reaction from the exposure to 2-butoxyethanol. Although the skin of the exposed fingers was not irritated, it was wrinkled and seemed to be more rigid and less elastic after exposure. This effect reached a maximum at 2-4 hours after the end of the exposure. The volume and skinfold thickness of the exposed fingers decreased, then returned to normal. A drying pattern with small fissures appeared on the skin, which disappeared after l-2 days.

Volunteers (214 men and women) were given 0.2 mL 10% 2-butoxyethanol in patches applied to the intrascapular area of the back, either to the right or left of the midline (CMA 1992; Greenspan et al. 1995). A 10% concentration was used because it is the highest concentration of 2-butoxyethanol found in cosmetic products, although concentrations as high as 50% can be found in cleaning products which should be diluted before use (OECD 1997). The entire study extended over a 6-week period and involved three phases: induction, rest, and challenge. The induction phase consisted of nine consecutive applications of 2-butoxyethanol and was assessed after 24-72 hours of patch application. The subjects were required to remove the patches approximately 24-72 hours after application; tbey were evaluated and identical patches reapplied. Following the ninth evaluation, the subjects were dismissed for a 14-day rest period. The challenge phase of the experiment was initiated during the 6th week of the study, with identical patches applied to sites previously unexposed to 2-butoxyethanol. These patches were removed by subjects after 24 hours, and the sites were graded 48 and 72 hours after application. Subjects who had six applications or more, with subsequent readings during induction, and at least one reading during challenge, were considered a completed case. One individual had erythema on the second challenge, 7 had barely perceptible erythemaon the first challenge, and 12 had barely perceptible erythema on the second challenge. The study authors concluded that there were no dermal effects of 10% 2-butoxyethanol under the conditions applied in this study; 201 subjects completed the study.

Water or undiluted 2-butoxyethanol was applied four times daily (0.35 mL per application or 1.4 mL/day) on gestational days 7-16 to the shaved interscapular skin of pregnant Sprague-Dawley rats with an automatic pipetter. Progressive necrosis of the tail (which may have been associated with the toxic effects of 2-butoxyethanol on red blood cells and subsequent vascular response) and rough coats were noted in some treated rats (Hardin et al. 1984). Severe skin irritation has been noted in guinea pigs after application of 2-butoxyethanol (dose not specified) (Eastman Kodak 1988). In another study, female New Zealand rabbits exposed to ≥72 mg/kg for 8 hours developed cutaneous lesions accompanied by necrosis of epidermis and dermis on the 4th day after exposure (Duprat and Gradiski 1979). Skin lesions healed within a 2-week period. In other studies, application of 0.5 mL undiluted compound (Rohm and Haas 1983), unspecified amounts of undiluted compound (Dow 1958,1981; Eastman Kodak 1988), or doses of 200 or 252 mg/kg 2-butoxyethanol (Dow 1959) to rabbit skin resulted in hyperemia, edema, slight exfoliation, and/or slight-to-moderate irritation. A single 6-hour occluded dermal application of 451 or 902 mg/kg 2-butoxyethanol to male and female New Zealand white rabbits caused slight necrosis in females at both doses and erythema in both sexes at both doses (Union Carbide 1980a). However, in a companion study in which male rabbits were exposed to the same doses for 24 hours, erythema and necrosis were noted at the high dose only (Union Carbide 1980b). Erythema was also seen in male and female New Zealand white rabbits exposed dermally to 18-361 mg/kg/day of 2-butoxyethanol for a total of nine applications over an 11 -day period with an exposure duration of 6 hours per day; male and female rabbits exposed to 271 mg/kg/day for nine times over 11 days also showed dermal necrosis (Union Carbide 1980a). A single dose of 0.01 mL of undiluted 2-butoxyethanol applied to the clipped bellies of New Zealand white rabbits caused moderate capillary congestion in two of five rabbits (grade 2) (Union Carbide 1980b). Dermal irritation from 2-butoxyethanol exposure has been studied in New Zealand rabbits using both the Draize protocol (24-hour occluded exposure) and the European Economic Communities protocol (4-hour occluded exposure) (Zissu 1995). For both protocols, 0.5 mL of undiluted 2-butoxyethanol was placed on the skin. 2-Butoxyethanol was considered a severe irritant by the Draize protocol and an irritant by the European Economic Communities protocol. Slight-to-moderate erythema was noted in male and female New Zealand rabbits dosed with 10-150 mg/kg/day 2-butoxyethanol 6 hours per day, 5 days per week, for 90 days (CMA 1983). Slight-to-moderate scaling and flaking were occasionally noted during weeks 2 and 3 and recurred sporadically through 91 days.

2-Butoxyethanol acetate was used to evaluate a noninvasive human method and an *in vitro* cytotoxicity method as alternatives to the rabbit skin test for primary irritant effects (Jacobs et al. 1989). The human method included measurements of cutaneous blood flow values (CBFV) by laser Doppler flowmetry before and after patch application on the forearm; some experiments used 100% test substances (75 mg/cm^2)

applied for 48 hours and measured 12 hours later, and other experiments used 10% solutions of test substances (7.5 mg/ m^3) applied for 3 hours and measured 1, 24, 48, and 72 hours later. Cutaneous blood flow values (CBFV) obtained in the two series of experiments (10% for 3 hours and 100% for 48 hours) resulted in a ranking of 2, which is the minimal mean erythema needed to classify substances as skin irritants. The CBFV in humans correlated very well (r=0.99) with erythema scores obtained on rabbits.

In New Zealand white rabbits, patches soaked with 0.5 mL of undiluted 2-butoxyethanol acetate were applied to one site of the prepared skin surface (Jacobs et al. 1989). The untreated skin was used as a control. Erythema scores were obtained 1, 24, 48, and 72 hours after application of the undiluted substance for 4 hours. The erythema scores of exposure to 100% 2-butoxyethanol acetate for 4 hours in rabbits showed a ranking of 2, which is the minimal mean erythema needed to classify substances as skin irritants. New Zealand rabbits exposed dermally to unspecified doses of 2-butoxyethanol acetate for 24 hours exhibited dermal effects (Truhaut et al. 1979). When 2-butoxyethanol acetate was tested for primary irritation of the skin, four of six rabbits showed slight erythema (grade 1) at 24 hours. Dermal irritation from exposure to 2-butoxyethanol acetate has been studied in rabbits using both the Draize protocol (24-hour occluded exposure) and the European Economic Communities protocol (4-hour occluded exposure) (Zissu 1995). For both protocols, 0.5 mL of undiluted 2-butoxyethanol acetate was placed on the skin. 2-Butoxyethanol acetate was considered a moderate irritant by the Draize protocol and non-irritating by the European Economic Communities protocol.

Ocular Effects. Male and female volunteers exposed by inhalation to 98, 113, or 195 ppm 2-butoxyethanol for 4-8 hours experienced ocular irritation during exposure to 113 and 195 ppm (Carpenter et al. 1956). As discussed in Section 2.2.1.2, the ocular irritation was probably due to direct contact of the eyes with the 2-butoxyethanol vapor.

Pregnant Fischer 344 rats dosed by inhalation during gestational days 6-15 with 25-200 ppm 2-butoxyethanol exhibited periocular wetness at all concentrations. New Zealand white rabbits exposed to 100 and 200 ppm (but not to 25 ppm) during gestation exhibited the same effect (Tyl et al. 1984). As noted in Section 2.2.1.2, the periocular wetness was probably due to direct contact of the eyes with the 2-butoxyethanol vapor.

The eyes of rabbits instilled with unspecified amounts of undiluted 2-butoxyethanol (Dow 1958, 1959, 1981) or 0.1 mL of undiluted 2-butoxyethanol (Rohm and Haas 1983; Union Carbide 1980b) exhibited severe eye

irritation, moderate-to-extensive conjunctivitis, moderate cornea1 damage, and/or slight iritis (Dow 1958, 1959, 1981; Rohm and Haas 1983; Union Carbide 1980b). Moderate comeal injury was observed in rabbits in which 0.5 mL of a 15% dilution of 2-butoxyethanol was placed in the conjunctival sac (Union Carbide 1980b). No effects were observed with a dilution of 5%. A single 6-hour occluded dermal exposure of male and female New Zealand white rabbits to 451 or 902 mg/kg 2-butoxyethanol caused grey iris in males in the high-dose group (Union Carbide 1980a). No ocular effects were seen at the lower dose. In a companion study in which male rabbits were given the same dermal doses for 24 hours, iritis was observed at the high dose (Union Carbide 1980b). Lacrimation was also observed prior to death in New Zealand rabbits dosed with 2,000 mg/kg 2-butoxyethanol on abraded skin, and yellow cornea was observed in animals dosed with 500 mg/kg 2-butoxyethanol (Olin 1976). No ocular effects were seen upon ophthalmological examination of male and female New Zealand white rabbits exposed dermally to 0, 18, 90, 180, or 361 mg/kg/day of 2-butoxyethanol for a total of nine applications over an 11 -day period with an exposure duration of 6 hours per day (Union Carbide 1980a).

The relationship between changes in cornea1 thickness (swelling) and Drake eye irritancy scores was examined to see if the former test could give equivalent results and a reduction in the variability inherent in the Draize procedure (Kennah et al. 1989a). The eyes of New Zealand albino rabbits were instilled with 0.1 mL 2-butoxyethanol at concentrations of 10%, 20%, 30%, 70%, or 100% (dilutions were prepared in polyethylene glycol as weight/weight percentage solutions) in the conjunctival sac of one defect-free eye. The other eye served as a control. The cornea, iris, and conjunctiva were scored at 24, 48, and 72 hours and at 7, 10, 14, and 21 days postdosing if irritation persisted. Cornea1 thickness was measured several hours before instillation of test material and thereafter at intervals coincident with the Draize scoring. A correlation with the Draize scoring procedure and cornea1 swelling was established. Eye irritation determination using this system showed that at 100% concentration, 2-butoxyethanol resulted in a Draize score of 66% with 19% coefficient of variation (CV), 181% cornea1 swelling with 9.9% CV, and was classified as severe. At 30% and 70% concentrations of 2-butoxyethanol, Draize scores were 39% and 49% with CV values of 13% and 44%; cornea1 swelling values were 146% and 181%, with CV values of 9.7% and 18%. The Draize scores indicated moderate irritation. At 10% and 20% concentrations of 2-butoxyethanol, Draize scores were 1% and 2%, with CV values of 100% and 70%; cornea1 swelling values were 91% and 113%, with CV values of 7.4% and 13%. The Draize scores indicated mild irritation.

Histological examination of the eyes revealed no adverse treatment-related ocular effects in male or female New Zealand rabbits dosed dermally with up to 150 mg/kg/day 2-butoxyethano16 hours per day, 5 days per week, for 90 days (CMA 1983).

The eyes of New Zealand rabbits instilled with an unspecified amount of 2-butoxyethanol acetate for 24 hours exhibited little effect (Truhaut et al. 1979). The eyes showed very little sign of irritancy, with only two of six rabbits showing slight conjunctival redness and discharge.

Body Weight Effects. 2-Butoxyethanol caused a 13-16% decrease in body weight gain during treatment in pregnant Sprague-Dawley rats to which undihrted 2-butoxyethanol was applied four times daily (0.12 mL, per application or 0.48 mL/day) to the shaved interscapular skin on gestational days 7-16 (Hardin et al. 1984). In guinea pigs (sex and strain not specified) exposed epicutaneously to 0.5 or 2.0 mL 2-butoxy-ethanol, and observed for 35 days, no effect on body weight was observed at the low dose (Wahlberg and Boman 1979). Mortality at the high dose was significant (65% within 1 week), and so no body weight measurements were made. No effects on body weight gain were observed in guinea pigs given a single dermal dose of 2,000 mg/kg and observed for 14 days (Shepard 1994a). The treated area was occluded for 24 hours. A slight but unspecified initial weight loss occurred in rabbits to which 200 mg/kg 2-butoxyethanol was applied to the skin for 24 hours (Dow 1959). No effect on body weight was seen in male New Zealand white rabbits exposed dermally to 0, 18, 90, 180, or 361 mg/kg/day of 2-butoxyethanol for a total of nine applications over an 11-day period with an exposure duration of 6 hours per day, but female rabbits exhibited a decrease in weight gain of 121% on day 3 at 361 mg/kg/day compared to control animals, and continued to lose weight until sacrificed on day 35 (Union Carbide 1980a). No adverse treatment-related effects on body weight were noted in male and female New Zealand rabbits that received dermal doses ≤ 150 mg/kg/day 2-butoxyethano16 hours per day, 5 days per week, for 90 days (CMA 1983).

Other Effects. Pregnant Sprague-Dawley rats treated dermally four times daily with 2-butoxyethanol (0.35 mL per application or 1.4 ml/day) during gestation exhibited dark stains around the muzzle and anogenital area (Hat-din et al. 1984). This may have resulted from grooming activities in the presence of hemoglobinuria. Female New Zealand white rabbits that died after dermal application of 2-butoxyethanol at ≥72 mg/kg exhibited hypothermia (Duprat and Gradiski 1979). Yellowed peritoneal fat was observed in male and female New Zealand white rabbits that died after a single 6-hour occluded dermal exposure to 902 mg/kg 2-butoxyethanol (Union Carbide 1980a). This effect was not noted in animals exposed to 451 mg/kg, a dose that resulted in the death of one of four females and no males.

2.2.3.3 Immunological and Lymphoreticular Effects

No studies were located regarding immunological or lymphoreticular effects in humans after dermal exposure to 2-butoxyethanol acetate or regarding lymphoreticular effects in humans after dermal exposure to 2-butoxyethanol. No studies were located regarding immunological effects in animals after dermal exposure to 2-butoxyethanol or 2-butoxyethanol acetate.

Volunteers (214 men and women) were given 0.2 mL 10% 2-butoxyethanol in patches applied to the intrascapular area of the back, either to the right or left of the midline (CMA 1992; Greenspan et al. 1995). A 10% concentration was used because it is the highest concentration of 2-butoxyethanol found in cosmetic products. The entire study extended over a 6-week period and involved three phases: induction, rest, and challenge. The induction phase consisted of nine consecutive applications of 2-butoxyethanol, and the subjects were assessed after 24-72 hours of patch application The subjects were required to remove the patches approximately 24-72 hours after application; they were evaluated and identical patches reapplied. Following the ninth evaluation, the subjects were dismissed for a 14-day rest period. The challenge phase of the experiment was initiated during the 6th week of the study, with identical patches applied to sites previously unexposed to 2-butoxyethanol. These patches were removed by subjects after 24 hours, and the sites were graded 48 and 72 hours after application Subjects who had six applications or more, with subsequent readings during induction, and at least one reading during challenge, were considered completed cases. One individual had erythema on the second challenge, 7 had barely perceptible erythema on the first challenge, and 12 had barely perceptible erythema on the second challenge. The study authors concluded that there was no evidence of sensitization to 10% 2-butoxyethanol in human subjects under the conditions applied in this study; 201 subjects completed the study. However, only a 10% dilution of 2-butoxyethanol was used; concentrations as high as 50% can be found in cleaning products which should be diluted before use (OECD 1997).

2-Butoxyethanol has also been tested for dermal sensitization in guinea pigs using the maximized Magnusson and Kliman test (Zissu 1995). The sensitization test was completed over a 4-week period. The sensitization exposure involved an injection of 2-butoxyethanol with Freund’s adjuvant. The animals were injected behind the shoulders at the beginning of the 1st week of this study. On the 8th day of the study, the animals received a 48-hour topical application of 2-butoxyethanol. The doses for the sensitization exposures were not stated. The guinea pigs were allowed to rest during the 3rd week. On the 24th day of the study, 0.5 mL of a 1% 2-butoxyethanol solution was applied for 48 hours to the left sheared flank using an occlusive patch. The

intensity of the erythema and edema was scored, and histopathological examinations were completed 24 hours after the removal of the patch. By this assay, 2-butoxyethanol did not result in dermal sensitization of guinea pigs.

Effects on lymphoreticular organs have been observed in animals after dermal exposure to 2-butoxyethanol. Congestion in the spleen, erythrocytic infiltration, and white atrophic pulp were noted in female New Zealand white rabbits that died after ≥72 mg/kg 2-butoxyethanol was applied to their clipped backs for 8 hours (Duprat and Gradiski 1979). Similarly, engorged spleen was noted in rabbits that died after acute dermal doses of ≥406 mg/kg (Carpenter et al. 1956). In addition, enlarged spleens were found in female New Zealand rabbits that died after a 6-hour occluded exposure to 902 mg/kg 2-butoxyethanol (Union Carbide 1980a), and dark red spleens were found in male rabbits that died after a 24-hour exposure to 451 mg/kg (Union Carbide 1980b). Gross examination of the thymus, spleen, lymph nodes, and bone marrow did not reveal any adverse effects in guinea pigs treated with a single dermal dose of 2,000 mg/kg and observed for 14 days (Shepard 1994a). The treated area was occluded for 24 hours. No effects on spleen or thymus weight and no histopathological lesions in the spleen, lymph nodes, or thymus were observed in male or female New Zealand rabbits that received dermal doses of ≤ 150 mg/kg/day 2-butoxyethano16 hours per day, 5 days per week, for 90 days (CMA 1983). Increased spleen weight, dark spleens, and spleen engorgement effects are probably due to hemolysis, which is discussed under Hematological Effects.

For 2-butoxyethanol acetate, histological examination of spleens revealed no lesions in New Zealand rabbits exposed dermally to doses of ≤ 10,000 mg/kg for 24 hours (Truhaut et al. 1979).

The highest NOAEL values and all LOAEL values from each reliable study for immunological or lympho-reticular effects in each species and duration category are recorded in Tables 2-6 and 2-7.

2.2.3.4 Neurological Effects

No studies were located regarding neurological effects in humans after dermal exposure to 2-butoxyethanol or 2-butoxyethanol acetate.

Prostration and narcosis prior to death were noted in female New Zealand white rabbits after 2-butoxyethanol was applied to their clipped backs for 8 hours at ≥ 72 mg/kg (Duprat and Gradiski 1979). Anorexia and lack of spontaneous movement were observed prior to death in New Zealand rabbits given occluded exposure to

2,000 mg/kg 2-butoxyethanol for 24 hours on abraded skin (Olin 1976). Muscular flaccidity was also seen in the rabbits at 1,000 and 2,000 mg/kg. Ataxia progressing to moderate-to-marked inactivity and death occurred in pregnant Sprague-Dawley rats treated dermally four times daily with 2-butoxyethanol(0.35 mL per application or 1.4 mL/day) on gestational days 7-16 (Hardin et al. 1984). Female, but not male, New Zealand rabbits receiving a 6-hour occluded dermal applicatiou of 902 mg/kg 2-butoxyethanol exhibited nystagmus and convulsions prior to death (Union Carbide 1980a). In an intermediate-duration study, histological examination of the brain and sciatic nerve revealed no pathological lesions in male or female New Zealand rabbits dosed dermally at ≤ 150 mg/kg/day 2-butoxyethanol6 hours per day, 5 days per week, for 90 days (CMA 1983).

For 2-butoxyethanol acetate, histological examination of the brain revealed no lesions in New Zealand rabbits exposed dermally to ≤ 10,000 mg/kg for 24 hours (Truhaut et al. 1979).

The highest NOAEL values and all LOAEL values from each reliable study for neurological effects in each species and duration category are recorded in Tables 2-6 and 2-7.

2.2.3.5 Reproductive Effects

No studies were located regarding reproductive effects in bumans after dermal exposure to 2-butoxyethanol or 2-butoxyethanol acetate.

Information regarding reproductive effects in animals after dermal exposure to 2-butoxyethanol or 2-butoxy-ethanol acetate is limited to data on reproductive organ weights and histological examination of reproductive organs No studies assessing reproductive function were Located. Gross changes at necropsy were not observed in the reproductive organs (epididymides, testes, ovaries, fallopian tubes, uterus, vagina, cervix, uteri) of guinea pigs given a single dermal(24 hours occluded) dose of 2,000 mg/kg and observed for 14 days (Shepard 1994a). No effect on testicular weight was found in male New Zealand white rabbits given dermal applications of 18-361 mg/kg/day 2-butoxyethanol for 6 hours per day for nine applications over 11 days (Union Carbide 1980a). The reproductive organ weights of female rabbits were not determined. Histological examination of testes, epididymides, seminal vesicles, prostate, mammary glands, ovaries, uterus, and vagina revealed no pathological lesions in male or female New Zealand rabbits dosed with 10, 50, or 150 mg/kg/day 2-butoxyethanol for 6 hours per day, 5 days per, week for 90 days (CMA 1983). A slight (5.2%), but

statistically significant, increase in relative testicular weight was noted for males in the 150-mg/kg/day dose group, but no effects on the weight of the ovaries were found in the females at any dose.

For 2-butoxyethanol acetate, histological examination of the testes and ovaries revealed no pathological lesions in male or female New Zealand rabbits exposed dermally to ≤ 10,000 mg/kg for 24 hours (Truhaut et al. 1979).

The highest NOAEL values and all LOAEL values from each reliable study for reproductive effects in each species and duration category are recorded in Tables 2-6 and 2-7.

2.2.3.6 Developmental Effects

No studies were located regarding developmental effects in humans after dermal exposure to 2-butoxyethanol or 2-butoxyethanol acetate or regarding developmental effects in animals after dermal exposure to 2-butoxyethanol acetate.

Water or undiluted 2-butoxyethanol was applied four times daily (0.12 mL per application or 0.48 mL/day) on gestational days 7-16 to the shaved interscapular skin of pregnant Sprague-Dawley rats (Hardin et al. 1984). No embryotoxic, fetotoxic, or teratogenic effects were detected in the offspring. At necropsy, no gross malformations were observed in the fetuses

The NOAEL value for developmental effects in rats after acute dermal exposure to 2-butoxyethanol is recorded in Table 2-6.

2.2.3.7 Genotoxic Effects

No studies were located regarding genotoxic effects in humans or animals after dermal exposure to 2-butoxy-ethanol or 2-butoxyethanol acetate. *In vitro* genotoxicity studies are discussed in Section 2.5.-

2.2.3.8 Cancer

No studies were located regarding carcinogenic effects in humans or animals after dermal exposure to 2-butoxyethanol or 2-butoxyethanol acetate.

2.3 TOXICOKINETICS

2-Butoxyethanol is well-absorbed following inhalation, oral, and direct dermal exposure. Dilution with water has resulted in more rapid absorption of 2-butoxyethanol following *in vitro* exposure of rat skin. Small amounts of the vapor can also be absorbed through the skin. There is little information concerning the absorption of 2-butoxyethanol acetate. 2-Butoxyacetic acid has been detected in persons occupationally exposed to 2-butoxyethanol acetate, and the compound was absorbed following direct dermal exposure of rabbits.

Metabolism of 2-butoxyethanol and 2-butoxyethanol acetate to 2-butoxyacetic acid and further metabolism to carbon dioxide is the major pathway of metabolism in both humans and animals. The production of ethylene glycol is a minor pathway of metabolism in both animals and humans. 2-Butoxyethanol and 2-butoxyacetic acid are widely distributed throughout the body, with no site of accumulation identified. Following all routes of exposure, urinary excretion of 2-butoxyacetic acid and its conjugates is the major route of elimination.

2.3.1 Absorption

2.3.1.1 Inhalation Exposure

Information describing human absorption of 2-butoxyethanol after inhalation exposure comes from both controlled experiments and occupational exposure. Seven male volunteers were exposed to 2-butoxyethanol at the Swedish occupational exposure limit (20 ppm or 0.85 mmol/ m^3) for 2 hours during light physical exercise on a bicycle ergometer (Johanson et al. 1986a). Expired air was collected at regular time intervals for estimation of the respiratory uptake of the solvent. Capillary blood was sampled during and after the exposure period and analyzed for 2-butoxyethanol. The respiratory uptake of 2-butoxyethanol averaged 10.1 µmol/minute (1.2 mg/minute) or 57% of the inspired amount. The concentration in blood reached a plateau level of 7.4 µmol/L (0.9 mg/L) within 1-2 hours. In another experimental study in humans, venous blood samples were collected from five healthy male volunteers prior to and after inhalation exposure to 20 ppm 2-butoxyethanol (the Swedish permissible exposure limit) for 2 hours during light physical exercise (Johanson and Johnsson 1991). Samples were collected at 0, 2, 4, and 6 hours from the start of exposure, and analysis indicated that 2-butoxyacetic acid, the major metabohte of 2-butoxyethanol, was found in all

samples except those collected prior to exposure. The minimum and maximum observed concentrations of 2-butoxyacetic acid in blood ranged from 20 to 57 µM (2.6-7.5 mg/L) with average concentrations of 22, 44, 39, and 28 µM at 1, 3, 4.9, and 7.1 hours, respectively. The 2-butoxyacetic acid blood level peaked after 2-4 hours.

Healthy male volunteers were exposed to 50 ppm 2-butoxyethanol vapor for 2 hours by mouth through a respiratory valve (Johanson and Boman 199 1) Capillary blood samples were collected at regular intervals and analyzed for 2-butoxyethanol. Duplicate experiments were carried out on each volunteer, with normal or increased temperature and humidity. The concentration of 2-butoxyethanol increased during the 1 st hour and appeared to approach steady state at about 3 µM during the 2nd hour. The respiratory uptake rate during mouth exposure was 1.3 mmole over a period of 2 hours, or 11 µmol/minute.

Seventeen people (2 women and 15 men), who were exposed to glycol ethers in a varnish production plant, were examined to determine their environmental and internal solvent exposure (Angerer et al. 1990). The workers in the production plant (n=12) were exposed to an average 2-butoxyethanol concentration of 1.1 ppm, among other solvents. Internal exposure was estimated by measuring free 2-butoxyethanol in blood as well as free 2-butoxyacetic acid in urine samples. Urine samples were taken pre- and postshift. The exposure of varnish workers to 2-butoxyethanol ranged from <0.1 to 8.1 ppm, compared to <0.1 ppm for workers in the store or the laboratory. The average permissible exposure limit was 20 ppm (Germany). Biological monitoring indicated 2-butoxyethanol levels in postshift blood of 121.3 µg/L for varnish production (range: <5.0-570.0 µg /L), compared to 49.4 µg /L for store workers (range: <5.0-143.1 µg/L) and <5.0 µg /L for laboratory workers. Pre- and postshift levels of 2-butoxyacetic acid in urine (mg/L) were 3.3 and 10.5 for varnish workers, 2.1 and 4.5 for store workers, and 0.2 and 4.2 for laboratory workers, respectively. Values were not normalized to creatinine. There was little correlation between external and internal levels of exposure, presumably because there was significant direct dermal exposure that was not measured. The study authors indicate that “At these working places glycol ethers were absorbed through the skin on a major scale. Because the workers were cleaning instruments, floor and skin with the solvents, skin contact was far from negligible.”

Individual exposure to 2-butoxyethanol acetate was measured by personal air sampling and biological monitoring of urine from 19 employees during an 8-hour workday in four silk-screen printing installations (Johanson et al. 1989). The samples were analyzed by gas chromatography. Urine was quantitatively collected from each subject two or three times during the day. 2-Butoxyethanol acetate was detected in the

personal air samples of 5 individuals at an average concentration of 0.44 ppm. Free 2-butoxyacetic acid was detected in the urine of 12 individuals at an average concentration of 8 µmol/L.

The data on absorption of 2-butoxyethanol after inhalation exposure in animals is scanty. In an early study, rats, guinea pigs, dogs, and rabbits exposed to 100-400 ppm 2-butoxyethanol for 4 hours excreted 2-butoxyacetic acid in the urine, with most of the metabolite excreted within 24 hours, indicating pulmonary absorption (Carpenter et al. 1956). Dogs and monkeys exposed to 100-398 ppm 2-butoxyethanol for 28-90 days also excreted 2-butoxyacetic acid in the urine.

In a more definitive study, the uptake, metabolism, and excretion of 2-butoxyethanol following 6-hour nose-only inhalation exposure of male Fischer 344 rats was determined at different inhaled concentrations (Sabourin et al. 1992a). Three to five rats per group were exposed to 4.3, 49, or 438 ppm 2-butoxyethanol for 6 hours for the determination of respiratory uptake and excretion. Additional groups of rats were used to determine fractional uptake of inhaled 2-butoxyethanol and body burden at the end of the exposure. Rats selected for body burden determination were euthanized, and the entire carcass was digested at the end of the 6-hour exposure period. Rats selected for metabolism determination were put into metabolism cages following the 6-hour exposure to determine excretion of the compound. The uptake was 4.49 µmol/ppm at 4.3 ppm, 4.58 µmol/ppm at 40 ppm, and 3.58 µmol/ppm at 438 ppm. A less-than-proportional amount of 2-butoxyethanol was inhaled at the high dose because of decreased minute volume.

In a more recent study, eight male Sprague-Dawley rats were exposed to 20 or 100 ppm 2-butoxyethanol 24 hours per day for 1, 2, 3, 4, 6, 8, 10, or 12 days (Johanson 1994). Urine was collected at 24-hour intervals. At the end of the exposure period, animals were killed and blood, muscle, liver, and testis were collected. Blood and tissue samples were analyzed for 2-butoxyethanol and 2-butoxyacetic acid by electron capture gas chromatography. No control group was used. The concentration of 2-butoxyethanol in the four tissues examined increased rapidly during the first l-3 days of exposure. The average peak concentration of 2-butoxyethanol in the blood was 15.1 µmol/L in the 20-ppm group and 72.3 µmol/L in the 100-ppm group, indicating linear uptake by blood. Respiratory uptake averaged 0.31 mmol/day at 20 ppm and 1.57 mmol/day at 100 ppm, also indicating linear pulmonary uptake at exposure concentrations of ≤ 100 ppm 2-butoxyethanol.

2.3.1.2 Oral Exposure

Case reports of intentional poisonings with 2-butoxyethanol provide some kinetic information for human oral exposure. A report on a woman admitted to the hospital after ingestion of 250-500 mL of a window cleaner containing 12% 2-butoxyethanol(467-933 mg/kg 2-butoxyethanol) in a suicide attempt indicated metabolic acidosis, hypokalemia, a rise in serum creatinine level, and markedly increased urinary excretion of oxalate crystals, indicating substantial absorption from the gastrointestinal tract (Rambourg-Schepens et al. 1988). Similar effects have been observed in other case reports of humans who ingested 2-butoxyethanol at 391-650 mg/kg (Bauer et al. 1992; Gijsenbergh et al. 1989). In the case reported by Rambourg-Schepens et al. (1988), kinetic evaluation of urinary excretion of 2-butoxyethanol indicated that 2-butoxyethanol was at the highest level of excretion at 1 day after ingestion.

Studies in animals indicate that absorption of 2-butoxyethanol after oral exposure is rapid and extensive (Ghanayem et al. 1987c). Male Fischer 344 rats were given a single oral dose of [^{14}C]2-butoxyethanol at 125 or 500 mg/kg. Rats were placed in metabolism cages and monitored for 48 hours after treatment. At 48 hours, animals were killed and the ^{14}C -radioactivity in each tissue, as well as in the blood, was determined. Detectable exhalation of radioactivity began l-2 hours after treatment, indicating rapid absorption and distribution. Approximately 18% and 10% of the administered doses of 125 and 500-mg/kg, respectively, was exhaled as $^{14}CO_2$ in 48 hours. Cumulative urinary excretion of radioactivity amounted to approximately 32% and 18% of the 125-and 500-mg/kg doses at 8 hours, to approximately 64% and 30% of the respective doses at 24 hours, and to approximately 70% and 40% of the respective doses at 48 hours. Fecal excretion of radioactivity amounted to 2-3% of the doses in 48 hours, and may represent biliary excretion (biliary metabolites were identified, see below) as well as unabsorbed 2-butoxyethanol. Thus, by adding the 48-hour excretion of radioactivity in the expired air and urine, at least 88% of the 125-mg/kg dose and at least 50% of the 500-mg/kg dose were absorbed. The fact that the percentage of radioactivity excreted was less at the higher dose than at the lower dose may indicate saturation of a metabolic pathway rather than lower absorption of the high dose, based on the finding that the urinary levels of the glucuronide conjugate of 2-butoxyethanol of the high-dose rats remained constant for 24 hours, while the glucuronide l&els declined after 8 hours in the low-dose rats (see Section 2.3.3 on Metabolism, below).

In male Fischer 344 rats given single gavage doses of 8.6 or 126 mg/kg [^{14}C]2-butoxyethanol, 59% of the low dose of radioactivity and 38-70% of the high dose of radioactivity were excreted in the urine during the first 24 hours (Corley et al. 1994). Excretion of $^{14}CO_2$ in the expired air amounted to 7.2% of the low-dose

and 8% of the high dose in 24 hours. Thus, at least 66.2% of the low dose and 46-78% of the high dose were absorbed.

Male Fischer 344/N rats were allowed access for 24 hours to uniformly labeled [^{14}C]2-butoxyethanol in drinking water at three doses (Medinsky et al. II 990). Elimination of radioactivity was monitored for 72 hours. 2-Butoxyethanol was administered at doses of 290, 860, or 2,590 ppm in drinking water, resulting in consumption of 237, 401, or 1,190 µmol//kg (28, 47, or 141 mgIkg/day). The majority of the ^{14}C was excreted in urine (50-60% of the dose as 2-butoxyacetic acid and 10% as ethylene glycol) or exhaled as CO_2 (8-10% of the dose). Less than 5% of the dose was exhaled as unmetabolized 2-butoxyethanol.

2.3.1.3 Dermal Exposure

Worker exposures to 2-butoxyethanol were evaluated by euvironmental and biological monitoring for 29 employees in five shops where window-cleaning agents had been used (Vincent et al. 1993). Each of the 29 employees was exposed to 2-butoxyethanol. Eleven men and two women were involved in cleaning cars in four garages. The remaining 16 were office cleaners. Only two of the workers who cleaned new cars wore protective gloves. The study authors suggested that the primary route of exposure was dermal, although some inhalation exposure necessarily occurred. Each of the window-cleaning agents sampled contained 2-butoxyethanol in concentrations ranging from 0.9% to 21.2% in volume. The duration of window-cleaner use ranged from 120 to 320 minutes per day, and quantities of cleaning agents ranged from 220 to 1,200 mL/day for individuals involved in cleaning new cars. The corresponding figures for cleaners of used cars were 20-50 minutes and 50-100 mL. For office cleaners, the corresponding figures were 15 minutes a day and <50 mL of cleaning agent used per day. For cleaners of new cars, the mean 2-butoxyethanol exposure was 2.33 ppm (range, <0.10-7.33 ppm), and the mean urinary 2-butoxyacetic acid concentration in endshift samples was 111.3 mg/g creatinine (range 12.7-371 mg/g creatinine). Mean preshift (average of Monday and Friday) urinary 2-butoxyacetic acid concentration was 17.9 mg/g creatinine (range, <2-98.6 mg/g creatinine). For cleaners of used cars, the mean 2-butoxyethanol exposure was 0.36 ppm (range, <0.10-l .52 ppm), and the mean urinary 2-butoxyacetic acid concentration in endshift samples was 6.3 mg/g creatinine (range, 2-24.4 mg/g creatinine). Mean preshift urinary concentration of 2-butoxyacetic acid was 4.8 mg/kg creatinine (range, 2-33 mg/g creatinine). For the office cleaners, the mean 2-butoxyethanol exposure concentration was 0.32 ppm (range, <0.30-0.73 ppm), and the mean urinary 2-butoxyacetic acid concentration in endshift samples was 2.1 mg/g creatinine (range, 2-3.3 mg/g creatinine). Mean preshift urinary concentration of 2-butoxyacetic acid was 2.1 mg/g creatinine (range, 2-4.6 mg/g

creatinine). Correlations of 2-butoxyethanol exposure in air and endshift concentrations of urinary 2-butoxyacetic acid from cleaners of new and used cars (for which the data were most complete) indicated a log-linear relationship. From these results, the study authors estimated that exposure to window cleaners containing 5.7-21.2% 2-butoxyethanol by volume for a period of 160 minutes would result in an endshift urinary 2-butoxyacetic acid concentration of 60 mg/g creatinine. A similar relationship was observed for urinary 2-butoxyacetic acid concentrations and utilization time for office cleaners. The positive correlations between exposure levels of 2-butoxyethanol and 2-butoxyacetic acid in the urine indicate dermal (and probably some pulmonary) absorption of 2-butoxyethanol by these workers.

Experimental evidence exists that dermal absorption of 2-butoxyethanol vapors may contribute to the overall absorption of 2-butoxyethanol during vapor exposure (Johanson and Boman 1991). Healthy male volunteers were exposed to 50 ppm 2-butoxyethanol vapor for 2-hour periods, first by mouth only through a respiratory valve, then by skin only while wearing respiratory protection. Capillary blood samples (finger prick) were collected at regular intervals and analyzed for 2-butoxyethanol. Duplicate experiments were carried out on each volunteer, with normal or increased temperature and humidity. In the mouth-only study, the concentration of 2-butoxyethanol increased during the 1st hour and appeared to approach steady state at about 3 µM during the 2nd hour. The respiratory uptake was 1.3 mmole over 2 hours or 11 µmol/rninute and the apparent blood clearance was 3.8 L/minute. In the percutaneous exposure, the concentration of 2-butoxy-ethanol increased to about 9 µM during the 2nd hour. The half-life of 2-butoxyethanol in the blood after skin exposure was about 34 minutes. The average concentration in blood and the calculated rate of uptake of 2-butoxyethanol were about 3-4 times higher during dermal exposure than during inhalation exposure, suggesting that about 75% (45-85%) of the total uptake during normal vapor exposure could be accounted for by dermal absorption. However, the experimental subjects wore only shorts, which is dissimilar from the usual occupational or consumer exposure scenario. In addition, it has been suggested that the finger prick method of blood sampling provides a sample of venous blood draining the skin, rather than a sample of systemic blood (Corley et al. 1997). Therefore, the estimate of dermal absorption by Johanson and Boman (1991) is most likely an overestimate.

A study of 6 men exposed one arm-only to 50 ppm 2-butoxyethanol ($^{13}C_2$ labeled on the ethylene glycol backbone) for 2 hours, in which blood samples were taken from the unexposed arm, and from a finger prick of the exposed arm, shows much lower dermal absorption of 2-butoxyethanol vapor (Corley et al. 1997) than noted by Johanson and Boman (1991). Concentrations of 2-butoxyethanol in the finger prick sample from the exposed arm were about 1,500 times greater than the blood concentration in the sample from the

unexposed arm. Concentrations of 2-butoxyacetic acid were about 37 times greater in the finger prick sample from the exposed arm compared to the sample from the unexposed arm, confirming that the finger prick method of blood sampling provides a sample of venous blood draining the skin. Based on data from the Corley et al. (1997) study and from the Johanson and Boman (1991) study, skin permeability coefficients (k_p) of 2 and 4 cm/hour were estimated for normal (23ºC, 29% relative humidity) and elevated (33°C, 71% relative humidity) temperature and relative humidity, respectively. The Corley (1994) model was further refined by modifying it to better reflect human metabolism as measured by urinary metabolite levels in Corley et al (1997), and to include a new permeability coefficient. Dermal uptake was estimated under worst-case resting conditions (respiration rates at their lowest, no clothing worn) and under more realistic conditions (35% of the body surface area exposed). Under the worst case conditions, 15-27% of the total uptake of 2-butoxyethanol vapor would be through the skin, while dermal uptake would be 4.4-8.4% of the total if only 25% of the body surface was exposed. Under simulated exercise (50 W, increased respiratory rates and cardiac output), uptake was estimated as 4.6-8.7% and 1.2-2.3% of the total assuming 100% or 25% of the body surface area was exposed, respectively.

The percutaneous absorption of liquid 2-butoxyethanol was investigated in 12 exposure experiments with five men (Johanson et al. 1988). All were nonsmokers and stated low or no consumption of alcohol. None had been exposed to industrial solvents. All had participated in a previous study of 2-butoxyethanol (Johanson et al. 1986a). The subjects kept two or four fingers immersed in neat 2-butoxyethanol for 2 hours. Capillary blood samples were collected from the other hand before, during, and up to 4 hours after the exposure and analyzed for 2-butoxyethanol. The presence of 2-butoxyethanol in blood and 2-butoxyacetic acid in urine confirmed that 2-butoxyethanol enters the systemic circulation in human males during *in vivo* dermal exposure. The shape of the uptake profile varied considerably among individuals and also among experiments. Percutaneous uptake rates were calculated from measured blood levels of 2-butoxyethanol with the use of kinetic parameters (clearance and volume of distribution) obtained in earlier experiments with the same subjects. The uptake rates ranged from 7 to 96 nmol/minute/cm^2 (0.83-/ 1.35 mg/minute/cm^2), with a geometric mean of 20 nmol/minute/cm^2 (2.36 mg/minute/cm^2). The results indicate that persons exposing large portions of their skin to 2-butoxyethanol are likely to absorb significant doses.

An *in vitro* study indicated that 2-butoxyethanol is absorbed through human abdominal epidermis (Dugard et al. 1984). Undiluted 2-butoxyethanol was applied to human abdominal epidermal membranes for a period of 8 hours. The absorption rate was 0.2 mg/cm^2/hour. In another *in vitro* study, a 10% solution of 2-butoxyethanol was applied to a 3- cm^2 area on frozen human arm skin, which was obtained at autopsy,

under semiocclusive or nonocclusive conditions (Bartnik et al. 1987). The percentage of absorption was 6.9% for the nonocclusive condition and 17.3% for the semiocclusive condition.

2-Butoxyethanol is also absorbed by animal skin. 2-Butoxyethanol was applied to the shaven skin of male and female rats on a 12- cm^2 area at a dose of 200 mg/kg (Bartnik et al. 1987). The application site was protected with a glass capsule to prevent oral uptake. Urine was collected at intervals of 0-8, 8-24, and 24-48 hours following application and stored at -80°C. Total radioactivity was determined for urine, cage rinse water, skin, and glass capsule. Percutaneous absorption was assessed by using the measurements of urinary excretion of ^{14}C from 0 to 48 hours following cutaneous application of 2-butoxyethanol. Radioactivity measured during the 48 hours following cutaneous application of radiolabeled 2-butoxyethanol showed that 20-23% of the applied dose was found in the urine, including cage rinse water, with no notable differences by sex. Over 95% of the radioactivity excreted in the urine was eliminated during the first 24 hours (metabolite not identified). Small amounts of radioactivity were found on the treated skin of male (4.7%) and female (8.3%) rats. Percutaneous absorption was determined to be 25-29% of the applied dose.

Percutaneous absorption was measured after three different doses of the [^{14}C]2-butoxyethanol (61, 181, or 299 mg/kg) were applied to same-sized areas on the clipped backs of Fischer 344/N rats; the sites of application were nonoccluded but were protected with a perforated covering (Sabourin et al. 1992b, 1993). The rates of excretion of the ^{14}C-labeled parent compound and metabolites by different routes were measured, as well as the amount of ^{14}C remaining in the carcass. Within the dose range studied, the absorption and metabolism of 2-butoxyethanol by Fischer 344/N rats was linearly related to the dermally applied dose. The absorption was approximately 21-26%, regardless of the dose administered. A small fraction of the applied dose (0.3-2%) was still present at the application site 72 hours following dosing, representing unabsorbed parent compound, dermally excreted parent compound, or metabolites in the skin. The remainder of the applied dose was recovered as volatilized radioactivity. The total recovery was 78-90%. The majority of the absorbed dose was excreted in the urine. Peak levels of radioactivity were reached at about 1 hour, with a half-life of about 4 hours.

The percutaneous absorption rate and elimination kinetics of 2-butoxyethanol were estimated in the guinea pig (Johanson and Femstrom 1986). An intravenous bolus dose of 5 or 11 mg/kg was administered into the jugular vein of 10 pentobarbital-anesthetized animals in order to calculate total clearance and mean residence time of the compound in guinea pigs. Following the intravenous dose, the apparent total clearance and mean residence time of 2-butoxyethanol in the blood were calculated to be 128 mL/minute/kg and 4.7 minutes,

respectively. At 2.5 hours after the intravenous dose, which was sufficient time for clearance of the intravenous dose, 1 mL neat 2-butoxyethanol was applied in two sealed glass rings on the clipped back of the animal for 2 hours. The concentration of 2-butoxyethanol in blood rose rapidly following the dermal application, and then approached a plateau level of 21 , µmol/L (2.482 mg/L) during the latter half of the exposure period. The absorption rate through the skin was estimated to be 0.25 (range, 0.05-0.46) µmol/minute/cm^2 (29.6 µg/minute/cm^2). The percutaneous uptake rate of 2-butoxyethanol in the present study was comparable to that obtained *in vitro* with human skin (i.e., 0.03 µmol/minute/cm [3.55 µg/minute/ cm^2]) once species differences were taken into account. The time lag of 21-60 minutes observed in the present study is in accordance with that reported for the penetration of 2-butoxyethanol through human skin *in vitro* (Dugard et al. 1984).

In *in vitro* experiments, frozen human skin from the flexus side of arms (obtained at autopsy), fresh dorsal skin of hairless rats, and frozen dorsal skin of pigs were used in a comparison absorption study of 2-butoxyethanol (Bartnik et al. 1987). Connective tissues and fat were removed. The solutions of [^{14}C]2-butoxyethanol were applied to a skin area of 3 cm^2 for human skin and 5 cm^2 for animal skin for 1 hour. The solutions (30 µL) of 2-butoxyethanol administered were as follows: 100% 2-butoxyethanol; 3.5% 2-butoxyethanol in water; 10% 2-butoxyethanol in water; 3.5% or 10% 2-butoxyethanol with 5% linear sodium dodecylbenzene sulfonate (LAS) in water; and 3.5% or 10% 2-butoxyethanol with 5% isopropanol in water. Both semioccluded and nonocclusive conditions were used. Absorption was determined after exposure durations of 1, 6, and 16 hours (semiocclusive) in rat skin 6 hours (semiocclusive) in pig skin, and 1 hour nonocclusive in rat skin. 2-Butoxyethanol at 3.5% (0.21 mg/ cm^2) in water was also tested after 10, 30, and 60 minutes under nonocclusive conditions in both rat and pig skin. Absorbed and nonabsorbed radioactivity levels were determined. In rat skin under semiocclusion, the applied dose of 2-butoxyethanol was rapidly absorbed (19% of 100% 2-butoxyethanol, 62.7% of 10% 2-butoxyethanol, and 45.6% of 3.5% 2-butoxyethanol in water at 1 hour) and after 16 hours was almost completely absorbed through the skin (94.3% of 100% solution, 82.6% of 10% solution, and 88.4% of 3.5% solution). The amount of penetration depended on time as well as concentration. The penetration of pure 2-butoxyethanol was initially slower than from aqueous solutions, but was more complete after 16 hours. In nonoccluded rat skin, 5.6% of the 100% 2-butoxyethanol, 10.4% of the 10% solution, and 11 .1% of the 3.5% solution were absorbed within 1 hour. In pig skin under semiocclusion, the applied dose of 2-butoxyethanol was less rapidly absorbed (2-3 times slower) than through rat skin. The penetration of pure 2-butoxyethanol was slower than from aqueous solutions and reached 11.2% after 6 hours, compared with 36.9% of the 10% solution and 47.5% of the 3.5% solution of 2-butoxyethanol. Isopropanol and LAS had little effect on absorption. In nonoccluded skin

treated with 3.5% 2-butoxyethanol, rat skin absorbed 10.4% in 10 minutes, 11.5% in 30 minutes, and 11.0% in 60 minutes, while pig skin absorbed 2.8% at 10 minutes, 6.6% at 30 minutes, and 5.4% at 60 minutes. In the comparison of *in vitro* skin penetration for humans, rats, and pigs, the data for 100 µg/cm^2 exposure for 1 hour were presented. For nonocclusive exposure, the percentages of absorption were 11% for rat skin, 8.6% for pig skin, and 6.9% for human skin. For semiocclusive exposure, the percentages of absorption were 43.3% for rat skin, 17.7% for pig skin, and 17.3% for human skin. Thus, the absorption pattern was: hairless rat>>>pig≥ human skin.

In a study on 2-butoxyethanol acetate in groups of six New Zealand rabbits to which the compound was applied dermally under occlusion for 24 hours, the doses of 3,319, 3,957, 4,766, 5,957, and 10,000 mg/kg were reported to correspond to absorbed doses (mg/kg±SE) of 610±310, 910±140, 1,130±390, 1,830±290, and 2,200±530 mg/kg, respectively (Truhaut et al. 1979). The absorbed doses were determined by comparing the amount of 2-butoxyethanol acetate applied under occlusion with the amount of 2-butoxyethanol acetate remaining after 24 hours of contact with the skin. The absorbed doses showed wide variability, which may be because this method is not very accurate.

2.3.2 Distribution

2.3.2.1 Inhalation Exposure

Some information describing distribution of 2-butoxyethanol and its major metabolite, 2-butoxyacetic acid after human inhalation exposure can be found in reports of controlled experiments. Venous blood samples were collected from five healthy male volunteers prior to and after inhalation exposure to 20 ppm 2-butoxyethanol during light physical exercise for 2 hours (Johanson and Johnsson 1991). 2-Butoxyacetic acid was found in all blood samples except those collected prior to exposure. Binding of 2-butoxyacetic acid to blood components is indicated by the low apparent volume of distribution of approximately 15 L, which is approximately equal to the volume of extracellular water, and the fact that the renal clearance rate is about one-third of the glomerular filtration rate. In another human study, seven male volunteers were exposed to 2-butoxyethanol for 2 hours at 20 ppm during light physical exercise on a bicycle ergometer (Johanson et al. 1986a). For 2-butoxyethanol, the apparent value of steady-state volume of distribution was 54 L and the mean residence time was 42 minutes. 2-Butoxyethanol was no longer detectable in the blood 2-4 hours after the end of exposure (Johanson et al. 1986a).

In animal studies, the body burden of [^{14}C]2-butoxyethanol following 6-hour nose-only inhalation exposure of male Fischer 344 rats was determined at several inhaled concentrations (Sabourin et al. 1992a). The uptake (4.49 µmol/ppm at 4.3 ppm; 4.58 µmol/ppm at 49 ppm; 3.58 µmol/ppm at 438 ppm) and metabolism (0.79 µmol/ppm at 4.3 ppm; 0.95 µmol/ppm at 49 ppm; 0.88 µmol/ppm at 438 ppm) of 2-butoxyethanol, expressed as [^{14}C]2-butoxyethanol equivalents, were essentially linear up to 438 ppm. Most (greater than 80%) of the [^{14}C]2-butoxyethanol-derived material in blood was in the plasma. 2-Butoxyacetic acid was the major metabolite of 2-butoxyethanol in plasma. Ratios of ethylene glycol to 2-butoxyacetic acid in plasma were higher than those in urine. The 2-butoxyethanol-derived ^{14}C in plasma rapidly became associated with the acid-precipitable (protein) fraction, probably because of binding of metabolites to proteins or incorporation of the 2-butoxyethanol metabolites into the carbon pool.

In a more recent study, eight male Sprague-Dawley rats were exposed to 20 or 100 ppm 2-butoxyethanol 24 hours per day for 1, 2, 3, 4, 6, 8, 10, or 12 days (Johanson 1994). Urine was collected at 24-hour intervals. At the end of the exposure period, animals were killed and blood, muscle, liver, and testis were collected. Blood and tissue samples were analyzed for 2-butoxyethanol and 2-butoxyacetic acid by electron capture gas chromatography. The concentrations of 2-butoxyethanol in blood, in the 20-ppm group were as follows: 7.5-29.0 µmol/L, with an average of 15.1 µmol/L; muscle, 0.9-13.0 µmol/kg, average of 9.1 µmol/kg; testis, 1.3-11.6 µmol/kg, with an average of 3.9 µmol/kg; and liver 5.1-16.4 µmol/kg, with an average of 10.8 µmol/kg. In the 100-ppm group, the concentrations were as follows: blood, 23.0-103 µmol/L, with an average of 72.3 µmol/L, muscle, 13.6-49.8 µmol/kg, with an average of 30.4 µmol/kg; testis, 4.7-36.1 µmol/kg, average, with an average of 12.6 µmol/kg; and liver, 42.4-129 µmol/kg, with an average of 83.8 µmol/kg. Although data points were reported on exposure days 1-12, the averages were calculated for 2-12 exposure days. The concentration of 2-butoxyethanol increased rapidly in the tissues during days l-3 and continued to increase, but more slowly, during the remaining days. Tissue concentrations in the 100-ppm group conformed to expected linear kinetics. Concentration ratios (100 ppm/20 ppm) were 5.1 for blood, 3.5 for muscle, 3.6 for testis, and 7.5 for liver. The kinetic pattern of 2-butoxyacetic acid was similar to 2-butoxyethanol. The concentrations in the 20-ppm group were as follows: blood, 31.8-48.8 µmol/L, with an average of 41.0 µmol/L; muscle, 5.8-14.9 µmol/kg, with an average of 9.3 µmol/kg; testis, 9.7-23.3 µmol/kg, with an average of 14.1 µmol/kg; and liver, 9.5-26.4 µmol/kg, with an average of 16.4 µmol/kg. In the 20-ppm group, the concentrations were as follows: blood, 125-226 µmol/L, with an average of 179 µmol/L; muscle, 17.9-63.6 µmol/g, with an average of 36.2 µmol/kg; testis, 11.6-48.5 µmol/kg, with an average of 26.7 µmol/kg; and liver, 52.8-107 µmol/kg, with an average of 85.2 µmol//kg. Again, the data points were reported on exposure

days l-l 2, but the averages were calculated for 2-12 exposure days. As with 2-butoxyethanol, the 2-butoxyacetic acid concentration increased rapidly in the tissues during the first l-3 days and then more slowly during the remaining days. Tissue concentrations in the 100-ppm group conformed to expected linear kinetics. Concentration ratios (100 ppm/20 ppm) were 4.4 for blood, 4.7 for muscle, 2.3 for testis, and 5.3 for liver. Thus, the disposition of 2-butoxyethanol and 2-butoxyacetic acid was linear at the exposure concentrations used, and although the number of tissues examined was limited, the data indicated extensive distribution.

2.3.2.2 Oral Exposure

Studies in animals indicate that distribution of 2-butoxyethanol after oral exposure is rapid (Ghanayem et al. 1987c). Male Fischer 344 rats were given a single oral dose of [^{14}C]2-butoxyethanol of 125 or 500 mg/kg. Rats were placed in metabolism cages and monitored for 48 hours after treatment. At 48 hours, animals were killed and the ^{14}C -radioactivity in each tissue, as well as in the blood, was determined. Tissue distribution of 2-butoxyethanol revealed that 2-butoxyethanol is distributed to all tissues, with the highest levels (determined 48 hours after dosing) detected in the forestomach (654 nmol/g at 125 mg/kg and 7,606 nmol/g at 500 mg/kg). The increase in the tissue concentration in rats treated with 500 mg/kg (as compared to that in rats treated with 125 mg/kg) was not proportional to the increase in dose. While the ratio of the administered doses was 1:4, the ratio of the tissue concentration in most tissues was greater than 1: 10. This may be related to saturation of 2-butoxyethanol metabolism at the high dose. There seems to be a positive correlation between tissue levels and the tissues exhibiting toxicity. Other levels were as follows: liver, 107 nmol/g at 125 mg/kg and 1,666 nmol/g at 500 mg/kg; skin, 75 nmol/g at 125 mg/kg and 806 nmol/g at 500 mg/kg; kidney, 69 nmol/g at 125 mg/kg and 1,304 nmol/g at 500 mg/kg; lung, 68 nmol/g at 125 mg/kg and 1,352 nmol/g at 500 mg/kg; the glandular stomach, 61 nmol/g at 125 mg/kg and 1,743 nmol/g at 500 mg/kg; spleen, 54 nmol/g at 125 mg/kg and 1,149 nmol/g at 500 mg/kg; fat, 40 nmol/g at 125 mg/kg and 162 nmol/g at 500 mg/kg; heart, 36 nmol/g at 125 mg/kg and 927 nmol/g at 500 mg/kg; blood, 34 nmol/g at 125 mg/kg and 1,288 nmol/g at 500 mg/kg; testes, 22 nmol/g at 125 mg/kg and 659 nmol/g at 500 mg/kg; and muscle, 17 nmol/g at 125 mg/kg and 571 nmol/g at 500 mg/kg.

In a companion study in male Fischer 344 rats, 2-butoxyethanol was given orally by gavage at 500 mg/kg alone or after pretreatment with pyrazole, which was found to inhibit the metabolism of 2-butoxyethanol to 2-butoxyacetic acid (Ghanayem et al. 1987b) (see Section 2.3.3). The levels of 2-butoxyethanol in tissues measured 48 hours after dosing with 2-butoxyethanol alone were those reported above by Ghanayem et al.

(1987c) for the 500-mg/kg dose. In rats pretreated with pyrazole followed by 500 mg/kg [^{14}C]2-butoxyethanol, the levels of 2-butoxyethanol-derived radioactivity in these tissues were appreciably lower: 2,384 nmol/g in forestomach, 417 nmol/g in liver, 144 nmol/g in skin, 282 nmol/g in kidney, 231 nmol/g in lung, 380 nmol/g in glandular stomach, 303 nmol/g in spleen, 120 nmol/g in fat, 102 nmol/g in heart, 135 nmol/g in blood, 104 nmol/g in testes, and 65 nmol/g in muscle (Ghanayem et al. 1987b). Analysis of radioactivity in the liver revealed that 2-butoxyacetic acid was the only radioactive species. These observations, along with the effects of pyrazole on the metabolic pathway (see Section 2.3.3) and excretion (see Section 2.3.4.2) of 2-butoxyethanol, indicated that pyrazole enhanced the elimination of 2-butoxyethanol and protected the rats from its hematoxic effects.

2.3.2.3 Dermal Exposure

Three different amounts of the [^{14}C]2-butoxyethanol (61, 181, or 299 mg/kg) were applied to same-sized areas on the clipped backs of Fischer 344/N rats, and nonoccluded percutaneous absorption was measured (Sabourin et al. 1992b, 1993). A small amount of the applied dose (0.3-2%) was still present at the application site 72 hours following dosing, representing unabsorbed parent compound, absorbed parent compound, or metabohtes in the skin. The majority of the absorbed dose was excreted in the urine. At 72 hours, 7-l 6% of the absorbed radioactivity remained in the carcass. Over 80% of the blood radioactivity was associated with the plasma, and less than 20% was associated with the red blood cells.

2.3.2.4 Other Routes of Exposure

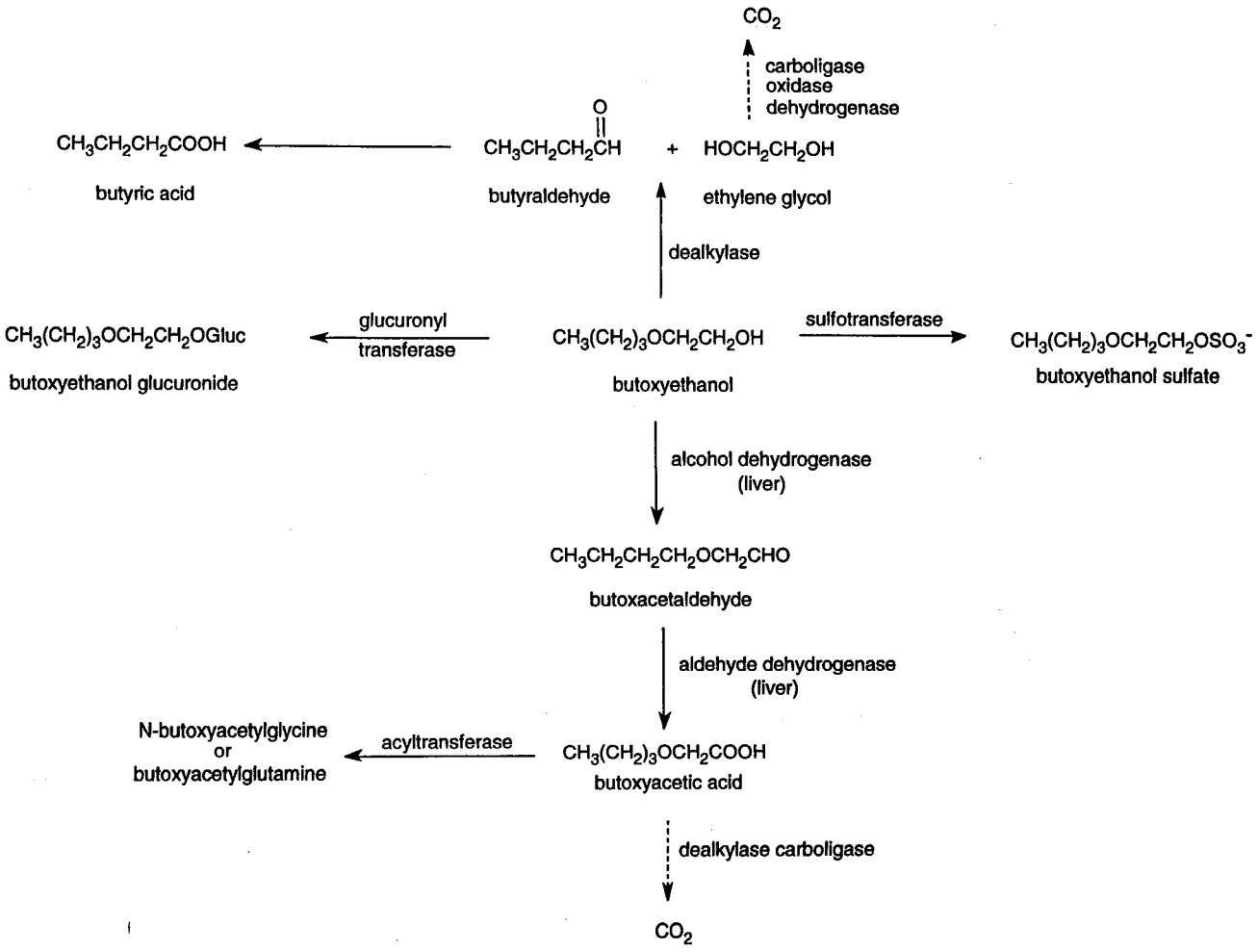
After injection of male Wistar rats in the scapular region with a single subcutaneous dose of 118 mg/kg [^{14}C]2-butoxyethanol, not exceeding 99 μCi/kg, the tissue distribution of radioactivity was determined (Bartnik et al. 1987). With the exception of plasma, all organs showed a higher relative specific radioactivity than blood 72 hours after dosing. If the specific radioactivity found in blood is given a value of 1, values for specific radioactivity (relative to blood) were as follows: 3.02 in liver, 1.99 in kidney, 6.62 in spleen, 1.55 in testis, 7.39 in thymus, 0.93 in plasma, 1.14 in carcass, 2.74 in fat, and 1.25 in sternum. However, the percentages of the applied dose of radioactivity in these tissues were reported to be 0.49% in liver, 0.07% in kidney, 0.06% in spleen, 0.08% in testis, 0.06% in thymus, 0.16% in plasma, 4.05% in carcass, and 1.60% in fat. The percentage of applied radioactivity in blood was 0.32%. The percentage of applied radioactivity in the sternum was not calculated.

2.3.3 Metabolism

There is no available evidence to suggest that the route of administration has any substantial effect on the subsequent metabolism of 2-butoxyethanol in humans. However, data from animals indicate that route of exposure may make some difference in the relative amounts of metabolites produced (Medinsky et al. 1990; Sabourin et al. 1992a, 1992b, 1993). Data regarding metabolism of 2-butoxyethanol have been obtained from a large volume of work done on other glycol ethers with specific experiments carried out on 2-butoxyethanol (summarized in EPA 1984; Miller 1987; NIOSH 1990). The metabolic scheme for 2-butoxyethanol is shown in Figure 2-6. 2-Butoxyethanol is oxidized in the liver by alcohol dehydrogenase to butoxy-acetaldehyde, which is further oxidized to 2-butoxyacetic acid by acetaldehyde dehydrogenase. In animals and humans, 2-butoxyacetic acid can be conjugated with glycine to form *N*-butoxyacetylglycine, or it can be broken down to CO_2 (Ghanayem et al. 1987b; Marxhall 1982; Medinsky et al. 1990; Rettenmeier et al. 1993). 2-Butoxyethanol can also be dealkylated by CYP2El to form ethylene glycol and butyraldehyde; ethylene glycol is then broken down to oxalic acid and ultimately to CO_2. Rettenmeier et al. (1993) identified an additional amino acid conjugate, *N*-butoxyacetylglutamine, in human urine samples, which accounted for an average of 50% of the total 2-butoxyacetic acid detected; the remainder was free. The formation of the *N*-butylacetylglutamine conjugate in humans has been confirmed in humans exposed (one arm only) to 2-butoxyethanol; 213 of total urinary 2-butoxyacetic acid was conjugated with glutamine (Corley et al. 1997). In rats, 2-butoxyethanol can also be detoxified by conjugation with sulfate to form butoxyethanol sulfate, or with glucuronic acid to form the glucuronide (Ghanayem et al. 1987b; Ghanayem et al. 1987c). Clinical reports and animal experiments that have contributed to the elucidation of this metabolic scheme are described below. Corley et al (1997) found no free or conjugated ethylene glycol or its metabolite, glycolic acid, in the urine.

A report on a woman admitted to the hospital after ingestion, in a suicide attempt, of 250-500 rnL, of a window cleaner containing 12% (467-933 mg/kg) 2-butoxyethanol indicated that metabolic acidosis, hypokalemia, a rise in serum creatinine level, and a markedly increased urinary excretion of oxalate crystals were observed, suggesting a metabolic pattern after absorption from the stomach (Rambourg-Schepens et al. 1988). The high output of the oxalate in the urine was explained by the metabolism of 2-butoxyethanol to ethylene glycol, which was further metabolized to oxalic acid. This assumption was supported by the existence of metabolic acidosis coincidental with the urinary peak oxalate level; however, 2-butoxyacetic acid can also cause metabolic acidosis. Similar metabolic effects have been reported by others (Bauer et al. 1992; Gijsenbergh et al. 1989).

Figure 2-6. Scheme for Metabolism of 2-Butoxyethanol*



*Derived from the work of Ghanayem et al. 1987b; Marshall 1982; Medinsky et al. 1990

Venous blood samples were collected from five healthy male volunteers prior to and after inhalation exposure to 20 ppm 2-butoxyethanol (the Swedish permissible exposure limit) for 2 hours under light physical exercise (Johanson and Johnsson 1991). Blood samples were collected at 0, 2, 4, and 6 hours from the start of exposure and analyzed by gas chromatography. The minimum and maximum observed concentrations of free 2-butoxyacetic acid in blood ranged from 20 to 57 µM (2.6-7.5 mg/L) with average concentrations of 22, 44, 39, and 28 µM at 1, 3, 4.9, and 7.1 hours, respectively. Free 2-butoxyacetic acid was found in all samples except those collected prior to exposure. These concentrations were about two orders of magnitude lower than those causing swelling and hemolysis of human erythrocytes *in vitro* (Ghanayem 1989). The 2-butoxyacetic acid blood level peaked after 2-4 hours. The decrease between 4-6 hours indicates an average half-time of 2-butoxyacetic acid in blood of about 4 hours, which is in accordance with half-times in urine observed by Johanson et al. (1986a) in humans similarly exposed (see Section 2.3.4.1). An interfering peak with a retention time similar, but not identical, to 2-butoxyacetic acid was found in all samples with the area of the peak corresponding to 1.8 µM 2-butoxyacetic acid. 2-Butoxyacetic acid was also detected in the urine of seven male subjects exposed to 20 ppm of 2-butoxyethanol for 2 hours (Johanson et al. 1986a). These subjects excreted 15.2-55% of the respiratory uptake in the urine.

The effect of genetic polymorphism of CYP 2El was studied in 32 male workers exposed to 2-butoxyethanol (Haufroid et al. 1997). Only one worker expressed the c2 allele (heterozygote c1/c2), while the other 30 workers were homozygous for c1. The heterozygous worker was the only worker for whom urinary 2-butoxyethanol acid was similar before and after his work shift, although he was exposed to 0.53 ppm 2-butoxyethanol. The study author indicated that the reduced 2-butoxyacetic acid excretion in their heterozygous subject was likely a result of increased activity of the c2 compared to the c1 allele; however, no firm conclusions can be drawn since only free 2-butoxyacetic acid was measured and it is therefore unknown how much conjugated 2-butoxyacetic acid was in the urine.

In an early study, rats, guinea pigs, dogs, and rabbits exposed to 100-400 ppm 2-butoxyethanol for 4 hours excreted free 2-butoxyacetic acid in the urine (Carpenter et al. 1956). Dogs and monkeys exposed to 100-398 ppm 2-butoxyethanol for 28-90 days also excreted free 2-butoxyacetic acid in the urine. These data indicate that the metabolism of 2-butoxyethanol to 2-butoxyacetic acid is a common pathway in mammalian species.

Other studies in animals also indicate that 2-butoxyethanol is metabolized to 2-butoxyacetic acid (Ghanayem et al. 1987c). Male Fischer 344 rats were given a single oral dose of [^{14}C]2-butoxyethanol at 125 or

500 mg/kg. Rats were placed in metabolism cages and monitored for 48 hours after treatment. Urine, feces, and bile were collected for metabolite analysis. Urinary metabolites detected at any one time point included free 2-butoxyacetic acid as the primary constituent (74-100%), the 2-butoxyethanol-glucuronide (0-24%), and a minor amount of the 2-butoxyethanol-sulfate conjugate (0-3%) detected only at the lower dose. In the animals treated with 500 mg/kg 2-butoxyethanol, a larger proportion of the administered dose was detected as the glucuronide, suggesting that the enzymes were saturated in the high-dose animals. In the bile, flow increased significantly at 0.5 hour after treatment and remained above pretreatment levels for the following 4 hours. Biliary metabolites included free 2-butoxyacetic acid, the 2-butoxyethanol-glucuronide, and the parent compound. The parent compound was detected only in the first 2 hours after treatment. In contrast to the urine, the 2-butoxyethanol-glucuronide was the major metabolite in the bile, while 2-butoxyacetic acid increased with time at the expense of the glucuronide.

In studies designed to investigate the metabolic basis of 2-butoxyethanol-induced hematotoxicity in male Fischer 344 rats, treatment with pyrazole, an alcohol dehydrogenase inhibitor, protected the rats against 2-butoxyethanol hematotoxicity and prevented the metabolism of 2-butoxyethanol to 2-butoxyacetic acid (Ghanayem et al. 1987b). Inhibition of 2-butoxyetbanol metabolism to 2-butoxyacetic acid was accompanied by an increase in the production of the glucuronide and sulfate conjugates of 2-butoxyethanol. Rats treated with pyrazole and 2-butoxyethanol exhibited a 10-fold decrease in the proportion of free urinary 2-butoxyacetic acid to 2-butoxyethanol-glucuronide and 2-butoxyethanol-sulfate (total production), compared to rats treated with 2-butoxyethanol alone. Pretreatment of rats with cyanamide, an aldehyde dehydrogenase inhibitor, also protected the animals against the hematotoxicity and modified 2-butoxyethanol metabolism in a manner similar to that with pyrazole. Further experiments indicated that hematotoxicity could result from treatment with 2-butoxyethanol, the metabolic intermediate butoxyacetaldehyde, or the ultimate toxic metabohte 2-butoxyacetic acid. Cyanamide also served to protect from the butoxyacetaldehyde-induced toxicity. Pyrazole significantly increased bile flow, even more than 2-butoxyethanol alone, and excretion of radioactivity from 2-(l-[^{14}C]butoxy)ethanol in bile was higher in rats pretreated with pyrazole (16%) than in rats given 2-butoxyethanol alone (8%). The composition of biliary metabolites also differed in animals treated with 2-butoxyethanol alone compared with animals treated with 2-butoxyethanol in conjunction with pyrazole. Free 2-butoxyacetic acid constituted 10%, 21%, and 46% of the total radioactivity excreted in the bile at 0-l, 2-4, and 6-8 hours, respectively, after dosing with 2-butoxyethanol alone. In contrast, no 2-butoxyacetic acid was detected in any bile fractions from animals treated with pyrazole. In these rats, approximately 12% of the radioactivity was excreted in the 1st hour as unchanged 2-butoxyethanol, with the remaining portion being the glucuronide. No other metabolites were detected in the bile. Further evidence of

the requirement for metabolic activation prior to the onset of hematotoxicity was observed with the administration of deuterium-labeled 2-butoxyethanol. The onset of hematotoxicity was significantly delayed, indicating the slower oxidation of 2-butoxyethanol due to the deuterium substitution in place of the somewhat smaller hydrogen atoms. This study provided conclusive evidence of the following: there is a good correlation between the 2-butoxyethanol-induced hematotoxicity and the amount of free 2-butoxyacetic acid in the urine; alcohol and aldehyde dehydrogenase are essential to the metabolic activation of 2-butoxyethanol to 2-butoxyacetic acid; and hematotoxicity induced by 2-butoxyethanol can be attributed to its metabolite, 2-butoxyacetic acid.

Experiments designed to determine the effect of age on the toxicity and metabolism of 2-butoxyethanol indicate that in rats, young animals (4-5 weeks old) are more resistant to the toxic effects than are older rats (9-13 weeks old) receiving an equivalent dose (Ghanayem et al. 1987a). Young rats excreted less free 2-butoxyacetic acid than adult rats, and more 2-butoxyethanol-glucuronide. No sulfate conjugate of 2-butoxyethanol was detected in the urine of young rats. High-performance liquid chromatographic analysis of the urinary metabolites of 2-butoxyethanol in adult and young rats showed that the ratio of free 2-butoxyacetic acid to the total amount of the glucuronide and sulfate conjugates of 2-butoxyethanol (previously thought to reflect an activation/detoxification index of 2-butoxyethanol) was significantly higher in older rats. An unknown metabohte was found in the urine of both young and adult rats in amounts of 6-8% and 1%, respectively. Additional studies on the effect of dose, age, and inhibition of metabolism on the toxicity of 2-butoxyethanol indicated that total metabolism was dose dependent, but there was no effect of dose on half-life or volume of distribution (Ghanayem et al. 1990a). There was no effect of age on half-life, volume of distribution, or clearance of 2-butoxyethanol. Inhibition of metabolism of 2-butoxyethanol with pyrazole (inhibition of alcohol dehydrogenase) or cyanamide (inhibition of aldehyde dehydrogenase) significantly increased the half-life and decreased the clearance. Although pyrazole had no effect on volume of distribution, cyanamide pretreatment caused a decrease in the volume of distribution for 2-butoxyethanol; the reason for this was not clear. Analysis of toxicokinetic parameters of free 2-butoxyacetic acid indicated that the half-life and maximum plasma concentration were directly related to the age of the animals and the dose of 2-butoxyethanol. Pyrazole decreased the maximum plasma concentration and half-life of free 2-butoxyacetic acid, presumably because of inhibition of metabolism of 2-butoxyethanol. Cyanamide increased the half-life and decreased the plasma concentration of free 2-butoxyacetic acid. The study authors noted that the effects of cyanamide are probably related to the inhibition of the conversion of butoxyacetaldehyde to 2-butoxyacetic acid, but the reason that cyanamide increased the half-life of 2-butoxyacetic acid is unclear. Probenecid, an inhibitor of renal transport of organic acids, did not alter the plasma concentration,

volume of distribution, or clearance of free 2-butoxyethanol, but it did increase the half-life of free 2-butoxyacetic acid.

In an effort to identify underlying mechanisms associated with the development of tolerance to the hemolytic effects of 2-butoxyethanol, male Fischer 344 rats were treated with 125 mg/kg/day of unlabeled 2-butoxyethanol for 3 or 7 days by gavage followed by a single dose of 125 mg/kg of [^{14}C]2-butoxyethanol (40-50 µCi/kg) administered on day 4 or day 8, respectively (Ghanayem et al. 1992). Three untreated rats received the same dose of [^{14}C]2-butoxyethanol(40-50 µCi /kg). Tissue disposition and urine analyses of 2-butoxyethanol metabolites were performed. No quantitative or qualitative alterations of 2-butoxyethanol metabolism or its disposition were caused by repeated exposure to 2-butoxyethanol as compared to treatment with a single dose. The ratios of free 2-butoxyacetic acid, the 2-butoxyethanol-glucuronide and 2 butoxyethanol-sulfate conjugates, and parent 2-butoxyethanol excreted in the urine of rats treated for 4 or 8 days were relatively similar to those from rats treated with a single dose.

Male Fischer 344/N rats were allowed access for 24 hours to uniformly labeled [^{14}C]2-butoxyethanol in drinking water at three doses (Medinsky et al. 1990). Elimination of radioactivity was monitored for 72 hours. The labeled 2-butoxyethanol was administered at doses of 290, 860, or 2,590 ppm in drinking water, resulting in consumption of 237, 401, or 1,190 µmol/kg/body weight (28, 47, or 141 mg/kg/day). The majority of the ^{14}C was excreted in urine, with 50-60% of the dose as free 2-butoxyacetic acid, or exhaled as CO_2 (8-10% of the dose); less than 5% of the dose was exhaled as unmetabolized 2-butoxyethanol. Ethylene glycol was excreted in urine, representing approximately 10% of the dose of 2-butoxyethanol. Unmetabolized 2-butoxyethanol and its glucuronide conjugate were also identified in the urine of animals dosed with 2-butoxyethanol.

A metabolism study was conducted to assist in validation of a PBPK model for 2-butoxyethanol kinetics (Corley et al. 1994). Two groups of male Fischer 344 rats with indwelling jugular cannulae received radiolabeled 2-butoxyethanol at doses of 8.6 and 126 mg/kg by gavage in water. Animals were immediately placed in metabolism cages for collection of urine, feces, and CO_2. Samples of blood were taken from each rat at 1, 3, 6, 12, and 24 hours postdosing for determination of 2-butoxyethanol and free 2-butoxyacetic acid. Urine was collected at intervals of 0-12 and 12-24 hours postdosing. Urine samples were analyzed for total radioactivity and metabolite profile. The overall disposition of 2-butoxyethanol in this study was similar to that reported by Ghanayem et al. (1987c). At the low dose, approximately 59% of the dose was excreted in the urine during the first 24 hours, and 7.2% was excreted as CO_2. At 126 mg/kg, 38-70% of the dose was

eliminated in the urine, and 8% was eliminated as CO_2. Metabolite profiles in the urine were similar to those reported by Ghanayem et al. (1987c) and Medinsky et al. (1990), and indicated that nearly 40% of the radioactivity in the urine was present as free 2-butoxyacetic acid during the first 12 hours following administration of 8.6 mg/kg, and nearly 65% after 126 mg/kg. An additional 15% of the radioactivity in the urine was present as the glucuronide conjugate of 2-butoxyethanol at the lower dose, and 10% was present at the higher dose. There was no evidence of further conjugation of 2-butoxyacetic acid under the enzyme/ hydrolysis conditions used in this study. Ethylene glycol was present in smaller quantities (2-l 1% of the total radioactivity). An explanation of the analytic preparation is provided in Chapter 6.

The metabolism of 2-butoxyethanol following 6-hour nose-only inhalation exposure of male Fischer 344 rats was determined at different inhaled concentrations (Sabourin et al. 1992a). The uptake and metabolism of 2-butoxyethanol were essentially linear up to 438 ppm. From 17% to 24% of the inhaled compound was metabolized. Most (64-76%) of the inhaled [^{14}C]2-butoxyethanol was eliminated in the urine, with free 2-butoxyacetic acid being the major urinary metabolite (37-43%), accompanied by lesser amounts of ethylene glycol(8-16%) and 2-butoxyethanol glucuronide (3-10%). A small proportion (5-8%) of the retained 2-butoxyethanol was exhaled as $^{14}CO_2$. Most (>80%) of the [^{14}C]2-butoxyethanol-derived material in blood was in the plasma. Free 2-butoxyacetic acid was the major metabolite of 2-butoxyethanol in plasma. Ratios of ethylene glycol to 2-butoxyacetic acid in plasma were higher than those in urine. Urinary metabolites were monitored at various times up to 66 hours postexposure. More than 88% of the total radioactivity excreted was excreted during the first 41 hours. Free 2-butoxyacetic acid was the major metabolite in the urine at all exposure concentrations; ethylene glycol and 2-butoxyethanol glucuronide were also found in lesser amounts. As the exposure concentration increased, the proportion of two unidentified minor metabolites increased, and the time required for urinary excretion of [^{14}C]2-butoxyethanol equivalents also increased. At 49 ppm, metabolism to the glucuronide was favored during exposure, whereas metabolism to free 2-butoxyacetic acid and ethylene glycol was favored at later times postexposure. The pattern was even more apparent at the higher dose. CO_2 production most closely followed ethylene glycol production. The study authors suggested that these results indicate that, in rats, overall metabolism of 2-butoxyethanol to 2-butoxyacetic acid, the hemolytic metabolite, was linearly related to the exposure concentration up to a concentration that caused severe toxicity (438 ppm). Assuming that the toxicity of inhaled 2-butoxyethanol is directly proportional to the formation of 2-butoxyacetic acid, the authors suggested that the toxicity of inhaled 2-butoxyethanol can be expected to be linearly related to the exposure concentration up to exposure concentrations that cause mortality.

Three different amounts of [^{14}C]2-butoxyethanol (61, 181, and 288 mg/kg) were applied to same-sized areas on the clipped backs of Fischer 344/N rats, and nonoccluded percutaneous absorption was measured (Sabourin et al. 1992b, 1993). The rates of excretion by different routes of the ^{14}C -labeled parent compound and metabolites were measured, as well as the amount of ^{14}C remaining in the carcass. Within the dose range studied, the absorption and metabolism of 2-butoxyethanol by Fischer 344/N rats were linearly related to the dermally applied dose. Free 2-butoxyacetic acid was the major metabolite. The 2-butoxyethanol-glucuronide was also detected in the urine. The formation of small amounts of ethylene glycol indicated cleavage of the ether bond. Plasma metabolites were evaluated at the mid-dose, in which no significant hemolysis was detected. Over 80% of the blood radioactivity was associated with the plasma and less than 20% was associated with the red blood cells. The concentration of total plasma metabolites was nine times higher than that of the parent compound, 2-butoxyethanol, in the plasma. Peak levels of radioactivity were reached at about 1 hour, with a half-life of about 4 hours. Free 2-butoxyacetic acid was the major metabolite and represented 53-75% of the plasma radioactivity.

Comparison of the profiles of urinary metabolites from inhalation, oral, and dermal exposure of rats to 2-butoxyethanol at low, middle, and high doses suggests that the route of administration influences the relative amount of metabolites (Medinsky et al. 1990; Sabourin et al. 1992a, 1992b, 1993; Shyr et al. 1993). In all exposure routes, free 2-butoxyacetic acid is the major metabolite of 2-butoxyethanol, but the relative proportions of minor metabolites can be influenced by route of exposure. Exposure by oral (drinking water) or inhalation routes generally favors production of ethylene glycol as compared to the glucuronide, with the exception of low-level drinking water exposure and the high-dose inhalation exposure, after which the amounts of the two metabolites were not statistically different. Dermal exposure favors production of the 2-butoxyethanol-glucuronide, possibly because of the more rapid administration of 2-butoxyethanol by this route, which in turn saturates the metabolic pathway and shift metabolism to 2-butoxyethanol-glucuronide conjugation.

The metabolism of 2-butoxyethanol (^{14}C -labeled at both ethanol carbons) by human and rat hepatocytes has been studied *in vitro* (Green et al. 1996). Following a 4-hour incubation period, rat hepatocytes produced more ethylene glycol and more free 2-butoxyacetic acid at all concentrations tested (0.02, 0.2, 2, and 10 mM). As indicated in Table 2-8, there was evidence of saturation of the ethylene glycol pathway in both species at 0.2 mM, and of the 2-butoxyacetic acid pathway in rats at 10 mM, and in humans at 2 mM. The glucuronide conjugate of 2-butoxyethanol was tentatively identified in the incubation media of both species. Kinetic parameters, calculated using Eadie-Hofsee plots, were maximum velocity (V_{max}) of 113±58 and

Table 2-8. Metabolism of 2-Butoxyethanol by Human and Rat Hepatocytes *In Vitro*[a,b]

Concentration (mM)	Human hepatocytes % of total radioactivity as:			Rat hepatocytes % of total radioactivity as:		
	2-Butoxyethanol[c]	2-Butoxyacetic[d] acid	Ethylene glycol	2-Butoxyethanol[c]	2-Butoxyacetic[d] acid	Ethylene glycol
0.02	40.8±19.1	51.8±18.6	4.4±1.9	<0.4	77.3±0.4	8.6±1.5
0.2	54.7±18.6	42.2±18.5	3.1±1.8	0.3±0.5	91.5±1.4	5.9±1.0
2	83.3±8.5	14.5±8.3	1.6±0.9	7.5±9.3	82.6±6.1	4.2±1.2
10	93.1±2.0	4.6±1.1	1.5±0.8	66.1±8.0	28.2±3.8	2.6±1.3

[a]Values are the amounts detected after 4 hours of incubation and are the mean and standard deviation of four experiments for humans and three experiments for rats.
[b]Derived from Green et al. (1996).
[c]Values include the gluceronide conjugate of 2-butoxyethanol.
[d]Values are free 2-butoxyacetic acid.

74 1±265 nmol/hour/10^6 hepatocytes for humans and rats, respectively, and metabolism rate constants (K_m) of 0.9±0.3 and 0.9±0.5 mM for humans and rats, respectively.

The metabolism of 2-butoxyethanol acetate has not been extensively studied, presumably because 2-butoxy-ethanol acetate is assumed to be metabolized to 2-butoxyethanol in the body, and then to follow the same pathway as 2-butoxyethanol, since 2-butoxyacetic acid has been detected in human urine after 2-butoxyethanol acetate exposure (Johanson et al. 1989).

2.3.4 Elimination and Excretion

2.3.4.1 Inhalation Exposure

Elimination and excretion of 2-butoxyethanol after inhalation exposure appears to be similar in both humans and animals. Human studies suggest that excretion of 2-butoxyacetic acid is variable. Volunteers were exposed to 98 or 195 ppm 2-butoxyethanol for 8 hours (Carpenter et al. 1956). Urinalyses for free 2-butoxyacetic acid were conducted on urine samples collected during the 24 hours following the end of the exposure period. Results showed that at the 195-ppm exposure level, one male excreted 175 mg and one female excreted 300 mg of 2-butoxyacetic acid in the 24-hour period. Another male exposed to 195 ppm excreted only trace amounts of 2-butoxyacetic acid. Subjects exposed to 98 ppm (n=4) showed the following values for excreted 2-butoxyacetic acid: 100 mg (one female), 183 mg (one female), 75 mg (one male), 250 mg (one male). Since a substantial fraction of butoxyacetic acid can be excreted in the urine as the amino acid conjugate, some of the observed variability could come from the proportion conjugated.

Seven male volunteers were exposed to 2-butoxyethanol for 2 hours at the Swedish occupational exposure limit (20 ppm or 0.85 mmol/ m^3) during light physical exercise on a bicycle ergometer (Johanson et al. 1986a). Expired air was collected at regular time intervals for estimation of the respiratory uptake of the solvent. Capillary blood and urine were sampled, during and after the exposure period, and analyzed for 2-butoxyethanol and its metabolite, free 2-butoxyacetic acid. 2-Butoxyethanol was no longerdetectable in the blood 2-4 hours after the end of exposure. The apparent values of elimination half-time, mean residence time, total blood clearance, and steady-state volume of distribution were 40 minutes, 42 minutes, 1.2 L/minute, and 54 L, respectively, for 2-butoxyethanol. The half-life for 2-butoxyethanol in urine was 1.36 hours. The amount of 2-butoxyethanol excreted in urine was less than 0.03% of the total uptake, while that of 2-butoxyacetic acid ranged from 15% to 55%. The excretion rate of free 2-butoxyacetic acid in urine

as well as the concentration varied more than 10-fold among subjects. There were increases in the concentra-tion and excretion rate during the first few hours, with maximums reached after 5-12 and 2-10 hours, respectively. The half-life of 2-butoxyacetic acid in urine was 5.77 hours. The total urinary excretion averaged 496 µmol. Relative to the uptake of 2-butoxyethanol, the excretion averaged 41% on an equimolar basis.

Venous blood samples were collected from five healthy male volunteers prior to and after inhalation exposure to 20 ppm 2-butoxyethanol (the Swedish permissible exposure limit) during light physical exercise for 2 hours (Johanson and Johnsson 199 1). The low renal clearance of free 2-butoxyacetic acid (22-39 mL/minute) compared with the average glomerular filtration rate of about 125 mL/minute indicates extensive binding to blood proteins and poor tubular secretion of the substance. Binding of 2-butoxyacetic acid to blood components is also indicated by the low apparent volume of distribution of approximately 15 L, which is about the volume of the extracellular water compartment. The study authors applied a one-compart-ment model for the kinetics of 2-butoxyacetic acid in blood with an average half-life of 4.3 hours, extrapolated to 1 workday (8 hours) of continuous exposure, and predicted a blood level of 114 µmol (15.1 mg/L) and a steady-state of 157 µmol/(20.7 mg/L). The authors suggested that if dermal contact is avoided, inhalation exposure to 20 ppm 2-butoxyethanol, even for extended periods of time, is unlikely to cause adverse effects in humans.

Individual exposure to 2-butoxyethanol acetate was measured by personal air sampling (air samples collected from the breathing zone of workers and samples analyzed by gas chromatography) and biological monitoring of urine from 19 employees in an 8-hour work day in four silk-screen printing installations (Johanson et al. 1989). The samples were analyzed by gas chromatography. Urine was quantitatively collected from each subject two or three times during the day. Free 2-butoxyacetic acid could be detected in the low FM range in the personal air samples and in the urine. 2-Butoxyethanol acetate was detected in the personal air samples of five individuals, with an average concentration of 0.44 ppm. The detection limit was 0.015 ppm in air. Free 2-butoxyacetic acid was detected in the urine of 12 individuals, at an average concentration of 8 µM (0.132 mg/L).

Studies have addressed the issue of excretion of conjugated forms of 2-butoxyacetic acid in the urine of exposed individuals (Rettenmeier et al. 1993; Sakai et al. 1994). In the study by Rettenmeier et al. (1993), urine samples (end of shift on Friday and prior to shift on the following Monday) were taken from six lacquer workers who used a 2-butoxyethanol-containing detergent to clean car body parts. Samples from the end of

shift on Friday contained relatively large amounts of both 2-butoxyacetic acid and its glutamine conjugate, whereas only trace amounts of these metabolites were detectable in the Monday samples. The end of shift Friday urinary concentrations of 2-butoxyacetic acid averaged 1.49 mmolL (range: 0.13-5.9 1 mmol/L), and the urinary concentrations of the glutamine conjugate averaged 0.82 mmol/L (range: 0.12-2.45 mmol/L). The fraction of 2-butoxyacetic acid that was excreted in the urine as the amino acid conjugate, *N*-butoxyacetylglutamine, averaged 48% (16-64%) of the total 2-butoxyacetic acid excreted. Thus, the urinary excretion of 2-butoxyacetic acid (up to 45-fold) and the glutamine conjugate (up to 20-fold) varied widely in the six subjects, even though their exposure conditions were similar. In a similar study, Sakai et al. (1994) compared 2-butoxyacetic acid levels in the urine of workers exposed to 2-butoxyethanol, both with and without acid hydrolysis to convert conjugated metabolites to free 2-butoxyacetic acid. Conjugated metabolites accounted for 71% (44-92%) of the total 2-butoxyacetic acid found in the urine over a Monday through Friday work week. Sakai et al. (1994) attributed the differences in their data compared with the findings of Rettenmeier et al. (1993) to differences in the derivatization methods and the possible differences in the 2-butoxyethanol exposure concentrations. Furthermore, Sakai et al. (1994) found that the fraction excreted as the conjugate during the first 2 days of the work week was greater than the amount excreted during the remainder of the work week, indicating a gradual depression of metabolic capacity of 2-butoxyacetic acid conjugation over subsequent work days. Variability in the rate of conjugate formation probably accounts for the large variation in excretion of free 2-butoxyacetic acid observed in earlier studies of persons exposed to 2-butoxyethanol. In order to determine the amount of total 2-butoxyacetic acid, it is necessary to measure both free butoxyacetic acid as well as the conjugate form. An explanation of analytic preparation is provided in Chapter 6.

Animal studies indicate that 2-butoxyacetic acid, the urinary metabolite found in humans, is also found in laboratory animals after inhalation exposure (Carpenter et al. 1956; Ghanayem et al. 1987a; Jijnsson and Steen 1978; Sabourin et al. 1992a). Twenty-seven female rats were exposed to 400 ppm (-561 mg/d), 17 female rats to 200 ppm (~281 mg/d), and 12 female rats to 100 ppm (~ 140 mg/d) 2-butoxyethanol (Carpenter et al. 1956). The estimated amount of metabolite (free 2-butoxyacetic acid) excreted was calculated from the urine samples collected during the 24-hour exposure period. The estimated amounts of free 2-butoxyacetic acid excreted were as follows (mean value per animal): 14 mg from rats exposed to 400 ppm, 7 mg from rats exposed to 200 ppm, and 1 mg from rats exposed to 100 ppm. Concurrent studies showed that most of the metabolite was excreted within 24 hours. In the same experiment, guinea pigs exposed to 100 ppm (~ 194 mg/d) or 200 ppm (~387 mg/d) 2-butoxyethanol excreted 4-5 mg in 24 hours, dogs exposed to 200 (~4,162 mg/d) and 400 ppm (~8,325 mg/d) excreted 42-100 mg, and rabbits exposed

to 200 ppm (~1,936 mg/d) and 400 ppm(~ 3,872 mg/d) excreted 12-23 mg and 89-302 mg, respectively, A correlation between vapor concentration and excretion of 2-butoxyacetic acid was observed for all animals tested except dogs. Additional studies of monkeys and dogs exposed to doses of 100-385 ppm 2-butoxyethanol for 28-90 days were also conducted, but the information was scanty and did not serve to clarify relative excretion rate by different species (Carpenter et al. 1956). The excretion of 2-butoxyethanol following 6-hour nose-only inhalation exposure of male Fischer 344 rats was determined at different inhaled concentrations (Sabourin et al. 1992a). The majority of the inhaled [^{14}C]2-butoxyethanol was eliminated in the urine, with free 2-butoxyacetic acid being the major urinary metabolite, accompanied by lesser amounts of ethylene glycol and 2-butoxyethanol glucuronide. A small proportion (5-8%) of the retained 2-butoxy-ethanol was exhaled as $^{14}CO_2$. Ratios of ethylene glycol to 2-butoxyacetic acid in plasma were higher than those in urine. Feces accounted for l-2% of the absorbed dose, whereas the carcass contained 13-20%. After exposure, 2-5% of the absorbed 2-butoxyethanol was exhaled unchanged. Urinary metabolites were monitored at various times up to 66 hours postexposure. More than 88% of the total radioactivity excreted was excreted during the first 41 hours. Free 2-butoxyacetic acid was the major metabolite in the urine at all exposure concentrations. Ethylene glycol and 2-butoxyethanol glucuronide were also found in lesser amounts. As the exposure concentration increased, the proportion of two unidentified minor metabolites increased. As the exposure concentration increased, the time required for urinary excretion of [^{14}C]2-butoxy-ethanol equivalents also increased. At 4.3 ppm, about 60% of the urinary radioactivity was excreted during the 6-hour exposure, whereas at 438 ppm, only 10% was excreted during the exposure period. At 49 ppm, metabolism to the glucuronide was favored during exposure, whereas metabolism to free 2-butoxyacetic acid and ethylene glycol was favored at later times postexposure. The pattern was even more apparent at the higher dose. CO_2 production most closely followed ethylene glycol production and may represent the breakdown of this metabolite.

Experiments designed to determine the effect of age on the toxicity and metabolism of 2-butoxyethanol indicate that in rats, young animals (4-5 weeks old) are more resistant to the toxic effects than are older rats (9-13 weeks old) receiving an equivalent dose (Ghanayem et al. 1987a). A significantly higher percentage of the administered dose was exhaled as CO_2 in young rats compared to adult rats (22% versus 10%). Young rats excreted a higher percentage of the dose in the urine compared to adult rats (60% versus 40%). Young rats excrete less free 2-butoxyacetic acid than adult rats, and more 2-butoxyethanol-glucuronide; and no sulfate conjugate of 2-butoxyethanol was detected. High-performance liquid chromatography analysis of the urinary metabolites of 2-butoxyethanol in adult and young rats showed that the ratio of free 2-butoxyacetic acid to the total amount of the glucuronide and sulfate conjugates of 2-butoxyethanol (previously thought to

reflect an activation/detoxification index of 2-butoxyethanol) was significantly higher in older rats. An unknown metabolite was found in the urine of both young and adult rats at 6-8% and 1%, respectively.

In a more recent study, eight male Sprague-Dawley rats were exposed to 20 or 100 ppm 2-butoxyethanol 24 hours per day for 1, 2, 3, 4, 6, 8, 10, or 12 days (Johanson 1994). Urine was collected at 24-hour intervals. At the end of the exposure period, animals were killed and blood, muscle, liver, and testes samples were collected. Blood and tissue samples were analyzed for free 2-butoxyethanol and free 2-butoxyacetic acid by electron capture gas chromatography. No control group was used. Total blood clearance values were 2.3 L/hour/kg for the 20-ppm group and 2.2 L/hour/kg for the 100-ppm group, and rates were not significantly different The urinary excretion rate for 2-butoxyacetic acid in the 20-ppm group averaged 0.20 mmol/day, whereas the 100-ppm group excreted 1 .03 mmol/day. The observed urinary excretion rate corresponded to 64% of the calculated respiratory uptake and renal clearance of 0.53 L/hour for both groups.

2.3.4.2 Oral Exposure

A case report of a woman admitted to the hospital after ingestion in a suicide attempt of 250-500 mL of a window-cleaner containing 12% (467-933 mg/kg) 2-butoxyethanol, indicated that metabolic acidosis, hypokalemia, a rise in serum creatinine level, and a markedly increased urinary excretion of oxalate crystals were observed, suggesting a metabolic pattern after absorption from the stomach (Rambourg-Schepens et al. 1988). Kinetic evaluation of urinary excretion of 2-butoxyethanol, free 2-butoxyacetic acid, and oxalate indicated that 2-butoxyethanol was at the highest level of excretion at 1 day after ingestion (2-3 g/g creatinine). 2-Butoxyacetic acid peaked between the 2nd and 3rd days of ingestion at 40 g/g creatinine and was not detectable on day 8. Oxalate excretion was biphasic, reaching similar peak levels of approximately 40 g/g creatinine on day 1 and day 7 after ingestion. The high output of the oxalate in the urine was explained by the metabolism of 2-butoxyethanol to ethylene glycol which is further metabolized to oxalic acid. The biphasic aspect of the oxalate excretion was not specifically addressed by the study authors, but since the two pathways of metabolism (to 2-butoxyacetic acid and ethylene glycol-oxalic acid) occur simultaneously, the biphasic response may be the result of competition for the pathways. This assumption was supported by the existence of metabolic acidosis contemporary with the urinary peak oxalate level. Similar metabolic effects have been reported by others (Bauer et al. 1992; Gijsenbergh et al. 1989).

A 23-year-old woman weighing 64 kg was admitted to the hospital approximately 1 hour after ingesting 500 mL window-cleaner containing 2-butoxyethanol(391-469 mg/kg) and ethanol (Gijsenbergh et al. 1989).

The half-lives of ethanol (from the cleaning fluid) and 2butoxyethanol in the blood were 20 and 210 minutes, respectively. The half-life of 2-butoxyethanol was longer than expected, perhaps because of saturation of metabolism and competitive inhibition by ethanol. 2-Butoxyacetic acid was highest in the dialysis fluid (738 mg/L) at the initiation of dialysis and decreased thereafter. A concomitant increase in 2-butoxyacetic acid in the urine was seen with the initiation of dialysis, and concentrations reached a peak of approximately 8 g/g creatinine 1.3 hours after the start of dialysis. The half-life of free 2-butoxyacetic acid in the urine after the start of dialysis was 5.3 hours.

Studies in animals indicate that 2-butoxyethanol is excreted through the urine, lungs, feces, and bile after exposure (Ghanayem et al. 1987b, 1987c). Male Fischer 344 rats were given single oral doses of [^{14}C]2-butoxyethanol of 125 or 500 mg/kg. Rats were placed in metabolism cages and monitored for 48 hours after treatment. Expired air, urine, and feces were collected and monitored for “C-associated radioactivity. Bile was collected from the common bile duct. The major route of elimination was the urine (40-70%), followed by exhalation (-18%). A small portion was excreted in the bile (8% at the 500-mg/kg dose) 8 hours after dosing, and a small amount was detected in the feces (2-3%).

Ghanayem et al. (1992) studied the differences m elimination of single or multiple oral doses of 2-butoxyethanol administered to rats, and found no quantitative or qualitative alterations of metabolism or disposition caused by repeated exposure to 2-butoxyethanol as compared to treatment with a single dose. Elimination of $^{14}CO_2$ and 2-butoxyethanol-derived radioactivity in the urine of rats receiving multiple doses of 2-butoxy-ethanol was essentially the same as for rats receiving a single dose. The ratios of free 2-butoxyacetic acid, the 2-butoxyethanol-glucuronide and 2-butoxyethanol-sulfate conjugates, and parent 2-butoxyethanol excreted in the urine of rats treated for 4 or 8 days were relatively similar to those from rats treated with a single dose.

Male Fischer 344/N rats were allowed access for 24 hours to uniformly labeled [^{14}C]2-butoxyethanol in drinking water at three doses (Medinsky et al. 1990). Elimination of radioactivity was monitored for 72 hours. 2-Butoxyethanol was administered at doses of 290, 860, or 2,590 ppm in drinking water, resulting in consumption of 237, 401, or 1,190 μmol/kg/day (28, 47, or 141 mg/kg/day). The majority f the ^{14}C was excreted in urine as free 2-butoxyacetic acid (50-60% of the dose), or exhaled as CO_2 (8-10% of the dose). Less than 5% of the dose was exhaled as unmetabolized 2-butoxyethanol. Ethylene glycol was excreted in urine, representing approximately 10% of the dose of 2-butoxyethanol. Elimination in the feces was a minor pathway for 2-butoxyethanol(1.6-2.8% of the dose) . Unmetabolized 2-butoxyethanol and its glucuronide conjugate were also identified in the urine of animals dosed with 2-butoxyethanol. The time course for

excretion of 2-butoxyethanol metabolites during and after exposure to the lowest dose indicated that the maximum excretion occurs during the first 12-24 hours after the start of exposure. This time course corresponds to the active period (nocturnal) for these animals, and the period during which they are likely to drink the most water. No evidence of the glycine conjugate of 2-butoxyacetic acid was found. The amount of ^{14}C -derived radioactivity found in the carcasses of animals ranged from 3 to 23.7 μmoles (2.8-9.8% of the dose) for the low- to high-dose groups.

2.3.4.3 Dermal Exposure

Worker exposures to 2-butoxyethanol were evaluated by environmental and biological monitoring for 29 employees in five shops where window-cleaning agents bad been used (Vincent et al. 1993). Each of the 29 employees was exposed to 2-butoxyethanol. Eleven men and two women were involved in cleaning cars in four garages. The remaining 16 were office cleaners. Only two of the workers who cleaned new cars wore protective gloves. The study authors suggested that the primary route of exposure was dermal, although some inhalation exposure necessarily occurred. Each of the window-cleaning agents sampled contained 2-butoxyethanol in concentrations ranging from 0.9% to 21.2% in volume. Window-cleaner use times ranged from 120 to 320 minutes per day, and quantities of cleaning agents ranged from 220 to 1,200 m /day for individuals involved in cleaning new cars. The corresponding figures for cleaners of used cars were 20-50 minutes and 50-100 mL. For office cleaners, the corresponding figures were 15 minutes per day and <50 mL, of cleaning agent used per day. For cleaners of new cars, the mean 2-butoxyethanol exposure was 2.33 ppm (range: <0.10-7.33 ppm), and the mean urinary free 2-butoxyacetic acid concentration in endshift samples was 111.3 mg/g creatinine (range: 12.7-371 mg/g creatinine). The mean preshift (average of Monday and Friday) urinary free 2-butoxyacetic acid concentration was 17.9 mg/g creatinine (range: 2-98.6 mg/g creatinine). For cleaners of used cars, the mean 2-butoxyethanol exposure was 0.36 ppm (range: <0.10-1.52 ppm), and the mean urinary free 2-butoxyacetic acid concentration in endshift samples was 6.3 mg/g creatinine (range: 2-24.4 mg/g creatinine). The mean preshift urinary concentration of free 2-butoxyacetic acid was 4.8 mg/kg creatinine (range: 2-33 mg/g creatimne). For the office cleaners, the mean 2-butoxyethanol concentration was 0.32 ppm (range: <0.30-0.73 ppm), and the mean u&mry free 2-butoxyacetic acid concentration in endshift samples was 2.1 mg/g creatinine (range: 2-3.3 mg/g creatinine). The mean preshift urinary concentration of free 2-butoxyacetic acid was 2.1 mg/g creatinine (range: 2-4.6 mg/g creatinine). Correlations of 2-butoxyethanol exposure in air and endshift concentrations of urinary free 2-butoxyacetic acid from cleaners of new and used cars (for which the data were most complete) indicated a log-linear relationship. From these results, the study authors estimated that exposure to

window cleaners containing 5.7-21.2% 2-butoxyethanol by volume for a period of 160 minutes would result in an endshift urinary concentration of free 2-butoxyacetic acid of 60 mg/g creatinine. A similar relationship was observed for urinary free 2-butoxyacetic acid concentrations and utilization time for office cleaners.

The percutaneous absorption of 2-butoxyethanol was investigated in 12 exposure experiments with five male volunteers. All were nonsmokers and stated low or no consumption of alcohol (Johanson et al. 1988). None had been exposed to industrial solvents. All had participated in a previous study of 2-butoxyethanol (Johanson et al. 1986a). The subjects kept two or four fingers immersed in neat 2-butoxyetbanol for 2 hours. Urine was collected for 24 hours and analyzed for the metabolite free 2-butoxyacetic acid. The presence of 2-butoxyethanol in blood and of free 2-butoxyacetic acid in urine, detected by gas chromatography, confirmed that 2-butoxyethanol enters the systemic circulation in human males during even minimal dermal exposure. The half-life of 2-butoxyethanol during the decay phase ranged from 0.6 to 4.8 hours (geometric mean: 1.3 hours). The excretion rate of 2-butoxyacetic acid increased during the first hours after exposure, reached a maximum at about 5 hours (3 hours postexposure), and then declined, with an average half-life of 3.1 hours. Seventeen percent of the absorbed dose was excreted as 2-butoxyacetic acid in 24 hours. The cumulative excretion of free 2-butoxyacetic acid ranged from 8.7 to 3 13 μM (1.15-41.37 mg).

In six men exposed one-arm only to 50 ppm [$^{13}C_2$] 2-butoxyethanol for 2 hours, about two-thirds of the 2-butoxyacetic acid excreted in the urine was in the form of the N-butoxyacetyl glutamine conjugate (Corley et al. 1997). No free 2-butoxyethanol, free or conjugated ethylene glycol ether, or glycolic acid (a metabolite of ethylene glycol) were detected in the urine. 2-Butoxyacetic acid was eliminated in the urine primarily during the first 12-hour collection period.

In a study in male and female Wistar rats, [^{14}C]2-butoxyethanol was applied to the shaven skin on a 12- cm^2 area under a protective glass capsule at a dose of 200 mg/kg (Bartnik et al. 1987). Urine was collected at intervals of 0-8, 8-24, and 24-48 hours following application and stored at -80° C. Radioactivity of urine, cage rinse water, skin, and glass capsule was determined. Percutaneous absorption was assessed by using the measurements of urinary excretion of ^{14}C from 0 to 48 hours following cutaneous application of 2-butoxyethanol. Radioactivity in 48 hours following cutaneous application of radiolabeled 2-butoxyethanol showed that 20-23% of the applied dose was found in the urine, including cage rinse water, with no notable differences by sex. Over 95% of the radioactivity excreted in the urine was eliminated in 24 hours.

Three different amounts of the [^{14}C]2-butoxyethanol (61, 181, or 299 mg/kg) were applied to same-sized areas on the clipped backs of Fischer 344/N rats, and nonoccluded percutaneous absorption was measured (Sabourin et al. 1992b, 1993). The rates of excretion of the ^{14}C -labeled parent compound and metabolites were measured, as well as the amount of ^{14}C remaining in the carcass. The majority of the absorbed and metabolized glycol ether was excreted in the urine (82-83%); 3-5% was found as CO_2, 3-6% was found in the feces, and 3-13% was found in the carcass. The percentage of glycol ether found tended to increase with increasing dose, although this was not consistently the case. Urinary metabolites were analyzed up to the time at which <10% of the excreted radioactivity. Urine was collected up to 23 hours postexposure. Free 2-butoxyacetic acid was the main metabolite (66-70%), followed by the 2-butoxyethanol-glucuronide (13-15%), and ethylene glycol(4-6%). CO_2 expired in the air was 3-4% in this experiment. The formation of small amounts of ethylene glycol indicated cleavage of the ether bond. Minor amounts of unidentified metabolites made up the remainder of the detected radioactivity.

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

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

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

The PBPK model for a chemical substance is developed in four interconnected steps: (1) model representation, (2) model parametrization, (3) model simulation, and (4) model validation (Krishnan and Andersen 1994). In the early 1990s, validated PBPK models were developed for a number of toxicologically important chemical substances, both volatile and nonvolatile (Krishnan and Andersen 1994; Leung 1993). PBPK models for a particular chemical 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. This simplification, however, is desirable if the uptake and disposition of the chemical substance(s) is adequately described because data are often unavailable for many biological processes and using a simplified scheme reduces the magnitude of cumulative uncertainty. The adequacy of the model is, therefore, of great importance and thus the importance of model validation.

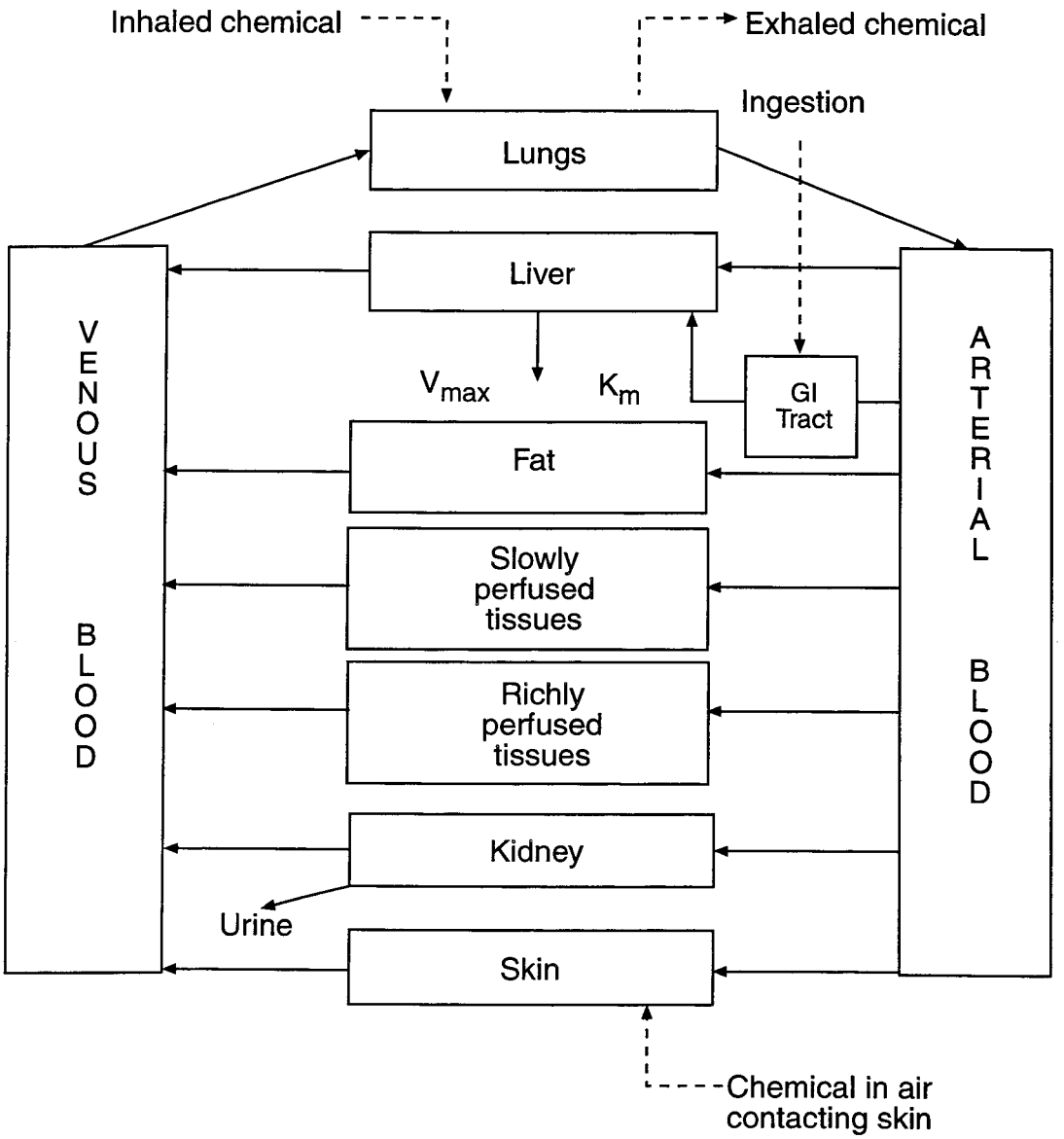
PBPK models improve the pharmacokinetic extrapolation aspects of the risk assessment process, which seeks to identify the maximal (i.e., safe) levels for human exposure to chemical substances (Andersen and Krishnan 1994). PBPK models provide a scientifically sound means to predict the target tissue dose of chemicals in humans who are exposed to environmental levels (for example, levels that might occur at hazardous waste sites) based upon the results of studies where doses were higher or were administered in different species. Figure 2-7 shows a conceptualized representation of a PBPK model.

PBPK models for 2-butoxyethanol are discussed below. No specific models were found for 2-butoxyethanol acetate. However, it is expected that 2-butoxyethanol acetate follows a similar model after absorption and conversion to 2-butoxyethanol. However, the absorption of 2-butoxyacetic acid may be different from 2-butoxyethanol because of its different chemical form.

2.3.5.1 Summary of PBPK Models

PBPK models of 2-butoxyethanol absorption, metabolism, disposition, and excretion have been published by Johanson and coworkers (Johanson 1986,199la; Johanson and Naslund 1988), Shyr et al. (1993), and Corley et al. (1994). They are presented in the order that they were published in the literature, since each

Figure 2-7. 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.

successive author used the data and results from the previous authors. Each successive model is more complete and more closely approaches modeling 2-butoxyethanol kinetics accurately. The Johanson model (Johanson 1986, 199 1 a; Johanson and Naslund 1988) addressed human inhalation exposure to 2-butoxyethanol during rest and exercise, whereas the Shyr model (Shyr et al. 1993) addressed high-low dose and route of administration extrapolation based on animal data. The Corley model (Corley 1996; Corley et al. 1994) combined aspects of each of the other two models and also addressed 2-butoxyacetic acid disposition and competing metabolic pathways. Because no chronic data are available for either humans or animals, these models have not been validated for chronic exposure to 2-butoxyethanol.

2.3.5.2 2-Butoxyethanol PBPK Model Comparison

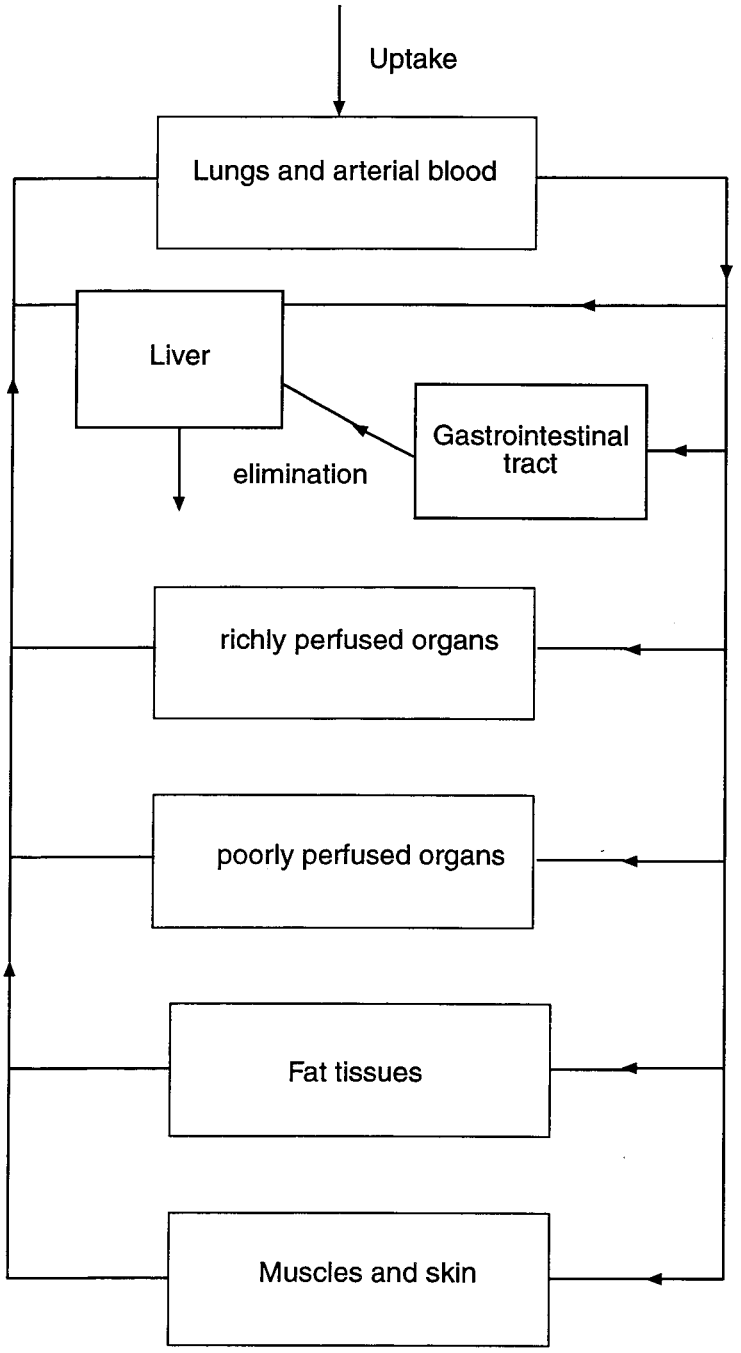
Three PBPK models that describe the disposition of 2-butoxyethanol have been identified from the open literature (late 1980s and forward). The first model (Figure 2-8) is based on human inhalation and perfused rat liver data, with later refinements (Johanson 1986, 1991a; Johanson and Naslund 1988). The second model (Figure 2-9) is based on rat data collected after inhalation, drinking water, and dermal exposure (Shyr et al. 1993). Shyr et al. (1993) also presented a model of the metabolic pathway of 2-butoxyethanol, as shown in Figure 2-10. The third model (Figure 2-l 1) was developed by Corley et al. (1994) and is an expansion of the Johanson model (Johanson 1986,199la; Johanson and Naslund 1988). It includes additional routes of exposure, physiological descriptions for rats, competing pathways for metabolism of 2-butoxyethanol, and measured partition coefficients for 2-butoxyethanol and 2-butoxyacetic acid. This model was further modified on the basis of data on human metabolites and dermal absorption of 2-butoxyethanol vapor (Corley et al. 1997).

2.3.5.3 Discussion of Models

The Johanson Model

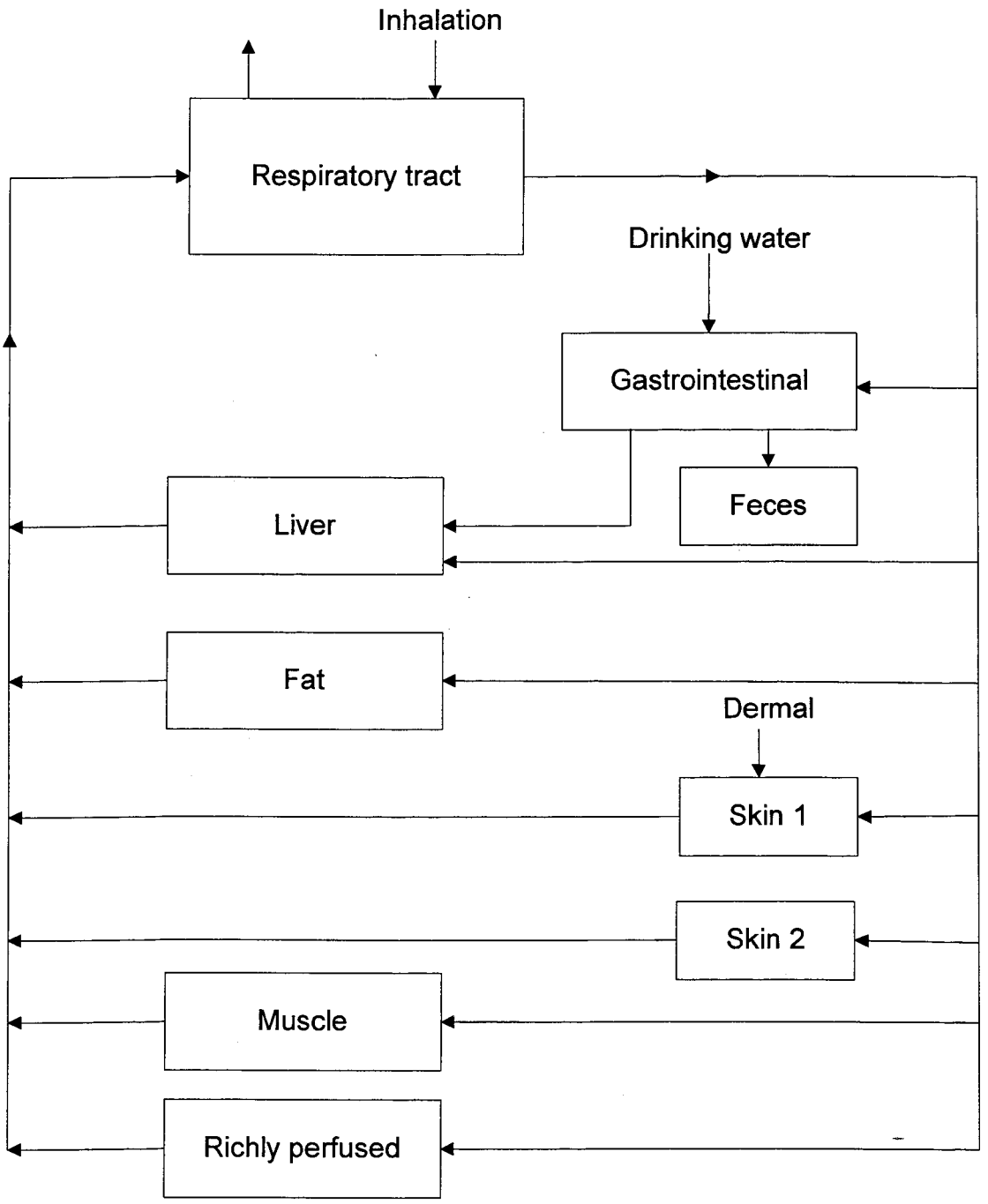
The Johanson model, with its refinements (Johanson 1986,199l a; Johanson and Naslund 198%), is one of the original PBPK models developed to describe and ultimately predict the effects of occupational inhalation exposure to 2-butoxyethanol and is depicted in Figure 2-8. Factors of exercise and co-exposure to ethanol are included.

Figure 2-8. Schematic Drawing of PBPK Model for Simulation of 2-Butoxyethanol Toxicokinetics*



*Derived from Johanson 1986

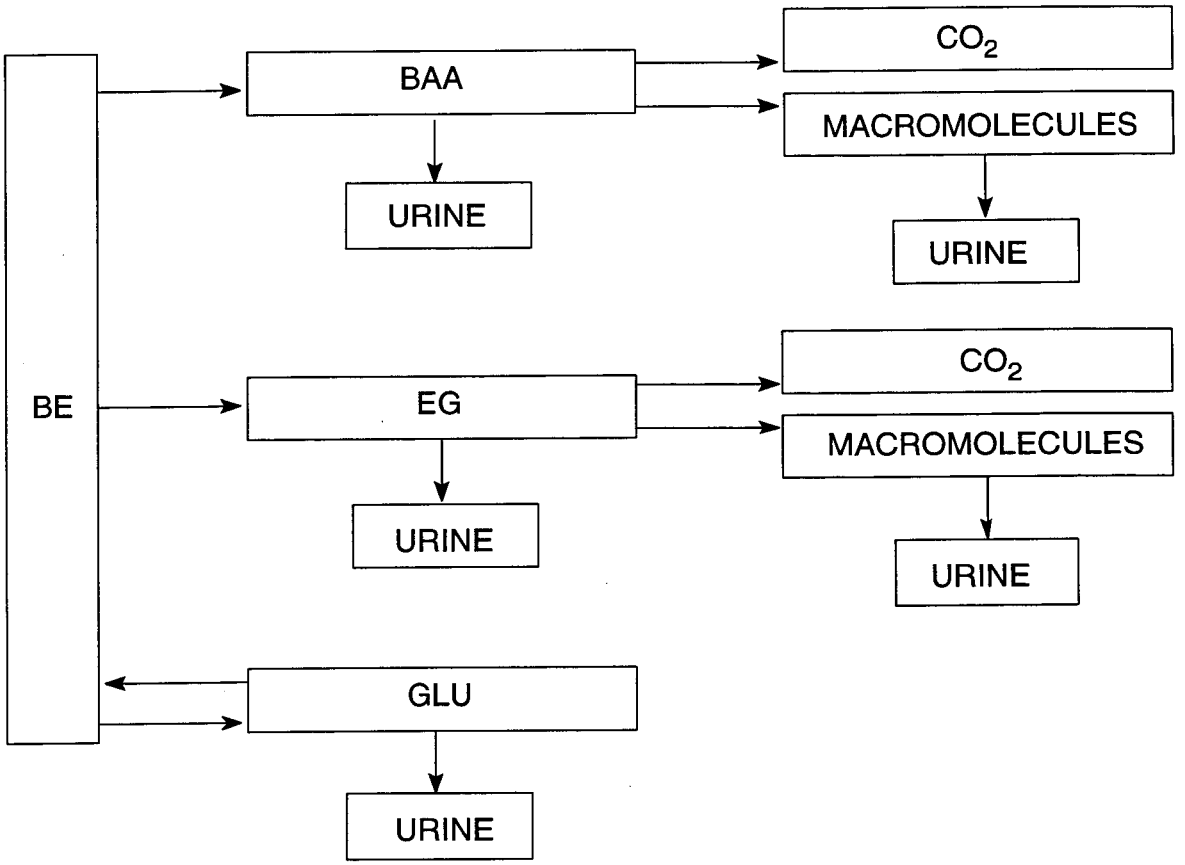
Figure 2-9. PBPK Model for 2-Butoxyethanol Disposition in Rats



Source: Shyr et al. 1993

Note: Skin 1 is the skin volume on which dermal dose was applied, and skin 2 is the rest of the skin volume.

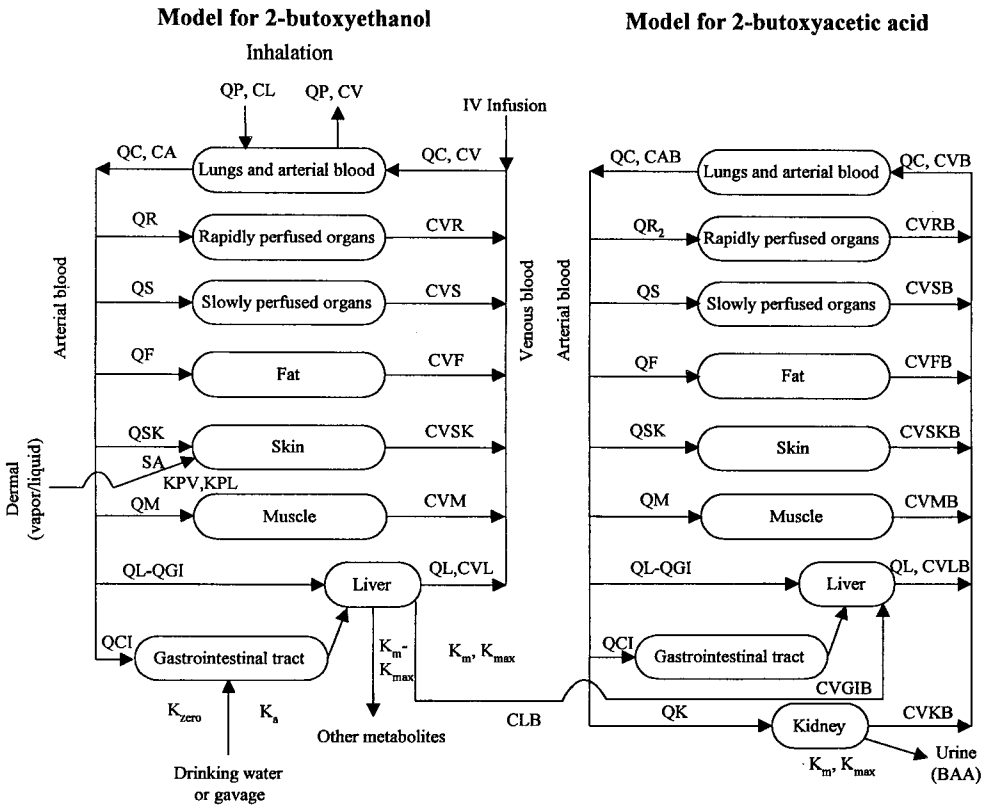
Figure 2-10. Model of Metabolic Pathway of 2-Butoxyethanol*



*Derived from Shyr et al. 1993

BAA = butoxyacetic acid; BE = 2-butoxyethanol; CO_2 = carbon dioxide; EG = ethylene glycol; GLU = glucuronide;

Figure 2-11. PBPK Model of 2-Butoxyethanol and 2-Butoxyacetic Acid



Source: Corley et al. 1994

Note: Physiologically based pharmacokinetic model used to describe PBPK model of 2-butoxyethanol and 2-butoxyacetic acid. In rats and humans following inhalation (whole-body or nose/mouth only), oral (gavage and drinking water), intravenous infusion, and dermal (vapor and liquid) routes of exposure. CL, concentration of BE in inhaled air (mg/liter); CX, concentration of BE in exhaled air (mg/liter); QP, alveolar ventilation (liter/hr); QC, cardiac output (liter/hr); Q_i, blood flow to "i" tissue (liter/hr); CA, arterial blood concentration of BE (mg/liter); CAB, arterial blood concentration of BAA (mg/liter); CV, pooled venous blood concentration of BE (mg/liter); CV_i, venous blood concentration of BE draining "i" tissue (mg/liter); CVB, pooled venous blood concentration of BAA (mg/liter); CV_iB, venous blood concentration of BAA draining "i" tissue (mg/liter); SA, surface area of skin exposed (cm^2); KPV, KPL, skin permeability coefficient for vapors or liquids, respectively (cm/hr); k_{zero}, zero-order rate of absorption of BE from water; k_a, first-order rate constant for absorption of BE from gavage dose (hr^{-1}); K_m, Michaelis constant for saturable process (mg/liter); V_{max}, maximum velocity for saturable process (mg/hr).

Risk assessment. This model successfully described the arterial blood concentrations of 2-butoxyethanol in a 70-kg man after inhalation exposure. Specifically, the model simulations indicated that for inhalation concentration of 20 ppm (the Swedish permissible exposure limit; U.S. limit = 25 ppm) for 2 hours accompanied by light physical exercise on an exercise bicycle, predicted arterial blood concentrations of 2-butoxyethanol agreed well with human experimental data (Johanson et al. 1986a). Simulations of inhalation exposure indicated that in humans, the concentration in blood increased in a linear manner throughout the 2-hour exposure, then declined rapidly to nearly non-detectable levels at 6 hours. By simulating different levels of physical activity at the same exposure level, but for 8 hours, mimicking a standard work day, Johanson's model was used to predict 2-butoxyethanol levels in blood after increased work load. As might be expected, the model predicts that with increased activity, and thus increased pulmonary blood flow and uptake of 2-butoxyethanol, arterial levels of 2-butoxyethanol increase proportionally during exposure, then decline rapidly after exposure ceases.

Simulations of co-exposure to ethanol (0.1% in the blood) during an S-hour exposure to 20 ppm 2-butoxyethanol with no exercise predict that the arterial levels of 2-butoxyethanol will be elevated because of a decrease in elimination rate (Johanson 1986). Because the increase is due to decreased elimination and not increased uptake, the rise and fall of blood concentration of 2-butoxyethanol are slower in the ethanol co-administration model than in the increased workload model.

Description of the model. The Johanson 2-butoxyethanol PBPK model was designed to describe organic solvent kinetics in general. Certain limiting assumptions were made in the model. For instance, the organs are lumped into compartments according to blood flow and fat content (Johanson 1986). Muscles and skin form a separate compartment because of their wide variation in blood flow at different workloads. In the model, solvent uptake is assumed to occur only in the Jung; elimination is assumed to occur in the liver. Instantaneous homogeneous distribution of the solvent in each compartment is assumed, and as a consequence, the solvent retained in the respiratory airways immediately reaches the arterial blood. A schematic representation of the Johanson model is shown in Figure 2-8, with only the inhalation route represented.

The tissue compartments included in the Johanson model are as follows: the lungs, presumed to be the only site of uptake, and arterial blood; the liver, presumed to be the only organ where transformation of 2-butoxyethanol takes place; the gastrointestinal tract in equilibrium with the liver; a group of richly perfused tissues; a group of poorly perfused tissues; a fat compartment; and muscles and skin. Support for the assumption

that the liver is the primary site of transformation of 2-butoxyethanol (as opposed to the kidney, for instance) comes from the investigator's observation that very little 2-butoxyethanol was recovered from the urine of men exposed to 2-butoxyethanol experimentally (Johanson et al. 1986a). All partition ratios between blood and other compartments were set to unity, based on preliminary studies on 2-butoxyethanol indicating a weak preference for water as compared to fat or protein. Diffusion equilibrium between end-capillary blood and tissue was assumed for all compartments.

Physiologic parameters used, biochemical constants, and partition coefficients in the model are shown in Table 2-9. Physiologic constants (organ volume, blood flows, etc.) and tissue and blood coefficients were taken from literature sources (Astrand 1983; Fiserova-Bergerova 1983a). Elimination constants for the liver were taken from experiments in the perfused rat liver (Johanson et al. 1986b). Venous equilibrium was assumed, and competitive inhibition between ethanol and 2-butoxyethanol was assumed.

Validation of the model. Since the model was developed using the data of Johanson et al. (1986a, 1986b), the model accurately reflects these data. Factors not taken into account in the model, but that may influence cardiac output and distribution and hepatic blood flow and thus, may have to be considered, include posture, thermal stress, food intake, pathological conditions, and the presence of other substances including drugs.

Target tissues. The model correctly described arterial blood concentrations of 2-butoxyethanol in human males during and after a 2-hour inhalation exposure to 20 ppm, accompanied by light physical exercise. Predictions of 2-butoxyethanol concentrations in muscle and skin may reflect blood sampled from peripheral veins, as would be accomplished in a biomonitoring program. The model shows that the rise and fall of 2-butoxyethanol in the arterial blood and lungs are faster than in the muscle and skin compartment. This suggests that venous blood sampling may be more useful than arterial sampling. A later refinement of the model included factors for "resting" and "working" skin and muscle, and data on tissueair partition coefficients were included, thus improving the predictive qualities of the model in an occupational setting (Johanson and Naslund 1988). Further refinement included modeling of the retention and excretion of solvent vapors in the various parts of the lung, including the trachea, main bronchi, lobar bronchi, segmental bronchi, subsegmental bronchi, small bronchi, bronchioles, terminal bronchioles, respiratory bronchioles, alveolar ducts, and alveoli, prior to equilibrating with other systemic compartments (Johanson 1991a). Data from 2-butoxyethanol experiments were included in the model (Johanson and Dynesius 1988; Johanson et al. 1986a).

Table 2-9. Parameters Used in the Johanson PBPK Model[a, b, c]

Parameters[d, e]	Volume (L)	Perfusion (L/Minute)			
		Rest	Light exercise	Middle exercise	Strenuous exercise
Pulmonary ventilation		6.0	22.0	38.0	54
Lungs and arterial blood	2.0	5.1	10.1	14.1	16.6
Gastrointestinal tract	2.4	1.2	1.2	0.9	0.5
Liver	1.5	1.6	1.6	1.3	0.9
Richly perfused organs	2.1	2.1	2.7	2.7	2.8
Poorly perfused organs	12.5	0.1	0.1	0.1	0.1
Fat tissues	14.5	0.25	0.65	1.0	0.8
Muscles and skin	33.0	1.0	5.0	9.0	12.0

[a]Derived from: Johanson 1986

[b]Volumes and blood flows were calculated from data compiled by Astrand (1983) and Fiserora-Bergerova (1983a).

[c] Metabolic constants were taken from *in vitro* experiments in perfused rat liver (Johanson et al. 1986b).

[d]Body weight = 70.0 kg.

[e] Metabolic and macromolecular binding constants: $V_{max}C$ (μmol/minute/g liver) = 0.730; K_m (μM) = 228

The data clearly indicate that, for accurate biomonitoring, sampling should take place during exposure because of the rapid disappearance of 2-butoxyethanol from tissue compartments. Even in the compartment that shows the longest residence time (poorly perfused organs), practically no 2-butoxyethanol is present the morning following exposure. The model suggests that bioaccumulation does not generally occur, although accumulation by a particular organ (or in a disease state) could not be ruled out. The study authors state that human data, combined with the physicochemical properties of the chemical, do not suggest that specific organ binding would substantially alter the total body burden. The model does not address the kinetics of the major metabolite, 2-butoxyacetic acid.

Species extrapolation. The Johanson model uses human data to predict occupational exposure parameters. The author notes that the validity of the assumptions used to extrapolate to humans from in vitrs hepatic elimination data on rats is not known.

High-low dose extrapolation. The model predicts nonlinearity due to saturated elimination at concentrations well above 100 ppm, even in combination with physical exercise and ethanol. The comparison between the simulated arterial blood levels and experimental arterial blood levels obtained after exposure to 20 ppm 2-butoxyethanol for 2 hours indicates linear kinetics at this concentration, which is lower than the PEL in the United States (PEL = 50 ppm, OSHA 1974). Thus, linear kinetics would be expected in ordinary occupational inhalation exposure, although low-dose extrapolation was not specifically addressed in this model.

Interroute extrapolation. The Johanson model was not used to predict the kinetic behavior of 2-butoxyethanol after dermal or oral exposure. The study author comments that saturation kinetics may occur after accidental or intentional ingestion of large amounts or after accidental dermal exposure to large volumes of 2-butoxyethanol, but not after exposure in a controlled occupational setting.

The Shyr Model

The Shyr model (Shyr et al. 1993) is based on experimental data from inhalation, drinking water, and dermal exposure experiments in rats (Figure 2-9). The major metabolite, 2-butoxyacetic acid, was included in the model in addition to the 2-butoxyethanol-glucuronide and ethylene glycol. A model formulation of the possible metabolic pathways based on experimental data was included (Figure 2-10).

Risk assessment. This model successfully described the differences in disposition of 2-butoxyethanol, based on urinary profiles of metabolites, in various exposure routes by taking into account the differences in absorption rate and by incorporating a minor pathway for metabolism (glucuronidation) by skin (Shyr et al. 1993). The model was reasonably successful in predicting the experimental rat data produced by Johanson et al. (1986a). The value of the model lies in the ability to extrapolate to the human situation from the routes of exposure likely to be used in experimental assays.

Description of the model. The Shyr 2-butoxyethanol PBPK model (Shyr et al. 1993) was based on an earlier PBPK model developed by Ramsey and Andersen (1984) to describe the disposition of styrene exposure in mice and rats, and on the Johanson (1986) model. A schematic representation of the Shyr model is shown in Figure 2-9, with inhalation, drinking water, and dermal routes represented. Parameters used in the model are summarized in Table 2-10.

The tissue compartments included in the Shyr model are as follows: respiratory tract; liver; gastrointestinal tract; fat; and a group of richly perfused tissues including kidney, bone marrow, and heart. Muscle and skin were separated into individual compartments to allow for the simulation of dermal exposure. The distribution of 2-butoxyethanol among compartments was assumed to be limited only by blood flow rate because the lipid solubility of 2-butoxyethanol allowed it to penetrate cell membranes rapidly. Liver was a major site of metabolism in the Shyr model with a minor amount of 2-butoxyethanol-glucuronide formed in the skin at the site of application for dermal exposure.

Drinking water exposure was simulated by estimating nocturnal drinking patterns of rats as a continuous activity. For dermal exposure, the rate of 2-butoxyethanol absorption through the skin was also estimated and adjusted according to the *in vitro* absorption data of Bartnik et al. (1987). A factor was included for dermal metabolism to the 2-butoxyethanol-glucuronide. Inhalation exposure was estimated using the model of Ramsey and Andersen (1984) with a factor for nasal absorption (Dahl et al. 199 1). Nasal absorption includes dissolution of the compound in the nasal mucous membrane upon inhalation, and vaporization and elimination from the membranes into expired air. Based on the work of Dahl et al. (199 1) in dogs, a high nasal uptake was assumed for compounds with high blood/air partitions. Increasing nasal absorption for soluble compounds is also suggested by Dahl et al. (1991). More recent work on kinetic models for respiratory uptake of inhaled volatiles has been done by Medinsky et al. (1993). Physiologic, biochemical constants and partition coefficients used in the model are shown in Table 2-10 Physiologic constants (organ volume, blood flows, etc.)

Table 2-10. Parameters Used in the Shyr PBPK Model[a]

Parameters for the rat	Value
Body weight (kg)	0.288
Percentage of body weight	
Liver	3.0
Fat	11.0
Skin included in application site	0.5
Skin not included in application site	9.5
Muscle	56.0
Gastrointestinal tract	3.0
Richly perfused tissues	4.6
Other	12.4
Flows (L/hour)	
Alveolar ventilation	15.1
Cardiac output	18.9
Percentage of cardiac output	
Liver	5.0
Muscle	9.6
Fat	6.0
Rapidly perfused tissues	57.0
Skin included in application site	0.12
Skin not included in application site	2.3
Gastrointestinal tract	20.0
Partition coefficients	
Blood/air	8000
Liver/blood	0.9
Fat/blood	0.7
Rapidly perfused tissues	0.9
Gastrointestinal/blood	0.9
Skin/blood	0.9
Muscle/blood	0.9
Biochemical parameters and rate constants	
$V_{max,ba}$ c (µmol/hour)	344
$K_{m,ba}$ (µmol/L)	317
$V_{max,eg}$ c (µmol/hour)	7.9
$K_{m,eg}$ (µmol/L)	23.2
$V_{max,glu}$ c (µmol/minute)	48.2
$K_{m,glu}$ (µmol/L)	472

[a]Derived from Shyr et al. 1993

[b]Includes tissues that are not perfused with blood (such as bone [not including marrow]), body fluids (such as gastrointestinal tract contents, urine, bile, and blood in major vessels not included in any tissue group), and hair.

ba = butoxyacetic acid; eg = ethylene glycol; glu = glucuronide; K_m = concentration at ½ V_{max}; V_{max} = maximum rate of metabolism

and tissue and blood coefficients were taken from Ramsey and Andersen (1984) or were taken from other literature sources.

Validation of the model. Since the model (Shyr et al. 1993) was developed concurrently with toxicokinetic experiments being conducted in the same laboratory, the model was used to predict the outcome of experiments that were yet to be run (Medinsky et al. 1990; Sabourin et al. 1992a, 1992b, 1993). Variables that were not predictive were modified according to the experimental data. Thus, the model was developed and refined in conjunction with the data it used for validation. The model was successful in predicting urinary metabolites from the three routes of exposure.

Target tissues. The model attempted to predict the urinary profiles of 2-butoxyethanol metabolites in rats after inhalation, oral (drinking water), and dermal exposure (Shyr et al. 1993). For 2-butoxyacetic acid, the major urinary metabolite, the values predicted by the model were in reasonable agreement with the experimental data. The exception was inhalation exposure at the high dose. The study authors indicated that the lack of agreement at the high dose for inhalation exposure may have been due to several factors, including a wide variability in the experimental data and an overestimation of the formation rate of 2-butoxyacetic acid in the liver at the high dose. For the other metabolites in the urine, ethylene glycol and 2-butoxyethanol glucuronide, the model predicted well for inhalation, oral, and dermal exposure. However, the predicted values for ethylene glycol after dermal exposure at the ow dose were relatively high. Shyr et al. (1993) presented a model formulation of the metabolic pathway of 2-butoxyethanol, leading to urinary excretion (Figure 2-10). Based on the data of Medinsky et al. (1990) and Sabourin et al. (1992a, 1992b, 1993), 2-butoxyethanol was primarily metabolized to 2-butoxyacetic acid, ethylene glycol, and the glucuronide of 2-butoxyethanol. Both ethylene glycol and 2-butoxyacetic acid could be metabolized further to CO_2. The unexcreted glucuronide was also available for hydrolysis back to 2-butoxyethanol. The rates at which 2-butoxyacetic acid and ethylene glycol were metabolized to CO_2 were proposed to be less than the rates at which these metabolites were excreted in the urine, since the amount of urinary metabolites was greater than the amount of CO_2 exhaled. Based on work done with ethylene glycol (Marshall 1982), small amounts of ethylene glycol and 2-butoxyacetic acid may be bound to macromolecules, such as protein, resulting in a longer retention time in the body. In the Shyr model (Shyr et al. 1993), the major compartment for 2-butoxyethanol metabolism was the liver, based on the study of Johanson et al. (1986b). Minor amounts of glucuronide are also assumed to be formed in the skin after dermal application, but there is little data to support this assumption.

Species extrapolation. Shyr et al. (1993) used the human inhalation data of Johanson et al. (1986b, 1988). Pulmonary ventilation and body weight data from Johanson et al. (1986a) were used, while other kinetic parameters were the same as the rat model. The metabolic parameters were adjusted for humans (K_m and V_{max}) according to Ramsey and Andersen (1984). The model adequately predicted urinary excretion of 2-butoxyacetic acid, which was similar to that of rats. Wben the model was applied to human dermal exposure data (Johanson et al. 1988), it overestimated 2-butoxyacetic acid excretion in the urine by 33%, as compared to experimental data.

High-low dose extrapolation. Three doses were used for each route of administration. The model accurately predicted the outcome with exception of predicting urinary 2-butoxyacetic acid excretion after the high-dose inhalation exposure (382 μmol absorbed dose). The study authors (Shyr et al. 1993) indicated that the discrepancy may have resulted from variability in the experimental levels of urinary 2-butoxyacetic acid and because the estimates did not include a factor for the unidentified metabohtes experimentally observed in rat urine after inhalation exposure. No low-dose extrapolation, mimicking doses to which most consumers would be exposed, was conducted.

Interroute extrapolation. The model (Shyr et al. 1993) accurately predicted the urinary metabolite profile of 2-butoxyethanol after inhalation, oral (drinking water), and dermal absorption in rats (Medinsky et al. 1990; Sabourin et al. 1992a, 1992b, 1993). The same set of parameters was used to predict data from all three routes of exposure. Metabolic pathways were proposed based on experimental data, and the relative significance of each pathway was quantified by fitting the model to the experimental data. The model predicted the change in profiles of urinary metabolites for different exposure routes.

The Corley Model

The Corley model (Corley et al. 1994) is an expansion of the model of human inhalation exposure by Johanson (1986). In the Corley model, disposition of 2-butoxyacetic acid was included, and the model was expanded to include data on rats, the most commonly used species in experimental toxicity tests of 2-butoxy-ethanol. This model was further modified on the basis of data on human metabolites and dermal absorption of 2-butoxyethanol vapor (Corley et al. 1997). other routes of exposure, including oral, dermal, and intravenous infusion, were included (Figure 2-l 1). This is a new model, however, and has not yet been subjected to extensive testing.

Risk assessment. In proposing the parameters necessary for a useful model to be used in the risk assessment of 2-butoxyethanol exposure, Corley et al. (1994) indicated that since human exposure is most likely by inhalation and dermal routes, these routes should be included in a model. In addition, formation of the toxic metabolite, 2-butoxyacetic acid, should be a focus of the model. Since species differences in hemolytic sensitivity to 2-butoxyethanol are known (Ghanayem and Sullivan 1993), these differences should be factored in also. The model was reasonably successful in simulating the concentration of 2-butoxyethanol in human blood after inhalation exposure (assuming no dermal absorption), in agreement with the data of Johanson (1986). In addition, total respiratory uptake of 2-butoxyethanol, the area under the 2-butoxyethanol blood concentration-time curve, and the amount of 2-butoxyacetic acid excreted in the urine were predicted with reasonable agreement. Corley et al. (1994) suggested that risk assessments can be strengthened through the use of an internal dose factor, such as the maximum concentration of 2-butoxyacetic acid in the blood, or the area under the curve for 2-butoxyacetic acid in blood in place of the administered dose, especially in interspecies comparisons. Maximum concentration is sensitive to dose rate (e.g., gavage bolus versus drinking water), whereas the area under the curve is sensitive to saturation of metabolism and elimination.

Description of the model. To develop the model Corley et al. (1994) used several types of data, including physiological constants for rats and humans, partition coefficients for 2-butoxyethanol and 2-butoxyacetic acid in various tissues, and biochemical constants describing the metabolism of 2-butoxy-ethanol and excretion of 2-butoxyacetic acid. The values used are presented in Tables 2-l 1 and 2-12.
Partition coefficients were available from the literature for 2-butoxyethanol in bloodair and salineair (Johanson and Dynesius 1988). Partition coefficients were determined experimentally for 2-butoxyethanol and 2-butoxyacetic acid in human blood and in the blood, liver, kidney, muscle, fat, skin, lung, stomach, small intestine, and cecum of male Fischer 344 rats(Corley et al. 1994). The metabolism of 2-butoxyethanol to 2-butoxyacetic acid was assumed to take place only in the liver through the alcohol/aldehyde dehydrogenase pathway, as described previously by Johanson (1986). A saturable metabolic pathway was added to account for all other possible pathways causing the disappearance of 2-butoxyethanol from the liver. Parameters for this pathway were estimated from the model of Medinsky et al. (1990) and scaled for humans. The assumption was made in the Corley model (1994) that 2-butoxyacetic acid was bound to proteins in the blood and was eliminated by a saturable process in the kidneys (i.e., renal acid transport) as suggested by the work of Ghanayem et al. (1990a). Incorporation of protein binding was necessary to reduce the amount of “free” 2-butoxyacetic acid available for elimination in order to closely simulate data in rats and humans. Elimination constants were estimated from the data of Ghanayem et al. (1990a). The maximum concentration (C_{max}) and the area under the curve for 2-butoxyacetic acid concentration in the blood were used as estimates of the

Table 2-11. Parameters Used in the Physiologically Based Pharmacokinetic Model for 2-Butoxyethanol and 2-Butoxyacetic Acid[a, b]

Parameters	Rat	Human	Estimation method[c]
Surface area (cm^2)	267	19,000	Fixed[d]
Body weight (kg)	0.23	70	Fixed
Percentage of body weight			
Liver	2.53	3.14	Fixed[e,f]
Kidney (2-butoxyacetic acid model)	(0.71)	(0.44)	Fixed[e,g]
Lung	1.17	1.15	Fixed[f]
Fat	7.0	23.1	Fixed[f]
Muscle	37.0	42.0	Fixed[h]
Gastrointestinal tract	3.4	3.4	Fixed[h]
Skin	10.0	5.1	Fixed[i]
Rapidly perfused	5.1	3.71	Fixed[g]
(2-butoxyacetic acid model)	(4.39)	(3.27)	Fixed[g]
Slowly perfused	24.8	9.4	Fixed[j]
Flows (L/hour)			
Alveolar ventilation	5.06	347.9	Fixed[f]
Cardiac output	5.06	347.9	Fixed[f]
Percentage of cardiac output			
Liver (including cardiac output from the gastrointestinal tract)	25.0	25.0	Fixed[g]
Gastrointestinal tract	21.0	21.0	Fixed[h]
Kidney (2-butoxyacetic acid model)	(25.0)	(25.0)	Fixed[g]
Fat	5.0	5.0	Fixed[g]
Muscle	12.0	15.0	Fixed[h]
Skin	5.0	5.0	Fixed[d]
Rapidly perfused	51.0	50 .0	Fixed[k]
(2-butoxyacetic acid model)	(26.0)	(25.0)	Fixed[k]
Slowly perfused	2	2	Fixed[l]
Partition coefficients for 2-butoxyethanol			
Blood/air	7965	7965	Fixed[m]
Liver/blood	1.46	1.46	Measured[n]
Kidney/blood	1.83	1.83	Measured[n]
Lung/blood	11.3	11.3	Measured[n]
Fat/blood	2.03	2.03	Measured[n]

Table 2-11 (*continued*)

Parameters	Rats	Humans	Estimation method[c]
Muscle/blood	0.64	0.64	Measured[n]
Gastrointestinal tract/blood	4.33	4.33	Measured[n]
Skin/blood	2.90	2.90	Measured[n]
Skin/air	7965	7965	Fixed[m]
Rapidly perfused blood	1.46	1.46	Measured[n]
Slowly perfused blood	0.64	0.64	Measured[n]
Partition coefficients for 2-butoxyacetic acid			
Liver/blood	1.30	1.30	Measured[n]
Kidney/blood	1.07	1.07	Measured[n]
Lung/blood	1.58	1.58	Measured[n]
Fat/blood	0.77	0.77	Measured[n]
Muscle/blood	1.31	1.31	Measured[n]
Gastrointestinal tract/blood	0.78	0.78	Measured[n]
Skin/blood	1.21	1.21	Measured[n]
Rapidly perfused blood	1.30	1.30	Measured[n]
Slowly perfused blood	1.31	1.31	Measured[n]
Metabolic constants			
2-Butoxyethanol-to-2-butoxyacetic acid			
K_{m1} (mg/L)	26.9	26.9	Fixed[h]
2-Butoxyethanol-to-others			
V_{max2} C (mg/hour/kg)	5	5	Fitted[o]
K_{m2} (mg/L)	0.5	0.5	Fitted[o]
Protein binding			
P (binding sites; mg/L)	164	164	Fixed[p]
K_d (dissoc. const.; mg/L)	46	46	Fixed[p]

Table 2-11 (*continued*)

Parameters	Rats	Humans	Estimation method[c]
Elimination rate constant for 2-butoxyacetic acid [in Urine]			
K_mE (mg/L)	0.5	0.5	Fitted[q]
Gavage absorption rate constants			
K_aS (hour^{-1}) water	1.0	1.0	Fitted[r]

[a]Derived from Corley et al. (1994)
[b]Values in parentheses are specific for the 2-butoxyacetic acid model unless otherwise designated.
[c]The model parameters were estimated independently and held fixed (fixed), measured in independent experiments (measured), or estimated by fitting the model to the data (fitted).
[d]McDougal et al. (1990)
[e]Dow historical control data (rat)
[f]Andersen et al. (1987)
[g]Corley et al. (1990)
[h]Johanson (1986)
[i]Fiserova-Bergerova (1983c)
[j]Total tissues listed represents 91% of body weight (the remaining 9% represents structural tissues with negligible blood perfusion). For mass balance, the volume of slowly perfused tissues (% body weight) was assumed to be 91 - (sum of other tissues).
[k]For mass balance, blood flow to rapidly perfused tissues (% cardiac output) was assumed to be 100 - (sum of other tissues).
[l]Blood flow to slowly perfused tissues from Corley et al. (1990) after accounting for muscle and skin.
[m]Johanson and Dynesius (1988). Skin:air assumed to be equal to blood:air.
[n]Determined *in vitro* using ultrafiltration (Jepson et al. 1992).
[o]Estimated by fitting model to data of Medinsky et al. (1990).
[p]Russell et al. (1987)
[q]Estimated by fitting model to data of Ghanayem et al. (1990a).
[r]Estimated by fitting model to data from Figure 3 and Tables 5 and 6.

Table 2-12. Human Physiological Parameters Adjusted for Light-to-Moderate Exercise[a,b]

Parameters	Value
Flows (L/hour)	
Alveolar ventilation	1,322
Cardiac output	603
Percentage of cardiac output	
Liver (including gastrointestinal)	16
Kidney (2-butoxyacetic acid model)	(11)
Fat	6
Muscle[c]	42
Gastrointestinal tract	12
Skin[c]	3
Rapidly perfused tissue (2-butoxyacetic acid model)	32 (21)
Slowly perfused tissue	1

[a]Derived from Corley et al. 1994

[b]50 watts on bicycle ergonometer

[c]Muscle and skin compartment of Johanson (1986) described separately and kidney compartment added for the BAA model. Blood flows to these compartments were adjusted according to Astrand (1983).

"internal dose" or "delivered dose" for extrapolations between species, routes, and high and low doses. Concentration-response relationships for 2-butoxyacetic acid-induced hemolysis in rat and human blood have been shown (Bartnik et al. 1987; Ghanayem 1989; Ghanayem et al. 1987, 1992; Udden 1994).

Validation of the model. The reliability of the model was tested using data sets reported by several laboratories, in addition to data produced by Corley et al. (1994). Data from previous reports were used to derive estimates of biochemical parameters describing dermal uptake of 2-butoxyethanol by humans (Ghanayem et al. 1990a; Johanson and Boman 199 1; Medinsky et al. 1990). Validation of these parameters was performed by comparing model predictions with other studies having relevant data. A simulation was conducted of 2-butoxyethanol and 2-butoxyacetic acid concentrations in the blood of young and old rats dosed intravenously with 2-butoxyetbanol. Reasonable agreement was obtained for simulations for the younger rats. The model tended to overpredict the concentration of 2-butoxyacetic acid at low doses soon after administration but was adequate thereafter. In older rats, adjustments to the parameters, including increasing the volume of the fat compartment, and reducing the rate of elimination of the kidneys to simulate the renal disease common in aged rats, improved the simulations.

The parameters associated with the disposition of 2-butoxyacetic acid were validated against experimental data produced by Corley et al. (1994). Reasonable agreement was reached for 2-butoxyacetic acid in the blood. Model simulations of the amounts of 2-butoxyacetic acid excreted by young male rats (16-18 weeks) and older rats (12 or 13 months) were compared with tbe data of Ghanayem et al. (1987a, 1987b). The best predictions were for young rats at doses where hemolysis did not occur. The model did not adjust for the toxic effects of the compound on the kidney or liver that may have been secondary to the hemolytic effect. The Corley model was responsive to the normal changes in age and weight in animals.

The Corley model achieved reasonable agreement between simulations and experimental data on 2-butoxyacetic acid excreted in the urine of rats after exposure to 2-butoxyethanol in the drinking water (Medinsky et al. 1990). Excellent agreement was also reached with the data of Sabourin et al. (1992a) after inhalation exposure of rats, for respiratory uptake of 2-butoxyethanol, total amounts of 2-butoiyethanol metabolized, and total amounts of 2-butoxyacetic acid excreted. The exception was for the total amount of 2-butoxyacetic acid excreted at the high dose, which caused hemolysis in the experimental animals (Sabourin et al. 1992a). The model over-predicted the amount excreted, another indication that the PBPK model did not account for hemolytic toxicity.

Simulations of the human inhalation data of Johanson et al. (1986a) for the concentration of 2-butoxyethanol in the blood were comparable to the original data, assuming no dermal uptake. Instead of using a standardized weight, the actual body weights of the volunteers in the study were used, and the parameters were scaled by body weight for each. The predicted pulmonary ventilation rate for the volunteers who engaged in light exercise agreed with those observed (Johanson et al. 1986a). Reasonable agreement was also obtained for respiratory uptake, area under the curve for 2-butoxyethanol blood concentration-time, and the amount of 2-butoxyacetic acid excreted in the urine. Simulations of the data of Johanson and Boman (1991), including dermal uptake through exposure to the vapor, suggested that dermal uptake was not as important as was indicated by Johanson and Boman (1991). When the assumption was made that the finger prick blood samples taken in the Johanson and Boman (1991) study were venous blood samples draining the skin, rather than mixed arterial blood, the model adequately predicted the data (Corley 1996). Data from an additional dermal exposure study, in which volunteers were exposed one arm only to 50 ppm 2-butoxyethanol for 2 hours and in which blood samples were taken from the unexposed arm (Corley et al. 1997) confirmed this theory. The Corley model was further refined by changing it to better reflect human metabolism as measured by urinary metabolite levels in Corley et al. 1997, and using a new permeability coefficient for dermal absorption of vapors The model was changed to make a majority of total urinary butoxyacetic acid in the form of the glutamine conjugate and to eliminate urinary excretion of free and conjugated butoxyethanol and free or conjugated ethylene glycol or its metabolite, glycolic acid.

Target tissues. Reasonable agreement was achieved between the model and experimental data for 2-butoxyacetic acid levels in the blood of rats after gavage exposure and in the urine of rats after drinking water exposure, respiratory uptake of 2-butoxyethanol, total amount metabolized, and total 2-butoxyacetic acid excreted after inhalation exposure of rats, as long as the doses were below those that caused hemolysis. In humans, reasonable agreement was achieved between the model and experimental data for respiratory uptake, the area under the curve for 2-butoxyethanol blood concentration-time, and the amount of 2-butoxyacetic acid excreted in the urine after inhalation exposure (Corley et al. 1994).

Species extrapolation. To compare species, Corley et al. (1994) ran simulations of the maximum concentration and area under the curve for 2-butoxyethanol and 2-butoxyacetic acid in rat and human blood after a 6-hour dermal exposure to aqueous 2-butoxyethanol. A 10% body surface exposure was assumed, with no evaporation of the chemical. Maximum absorption was predicted at 40% solutions, with less absorbed at higher and lower doses. Although the reasons for these results are not clear, they suggest that under normal circumstances, humans are unlikely to absorb enough 2-butoxyethanol through dermal exposure alone to cause

hemolysis. Following inhalation exposure, the model predicts that humans would not be expected to produce enough 2-butoxyacetic acid to result in hemolysis even if they were exposed to concentrations up to 1,160 ppm 2-butoxyethanol, the saturated vapor concentration (Corley 1996). This conclusion was reached even with the inclusion of dermal absorption of vapor, the estimate of which was subsequently reduced on the basis of Corley et al. (1997) human dermal absorption data.

High-low dose extrapolation. The model seems to predict well for animals at low and middle doses that do not cause hemolysis for oral (drinking water) and inhalation exposure (Medinsky et al. 1990; Sabourin et al. 1992a). The model was not designed to adjust for toxicity in the kidney and liver that may have been secondary to the hemolytic effects of 2-butoxyethanol. Low dose extrapolation, modeling doses similar to those at which most consumer exposure occurs, was not conducted.

Interroute extrapolation. The model seemed to predict well for different routes of exposure in animals, as long as the doses were below those causing hemolysis. The model predictions did not agree with human dermal absorption estimations of Johanson and Boman (1991) when it was assumed that finger prick blood samples represented arterial blood. The model predictions did agree with human data when it was assumed that finger prick blood samples represented venous blood draining the skin, and they predicted the one arm-only exposure scenario (Corley 1996). Human inhalation exposure was predicted well (Johanson et al. 1986a).

2.4 MECHANISMS OF ACTION

2-Butoxyethanol toxicity has been studied in some detail, and current understanding of 2butoxyethanol includes its metabolic fate, mechanism of toxicity, pharmacokinetic models for disposition, and impact of exposure on human health. These aspects of 2-butoxyethanol toxicity will be addressed in the following sections. The toxicity of 2-butoxyethanol acetate has been studied far less than 2-butoxyethanol toxicity, presumably because 2-butoxyethanol acetate is assumed to be metabolized to 2-butoxyethanol in the body.

2.4.1 Pharmacokinetic Mechanisms

Pharmacokinetic models of 2-butoxyethanol disposition have been developed (Bartnik et al. 1987; Corley et al. 1997; Corley et al. 1996; Corley et al. 1994; Ghanayem et al. 1990a; Medinsky et al. 1990; Sabourin et al. 1992a, 1992b, 1993; Shyr et al. 1993). Both *in vivo* and *in vitro* data indicate that metabolic activation of

2-butoxyethanol by the alcohol/aldehyde dehydrogenases is a prerequisite for the development of the characteristic hematotoxicity (Dow 1982; Ghanayem et al. 1987b, 1987c; ICI 1989). Metabolism is presumed to take place in the liver; excretion takes place primarily via the urine.

2-Butoxyethanol metabolism to its reactive intermediate, 2-butoxyacetic acid, presumably through the intermediate 2-butoxyacetaldehyde, takes place through oxidation by the alcohoValdehyde dehydrogenases. Nontoxic pathways include conjugation of 2-butoxyethanol with glucuronic acid, which is excreted in the urine; thus, the conjugate formation leads to elimination and the prevention of formation of toxic metabolites. Some evidence exists that ethylene glycol is also a metabolite of 2-butoxyethanol under certain circumstances through dealkylation; ethylene glycol formation may depend on the route of administration (Corley et al. 1994; Medinsky et al. 1990; Rambourg-Schepens et al. 1988; Sabourin et al. 1992a, 1992b, 1993; Shyr et al. 1993). At higher doses, saturation of the 2-butoxyacetic acid and ethylene glycol pathways favors the formation of glucuronide conjugates of 2-butoxyethanol (Sabourin et al. 1992a). Ethylene glycol itself has been shown to cause hepatic, renal, and developmental toxicity, as summarized in the ATSDR *Toxicological Profile for Ethylene Glycol and Propylene Glycol* (ATSDR 1993). However, ethylene glycol is a minor metabolite of 2-butoxyethanol, and the toxicity of 2-butoxyethanol in humans and animals can be understood on the basis of on its metabolism to 2-butoxyacetic acid.

2.4.2 Mechanisms of Toxicity

The most characteristic toxic effect of 2-butoxyethanol in animal models is on the hematologic system and includes elevated osmotic fragility of erythrocytes, hemolysis, decreased hematocrit, hemoglobinuria, hemoglobinemia, and hemolytic anemia (Bartnik et al. 1987; Carpenter et al. 1956; Corley et al. 1994; Dodd et al. 1983; Dow 1959; Duprat and Gradiski 1979; Eastman Kodak 1983; Ghanayem and Sullivan 1993; Ghanayem et al. 1987a, 1987b, 1990b, 1992; Grant et al. 1985; Hardin et al. 1984; Krasavage 1986; Nagano et al. 1979,1984; NTP 1993,1989; Sabourin et al. 1992a, 1992b, 1993; Union Carbide 1980a). Evidence of the early stages of hemolysis, characterized by erythrocytic swelling and detected as increased mean cell volume, increased hematocrit, and decreased mean cell hemoglobin concentration, has also been observed after exposure to 2-butoxyethanol (Bartnik et al. 1987; Dodd et al. 1983; Ghanayem and Sullivan 1993; Tyl et al. 1984). These effects have also been observed after 2-butoxyethanol acetate exposure (Truhaut et al. 1979). Compensatory erythropoiesis, occurring in response to hemolysis, results in an increase in reticulocytes, increased mean cell volume, increased or normal mean cell hemoglobin, and decreased mean cell hemoglobin concentration (McGrath 1993); it may also contribute to changes in hematological parameters, as was evident

in some animal studies of 2-butoxyethanol (Dodd et al. 1983; Ghanayem et al. 1987a, 1992; Grant et al. 1985; NTP 1989; Tyl et al. 1984).

Hemolysis can occur within the circulatory system, or outside the circulatory system in the spleen, bone marrow, or liver (McGrath 1993). Hemoglobinemia (the presence of free hemoglobin in the blood) and hemoglobinuria (the presence of free hemoglobin in the urine) are indicators of destruction of red blood cells within the circulatory system (Jones and Hunt 1983; McGrath 1993). Hemoglobin is normally contained in erythrocytes. When hemoglobin is released from damaged erythrocytes, it is detectable in the blood, is normally decomposed in the spleen and other organs of the reticula-endothelial system, and is eliminated as bilirubin. When the amount of free hemoglobin in the blood is such that the spleen and other organs of the reticula-endothelial system cannot completely remove it from the circulation, it appears in the urine (hemoglobinuria). When hemoglobinuria is detectable, it is reasonable to assume that hemolytic anemia is occurring. Hemoglobinuria is normally identified by the brownish or reddish-brown color of the urine (Jones and Hunt 1983). Increased hemosiderin, resulting from free hemoglobin in the circulatory system, may also be present in the kidney tubules (McGrath 1993). Hemolysis occurring outside the circulatory system is characterized by increased spleen size and weight, and hemosiderin deposits in the spleen and liver.

Most of the toxic effects of 2-butoxyethanol or 2-butoxyethanol acetate observed in animal studies may be secondary to the hemolytic effects of these chemicals. Free hemoglobin resulting from intravascular hemolysis can cause renal damage (pigment nephropathy) which may result in renal failure (Rosenstock and Cullen 1994). Hemolysis can cause accumulation of hemoglobin in renal tubular cells, and may also result in anemia, insufficient blood supply, and renal ischemic injury culminating in acute tubular necrosis. Adverse renal effects secondary to hemolysis, including tubular necrosis and other histopathological changes, have been noted in animals after inhalation (Carpenter et al. 1956), oral (Eastman Kodak 1983; Ghanayem et al. 1987a, 1987b; Krasavage 1986), or dermal exposure to 2-butoxyethanol (Duprat and Gradiski 1979; Union Carbide 1980a) and after inhalation, oral, or dermal exposure to 2-butoxyethanol acetate (Truhaut et al. 1979). Extravascular hemolysis results in accumulation of heme pigments in the liver and spleen; thus, necrosis, pigment deposition, and increased organ weight in these organs are most likely secondary to the hemolytic effects of 2-butoxyethanol (Dodd et al. 1983; Duprat and Gradiski 1979; Eastman Kodak 1983; Ghanayem et al. 1987a, 1987b; Grant et al. 1985; Krasavage 1986; NTP 1989, 1993). As a result of hemolysis, anemia and insufficient blood supply may occur, resulting in effects such as cold extremities (NTP 1989; Tyl et al. 1984) and necrosis of the tail tip (Dow 1981; Hardin et al. 1984; Nelson et al. 1984; Tyl et al. 1984).

Hematuria, the presence of whole red blood cells in the urine, can be identified by the reddish color of the urine (Jones and Hunt 1983). Hematuria results from hemorrhaging into the urinary system. Although hemoglobinuria and hematuria may both be present, whether or not the mechanisms of toxicity producing the two effects are related is unclear. It is possible that hematuria occurs after 2-butoxyethanol exposure via leakage of erythrocytes through renal structures that have been damaged by the presence of free hemoglobin resulting from hemolysis .

As indicated above in Section 2.3.3, metabolic activation of 2-butoxyethanol to 2-butoxyacetic acid via the alcohol/aldehyde dehydrogenase pathway is necessary for hematotoxicity. Ghanayem and coworkers (Ghanayem 1989; Ghanayem and Sullivan 1993) undertook studies to determine the mechanisms of hemolytic effects in rat and human erythrocytes *in vitro*, and to determine the relative sensitivity of human erythrocytes to 2-butoxyacetic acid. Incubation of 5 or 10 mM 2-butoxyethanol with rat blood resulted in no significant hemolysis; no evidence of 2-butoxyethanol metabolites was found (Ghanayem 1989). Incubation of rat erythrocytes with 20 mM 2-butoxyethanol resulted in hemolysis that was determined to be nonspecific in nature, because as with the 5- and 10 M doses, no metabolites of 2-butoxyethanol were detected at this higher dose. Addition of either alcohol or aldehyde dehydrogenase to whole blood with 2-butoxyethanol did not cause either hemolysis or production of 2-butoxyetbanol metabolites. Thus, the work of Ghanayem (1989) suggests that whole blood is not capable of metabolizing 2-butoxyethanol *in vitro* to 2-butoxyacetic acid, the hemolytic metabolite. Incubation of rat erythrocytes with butoxyacetaldehyde or 2-butoxyacetic acid caused time- and concentration-dependent swelling of red blood cells followed by hemolysis, which has also been observed *in vivo* (Ghanayem et al. 1990b). Butoxyacetaldehyde was less effective in causing hemolysis than was 2-butoxyacetic acid, suggesting that, although whole rat blood may contain enough aldehyde dehydrogenase to cause some conversion of the aldehyde to 2-butoxyacetic acid, the 2-butoxyacetic acid is the hemolytic agent. Addition of aldehyde dehydrogenase and its cofactors, followed by butoxyacetaldehyde, resulted in a significant increase in hemolysis, which could be decreased with the addition of cyanamide, an aldehyde dehydrogenase inhibitor, thus supporting the evidence that production of 2-butoxyacetic acid is the important step in 2-butoxyethanol-specific hemolysis (Ghanayem et al. 1989). Further experiments indicated that in rat erythrocytes, 2-butoxyacetic acid and butoxyacetaldehyde caused a decrease in blood adenosine triphosphate (ATP) concentration. The observation that 2-butoxyethanol-induced hemolysis is preceded by cellular swelling and that this was associated with the depletion of ATP, suggests that the red blood cell membrane is the target of toxicity, although the primary effect on the erythrocyte is not clear. Studies on the direct effect of 2-butoxyacetic acid on the red blood cell also suggest that the red blood cell membrane is the site of toxic insult (Ghanayem and Sullivan 1993). Human erythrocytes were relatively resistant to the effects

of 2-butoxyacetic acid *in vitro,* suggesting differences between the human cell membrane and that of the rat (Ghanayem 1989; Ghanayem and Sullivan 1993; ICI 1989). Studies of erythrocytes from guinea pigs, dogs, pigs, and cats indicated these species were resistant to the effects of 2-butoxyacetic acid, whereas rats, mice, and rabbit erythrocytes were susceptible (Ghanayem and Sullivan 1993; ICI 1989). Blood from mice, rats, hamsters, rabbits, and baboons exposed *in vitro* is susceptible to the hemolytic effects of 2-butoxyacetic acid, which produces time- and concentration-dependent increases in mean corpuscular volume and hematocrit at concentrations at or above 1 mM (Ghanayem and Sullivan 1993). Ghanayem et al. (1992) conducted a series of experiments on the *in vitro* effect of 2-butoxyacetic acid on blood from 2-butoxyethanol-treated rats. Red blood cells from rats exposed to 125 mg/kg 2-butoxyethanol for 3 days and allowed to recover for 7 days prior to bleeding were less sensitive to the hemolytic effects of 2-butoxyacetic acid than red blood cells from untreated rats. This work confirms the observation that young erythrocytes, formed in response to recovery from the hemolytic effects of the first exposure to 2-butoxyacetic acid, are more resistant to the hemolytic effects of 2-butoxyacetic acid than mature erythrocytes. This work suggests that chronic exposure to 2-butoxyethanol might result in tolerance since the aging erythrocytes would be lysed, and since the remaining circulating erythrocytes would be younger and more resistant to hemolysis. The resilience of newly formed red blood cells to 2-butoxyethanol exposure has been confirmed by Sivarao and Mehendale (1995). These investigators also showed that bleeding rats, which increases new red blood cell formation, provided protection against the hemolytic effects of 2-butoxyethanol.

In an effort to further elucidate the mechanism of toxicity of 2-butoxyethanol, Foster et al. (1987) dosed rats directly with 2-butoxyacetic acid once by gavage with 0, 174, 434, or 868 mg/kg/day 2-butoxyacetic acid in water. Hematuria resulted from direct exposure to a single dose of 2-butoxyacetic acid at 868 mg/kg/day, but not at 434 mg/kg/day or less. The results of this study confirmed that the metabolite of 2-butoxyethanol (i.e., 2-butoxyacetic acid) could produce hematological effects similar to those observed after 2-butoxyethanol exposure. It is unknown why such a high dose of 2-butoxyacetic acid was needed to produce hemolysis since this metabohte is thought to cause the hemolytic effects resulting from exposure to 2-butoxyethanol. It is possible that the differences in the rate of uptake and excretion of the metabolite may account for the need of a higher dose level.

Evidence of anemia has also been observed in cases of intentional human poisoning with 2-butoxyethanol (Bauer et al. 1992; Gijsenbergh et al. 1989; Rambourg-Schepens et al. 1988). It has been suggested that support with fluids (Bauer et al. 1992; Gijsenbergh et al. 1989; Rambourg-Schepens et al. 1988) and hemodialysis (Bauer et al. 1992; Gijsenbergh et al. 1989) may contribute to hematological effects in humans

following ingestion of large doses of 2-butoxyethanol (Udden 1996). Before hemolysis is considered as the mechanism for anemia in humans, Udden (1996) suggests that additional information regarding 2-butoxy-ethanol ingestion is needed, including review of morphology of red blood cells, reticulocyte count, mean cell volume, and tests for intravascular hemolysis (lactate dehydrogenase, haptoglobin, hemoglobinuria). Hemoglobinuria should be distinguished from myoglobinuria, a condition in which myoglobin is excreted in the urine. Myoglobin resembles blood hemoglobin but contains only one heme as part of the molecule.

Metabolic acidosis has been observed following exposure to 2butoxyethanol. Metabolic acidosis may be due to production of lactate and 2-butoxyacetic acid (Gijsenbergh et al. 1989). Metabolic acidosis, if left uncorrected, leads to hyperventilation and renal failure, which have been identified as possible toxic sequellae to 2-butoxyethanol exposure (Gijsenbergh et al. 1989; Litovitz et al. 1991; Rambourg-Schepens et al. 1988).

No specific mechanism of toxicity has been described for other toxic effects of 2-butoxyethanol, including respiratory, cardiovascular, gastrointestinal, neurological, reproductive, and developmental effects. Respiratory effects observed in humans include irritation of the nose and throat and a slight increase in nasal mucus discharge after inhalation or dermal exposure (Carpenter et al. 1956), and pulmonary edema or respiratory insufficiency after oral ingestion (Bauer et al. 1992; Gijsenbergh et al. 1989). In animals, inhalation exposure has been shown to cause rapid and shallow breathing, audible respiration, nasal discharge, perinasal encrustation, decrease in respiratory rate, and congested or hemorrhagic lungs (Carpenter et al. 1956; Dodd et al. 1983; Dow 1986; Duprat and Gradiski 1979; Kane et al. 1980; Tyl et al. 1984). After oral exposure, dyspnea has been observed (NTP 1989; Wier et al. 1987). Some of these effects, including changes in respiratory rate and hemorrhaging of the respiratory tissues, may be related to metabolic acidosis and hemolysis, respectively. After oral exposure, humans exhibit cardiovascular effects including tachycardia and low blood pressure (Bauer et al. 1992), but similar effects have not been observed in animals. Gastrointestinal effects, including emesis, have been observed in humans, monkeys, and dogs after inhalation exposure (Carpenter et al. 1956), but the cause is unknown. other gastrointestinal effects, including hyperkeratosis and acanthosis, and diarrhea observed in animals after oral dosing may be related to contact irritation of the gastrointestinal tissues (Eastman Kodak 1983; Krasavage 1986; NTP 1993). Hemorrhagic orred stomach or intestines observed after oral or dermal dosing in animals may be related to the hemolytic activity of 2-butoxyethanol (Union Carbide 1980a, 1980b). No mechanism has been described for neurological effects, including headache, CNS depression, and disturbed taste sensation in humans after inhalation exposure (Carpenter et al. 1956), and loss of coordination, excess salivation, and weakness in animals after inhalation, oral, or dermal exposure (Carpenter et al. 1956; Dodd et al. 1983; Dow 1959, 1972, 1981, 1986; Duprat and

Gradiski 1979; Eastman Kodak 1983; Hardin et al. 1984; Krasavage 1986; NTP 1989; Olin 1976; Union Carbide 1980a, 1980b; Wier et al. 1987). Coma has been observed after human poisoning (Bauer et al. 1992; Gijsenbergh et al. 1989; Rambourg-Schepens et al. 1988). With the exception of decreased pup weight observed in mice at 700 mg/kg/day (Heindel et al. 1990), developmental effects (including a decrease in viable implants and an increase in malformations in animals) reported occurred at 2-butoxyethanol doses that resulted in moderate to severe maternal toxicity (Hardin et al. 1987; NTP 1989; Tyl et al. 1984; Wier et al. 1987). Reproductive effects have been observed at doses that resulted in other adverse effects in adult animals (Hardin et al. 1987; Heindel et al. 1990; NTP 1989, 1993; Schuler et al. 1984; Tyl et al. 1984; Wier et al. 1987). Mechanisms for the reproductive and developmental effects of 2-butoxyethanol have not been identified.

Several *in vitro* studies have investigated the action of 2-butoxyethanol on cells in culture, the results of which may suggest additional or related areas of research that may lead to an explanation of 2-butoxyethanol's effect on erythrocytes. 2-Butoxyethanol has been shown to effectively block metabolic cooperation between Chinese hamster V79 cells in culture at nontoxic doses (Loch-Caruso et al. 1984). At higher doses, the cells died. Metabolic cooperation is a manifestation of the transfer of metabolites between cells coupled at a specific junction (gap junction), a process that has been implicated in cell division, morphological development, and differentiation. Disruption of this process could contribute to cellular death or dysfunction, teratogenesis, or tumor promotion. In another study, 2-butoxyethanol and 2-butoxyacetic acid were shown to be cytotoxic to Chinese hamster ovary cells in culture, as indicated by a reduction in cloning efficiency (Jack et al. 1985). In support of this observation, the study authors cited the work of others, Loch-Caruso et al. (1984) and Johnson et al. (1984), who observed that 2-butoxyethanol exposure resulted in developmental toxicity of the hydra. Jackh et al. (1985) also noted that the hemotoxicity of 2-butoxyethanol observed in small rodents *in vivo* and *in vitro* (Ghanayem et al. 1987a, 1987b, 1989, 1992) also suggests cytotoxicity. In another study, 2-butoxy-ethanol was shown to decrease maximum calcium uptake by rabbit sarcoplasmic reticulum vesicles and to markedly increase the subsequent calcium efflux rate (Mayahara et al. 1982). In addition, 2-butoxyethanol caused tetany in an isolated phrenic nerve-diaphragm preparation of the rat. The significance of these results with regard to *in vivo* toxicity is currently unknown.

2.4.3 Animal-to-Human Extrapolations

Recent physiologically based pharmacokinetic models have tried to address 2-butoxyethanol metabolism and disposition in an effort to derive animal-to-human extrapolations (Medinsky et al. 1993; Shyr et al. 1993). Other models have also been developed based on human data (Johanson 1986,199la; Johanson and Naslund 1988). Each model was multicompartmental and attempted to relate the generation of metabolites to end points of 2-butoxyethanol toxicity. Primary focus has been on absorption from the respiratory system and the skin (Johanson 199 1 a; Shyr et al. 1993). Absorption from the gastrointestinal tract has also been addressed (Shyr et al. 1993). Additional data have recently been published to more adequately model the metabolism and disposition of 2-butoxyethanol in rats (Johanson 1994), as well as additional animal and human data and revised modeling parameters that more adequately address issues of routes of exposure and competing metabolic pathways (Corley et al. 1997; Corley et al. 1994). With this additional work, the database and resulting model are more complete, and come close to adequately modelling data for both animals and humans Additional study, including regarding dermal uptake, protein binding parameters, renal elimination, mechanism of hemolysis of rat erythrocytes and its relevance in humans, and further evaluation of the metabolism and kinetics of 2-butoxyethanol and its metabohtes in animals with erythrocytic sensitivity similar to humans (less sensitive than rats), has been recommended to improve the ability of the model to predict outcome in different species (Corley et al. 1994).

In vitro studies have shown that, compared to rat blood cells, human red blood cells are insensitive to the hemolytic effects of 2-butoxyacetic acid. Concentrations of 2-butoxyacetic acid that caused hemolysis of rat red blood cells did not cause hemolysis of human red blood cells (Bartnik et al. 1987). For example, following a 60-minute exposure of rat red blood cells to 7.5 mM 2-butoxyacetic acid, 100% hemolysis was observed. Following a 60-minute exposure of human red blood cells to 15 mM 2-butoxyacetic acid, no hemolysis was observed (Bartnik et al. 1987). An *in vitro* study measuring more sensitive end points of red blood cell effects, i.e., mean cell volume and hematocrit, has also shown rat red blood cells to be more sensitive to 2-butoxyacetic acid compared to human red blood cells (Ghanayem and Sullivan 1993). For example, following a 1 -hour exposure to 2 mM 2-butoxyacetic acid, mean corpuscular volume was about 150% of controls in rat red blood cells and 105% of controls in human red blood cells; hematocrit was about 155% of controls in rat red blood cells and 105% of controls in human red blood cells. Red blood cells of rabbits, hamsters, mice, and baboons were also sensitive to 2-butoxyacetic acid, while red blood cells of cats, pigs, dogs, and guinea pigs were not sensitive to 2-butoxyacetic acid (Ghanayem and Sullivan 1993). The molecular mechanism for the resistance of red blood cells from some species but not others is not known. Until the mechanism is known, it

has been suggested that 2-butoxyethanol should be treated as a chemical with the potential for causing human toxicity (Ghanayem 1996).

2.5 RELEVANCE TO PUBLIC HEALTH

Overview

2-Butoxyethanol and, to a lesser extent, 2-butoxyethanol acetate are found in a wide variety of industrial and consumer products. For 2-butoxyethanol, the exposure scenario of most concern to the general public is inhalation or dermal absorption while using household cleaners, metal cleaners, spray lacquers, quick-dry lacquers, enamels, varnishes, varnish removers, and latex paints. 2-Butoxyethanol acetate is used in nitrocellulose lacquer, epoxy and acrylic enamels, latex coatings, and some ink and spot remover formulations. Individuals employed in industries that make or use 2-butoxyethanol or 2-butoxyethanol acetate (e.g., hospitals and medical facilities, silk screen shops, furniture finishers, print shops, paint manufacture) are probably exposed to the highest concentrations of atmospheric 2-butoxyethanol or 2-butoxyethanol acetate. In the general population, people residing around certain chemical manufacturing sites or living near waste sites containing 2-butoxyethanol or 2-butoxyethanol acetate may theoretically be exposed to concentrations higher than background air concentrations; however, neither of these chemicals has been measured in air near NPL sites.

Several *in vivo* and *in vitro* studies have conclusively demonstrated that 2-butoxyethanol and 2-butoxyethanol acetate can be absorbed through human skin. Dermal absorption can occur from exposure to the liquid form and small amounts of the vapor can be absorbed dermally. Since over half of the products containing 2-butoxyethanol marketed in the United States are intended for household use, the potential for dermal exposure is great. All-purpose cleaners comprise the largest proportion of consumer products containing 2-butoxyethanol. Consumers generally do not wear respiratory protection or protective gloves when performing general cleaning chores such as washing windows, mopping floors, cleaning cars, etc. Thus, the potential for exposure through both the derrnal and inhalation routes is substantial.

Occupational exposure to 2-butoxyethanol usually involves co-exposure to other solvents and chemicals. In several NIOSH Health Hazard Evaluations, effects reported by workers included eye, nose, and throat irritation; coughing; runny nose; headache; dizziness; lightheadedness; and nausea. Since personal breathing zone and workplace air samples analyzed for solvents and other chemicals (such as toluene, xylene, methyl

ethyl ketone, methyl isobutyl ketone, styrene) along with 2-butoxyethanol indicated that exposure levels for each chemical were below the NIOSH, ACGIH, and OSHA criteria, NIOSH concluded that the effects were probably due to the additive combination of the solvents.

Acute oral exposures of humans to large amounts of 2-butoxyethanol have been shown to cause coma and respiratory depression, in addition to hematotoxic effects Although this route of exposure is the least likely for the general population, the existence of many household products containing 2-butoxyethanol or 2-butoxy-ethanol acetate makes accidental poisoning a potential problem, particularly for children.

Acute- and intermediate-duration inhalation, oral, and dermal exposures to 2-butoxyethanol have been shown to cause death in animals. There is a report of human death following the ingestion of an unknown amount of cleaner containing 6.5% 2-butoxyethanol. Only scanty data are available for 2-butoxyethanol acetate. These data suggest that in animals 2-butoxyethanol acetate may be less lethal than 2-butoxyethanol. No studies were located regarding death in humans or animals after exposure to 2-butoxyacetic acid by any route.

The most noted systemic effect in animals resulting from acute and intermediate exposure to 2-butoxyethanol or 2-butoxyethanol acetate is hematotoxicity. Hemolysis occurs in several stages, including swelling, popping, reduction in volume, and hemoglobin leakage. Some or ah of these stages have been observed in at least three cases of human poisoning and in animal studies. Hemodialysis may have contributed to the hemolytic effects in humans. The hematotoxicity is characterized in humans by decreased hemoglobin content, progressive erythropenia, and hemoglobinuria, and in animals by swelling of the erythrocytes, followed by hemolytic anemia, and decreases in circulating red blood cells, hemoglobin concentration, and hematocrit. A causal relationship has been established for animals, with the proposed target being the red blood cell membrane. Hemoglobinuria observed in animals is a consequence of the hemolytic effects of the 2-butoxyethanol metabolite, 2-butoxyacetic acid. An *in vitro* study has shown that the red blood cells of rats, mice, hamsters, rabbits, and baboons are more susceptible to the hemolytic effects of 2-butoxyacetic acid than the red blood cells of guinea pigs, dogs, pigs, and cats. In contrast to animals, human red blood cells appear to be more resistant to the hemolytic effects of 2-butoxyacetic acid, although presumably the same over&mechanism is operational in humans.

Other notable systemic effects observed in humans exposed to high doses inelude respiratory depression, metabolic acidosis, alteration of cardiac rhythm, and hematuria. In animals, hepatic and renal toxicity have been observed, including hepatic necrosis and hemoglobin casts in the renal tubules and hematuria. Increased

spleen weight and splenic congestion have been observed after 2-butoxyethanol exposure in animals. Many of the effects observed may be due to the hemolytic effects of 2-butoxyethanol.

Data on effects of 2-butoxyethanol and 2-butoxyethanol acetate on male reproductive tissues in animals generally indicate that male reproductive organs are not affected, but one study indicated decreased sperm concentration in rats after intermediate-duration oral exposure to 2-butoxyethanol. However, no significant effects on spermatid heads (10^7/g testis), spermatid counts (mean/10^{-4} mL), or percent mobile spermatozoa were observed. Reproductive effects in female animals consist of vaginal bleeding and altered estrous cycles. Data on reproductive and developmental competence in animals indicate that at sufficiently high doses, especially those that result in adult animal toxicity, 2-butoxyethanol can adversely affect these processes. Reduced fetal viability and retarded skeletal ossification were the most frequently observed effects.

Limited *in vitro* data indicate that 2-butoxyethanol is not genotoxic. No studies were located regarding cancer in humans or animals after exposure to 2-butoxyethanol.

In studies of 2-butoxyethanol exposure in humans, the information provided on exposure levels and their correlation with observed effects were sufficient for derivation of a chronic inhalation MRL. Data from animal studies were used to derive other MRL's.

Minimal Risk Levels for 2-Butoxyethanol

Inhalation MRLS

- An MRL of 6 ppm has been derived for acute-duration inhalation exposure (14 days or less) to 2-butoxyethanol.

The MRL was calculated from a NOAEL of 50 ppm for hematotoxicity in pregnant rats in a study by Tyl et al. (1984). See Appendix A for details. In the study by Tyl et al. (1984), timed-pregnant Fischer 344 rats (n=36) were exposed to 2-butoxyethanol vapors by inhalation on gestational days 6-15 at concentrations of 0, 25, 50, 100, or 200 ppm for 6 hours per day. The animals were observed for clinical signs throughout the study, and food and water consumptions (withheld during exposures) was measured. Maternal body weights were taken on gestational days 0, 6, 9, 12, 15, and 21. The animals were sacrificed on gestational day 21 after blood samples were collected. Hematology and organ weights (uterus, liver, thymus, spleen, and kidney) were evaluated.

Hematologic determinations on dams (at 100 and 200 ppm but not 50 ppm) showed statistically significant reductions in erythrocyte (red blood cell) count and mean corpuscular hemoglobin concentration, and an increase in hemoglobin per cell (mean corpuscular hemoglobin) and size of the red blood cell (mean corpuscular volume). There were significant increases in hemoglobin and hematocrit at 200 ppm. Clinical signs that occurred with an incidence different from the controls were observed in 75% of dams in the 200-ppm group during the exposure period. They included the clear evidence of hematuria or hemoglobinuria, periocular wetness, perinasal encrustation, pale and cold extremities, and necrosis of the tail tip. Periocular wetness, clear evidence of hematuria, and perinasal discharge were also noted in 25-50% of the dams in the 100-ppm group. In the 25- and 50-ppm groups, 25% of the dams showed signs of periocular wetness. Maternal body weight gain was decreased 29% in the 100-ppm group at gestational days 6-15, compared to the control group. Exposure resulted in negative weight gain (weight loss during specific periods of time, at 200 ppm), decreased maternal body weight (gestational days 6-21, 1 l-12% at 200 ppm), decreased food consumption (gestational days 6-15, 13% at 100 ppm), and evidence of anemia (200 ppm) that was not observed at lower concentrations. Decreased water consumption (14%) was noted at 200 ppm. There was a reduction in maternal gravid uterine weight at 200 ppm. Relative kidney weight was elevated compared to controls at 200 ppm in the presence of decreased body weight. No treatment-related effects were observed on the weights of the liver.

Although periocular wetness was observed at high incidences in the rats at ≥25 ppm, indicating that 25 ppm is a minimal LOAEL for eye irritation (from direct contact of the eyes with the vapor during inhalation exposure), hemolysis is the most consistently observed critical effect of concern for exposure to 2-butoxyethanol.

Other studies have shown hematotoxicity after acute inhalation exposure at similar concentrations. Fischer 344 rats (eight males, eight females) were exposed for 9 days, 6 hours per day, 5 days per week to 2-butoxyethanol target concentrations of 0, 25, 100, or 250 ppm (mean average concentrations determined by gas chromatography to be 0, 20, 86, or 245 ppm, respectively) (Dodd et al. 1983). Rats were observed for signs of toxicity during exposure and for 14 days postexposure. Necropsies were performed on animals that died and on survivors killed at the end of the observation period. Biological assessments included daily animal observations, determination of body and organ weight (kidney, liver, lungs, and testes), and ophthalmologic, hematologic, and gross pathologic examinations. A few male rats exposed to 245 ppm were observed to have audible respiration and

nasal discharge. There was no effect on the hematologic parameters in rats exposed to 20 ppm 2-butoxyethanol. At 86 ppm, both sexes showed evidence of a significant, but less profound, effect on the erythroid parameters. At 245 ppm, both male and female rats showed significantly depressed red blood cell counts (20% below control values), hemoglobin, and mean corpuscular hemoglobin concentration, and significant increases in mean corpuscular volume, nucleated red blood cells, and reticulocytes. Red stained urine (hematuria) was also observed in rats of both sexes exposed to 245 ppm. Female rats exposed to 86 ppm showed significantly increased liver weights. Male and female rats of the 245 ppm group also showed increased liver weights. Decreased mean body weight gains (percentage not provided) were also found. A 14-day postexposure recovery showed substantial reversal of the affected blood parameters.

- An MRL of 3 ppm has been derived for intermediate-duration inhalation exposure (15-364 days) to 2-butoxyethanol.

The MRL was derived from a NOAEL of 25 ppm for hematotoxicity in female rats in a study by Dodd et al. (1983). See Appendix A for further details. The female rat was used since females appeared to have more severe hematological effects than the male rats. In the study by Dodd et al. (1983), Fischer 344 rats (16 males, 16 females) were exposed for 90 days, 6 hours per day, 5 days per week to 2-butoxyethanol target concentrations of 0, 5, 25, or 75 ppm (mean average concentrations determined by gas chromatography were 0, 5, 25, or 77 ppm, respectively). Rats were observed for signs of toxicity during exposure and for 14 days postexposure. Necropsies were performed on animals that died and on survivors killed at the end of the observation period. Biological assessments included daily animal observations, body weight and organ (kidney, liver, lungs, and testes) weight determinations, and ophthalmologic, hematologic, and gross pathologic examinations. In addition, red blood cell osmotic fragility tests, serum chemistries, urinalyses, and histologic examinations of selected tissues of the high 2-butoxyethanol concentrations were performed. No deaths occurred throughout the study. No clinical signs of toxicity were observed. There was a transient decrease in body weight gain (exposure weeks 2-4; data not shown) of females exposed to 77 ppm 2-butoxyethanol. After 6 weeks (3 1 exposures), female rats exposed to 77 ppm had slight but significant decreases (13% below control values) in red blood cell counts and hemoglobin concentrations, accompanied by au 11% increase above control values in mean corpuscular hemoglobin. At the end of the 90-day study, the hematologic effects seen in the exposed female rats either lessened or returned to control value ranges. The only significant hematologic finding in male rats was a 5% decrease in red blood cells at 77 ppm after 66 exposures. The red

blood cell osmotic fragility tests of both 2-butoxyethanol-treated rats and controls were similar. No treatment-related differences were found in body weight, organ weights, urine or serum chemistries, gross lesions, or microscopic lesions in males or females. In light of the results of the Dodd et al. (1983) study of acute-duration inhalation exposure of rats to doses up to 245 ppm, and the more severe hematological effects observed, which were accompanied by changes in liver weight, respiratory effects, and decreased body weight gain, it seems reasonable to accept the hematological effects seen in the intermediate inhalation exposure experiment as being the most sensitive indicator of toxicity. No other suitable supporting studies were found.

- An MRL of 0.2 ppm has been derived for chronic-duration inhalation (≥365 days) to 2-butoxyethanol.

The MRL was derived from NOAEL of 0.6 ppm for decreased hematocrit and increased mean corpuscular hemoglobin concentration observed in 31 male workers exposed to an average concentration of 0.6 ppm for 1 to 6 years (Baufroid et al. 1992). See Appendix A for further details. An uncertainty factor of 3 for human variability was used because the results of *in vitro* data suggest that 2-butoxyethanol does not cause significant hemolysis of normal and potentially susceptible erythrocytes (Udden 1996). In this study, 31 male workers exposed to 2-butoxyethanol were studied. Twenty workers were exposed to an average concentration of 0.75 ppm, and 11 workers were exposed to an average concentration of 0.46 ppm. The weighted average of the means for the two groups of workers is 0.6 ppm. The workers were also exposed to methyl ethyl ketone. Studies in animals indicate that methyl ethyl ketone does not produce hematologic effects (ATSDR 1992). Exposure concentrations of methyl ethyl ketone were not provided. Twenty-one unexposed men that worked for the same company served as controls. Urine was collected before the shift and at the end of the shift and assayed for free 2-butoxyacetic acid, retinol binding protein, and creatinine. Blood was collected and assayed for red blood cell counts, hemoglobin, hematocrit, mean corpuscular volume, mean corpuscular hemoglobin, mean corpuscular hemoglobin concentration, haptoglobin, reticulocytes, and osmotic resistance. Serum alanine aminotransferase and aspartate aminotransferase were also measured.

Urinary concentrations of 2-butoxyacetic acid ranged from not detected to 1.1 mg/g creatinine before shift (average 0.3), and 0.3 to 51.4 mg/g creatinine after shift (average 12.2) in persons exposed to an average of 0.75 ppm. Urinary concentrations of 2-butoxyacetic acid ranged from not detected to 3.4 mg/g creatinine before shift (average 0.6), and 0.6 to 20.4 mg/g creatinine after shift (average 9.2)

in persons exposed to an average of 0.46 ppm. A significant correlation ($r = 0.55$, $p = 0.0012$) was observed between end shift urinary 2-butoxyacetic acid concentrations and 2-butoxyethanol in air. Two small but statistically significant differences in hematology values were observed: a significant decrease ($p=0.03$) in hematocrit values (43.9% exposed, 45.5% controls) and a significant ($p=0.02$) increase in mean corpuscular hemoglobin concentration(33.6 g/dL exposed, 32.9 g/dL controls). These differences are consistent with hemolysis observed in animal studies, and might be early indicators of potential adverse effects in humans. However, because the changes in both hematocrit and mean corpuscular hemoglobin concentration were in the range of normal clinical values, the effect was not considered significant. Normal clinical values of hematocrit are reported as 40-54%, with a level of 30% or less considered to indicate moderate to severe anemia (Fischbach 1992). Normal clinical values of mean corpuscular hemoglobin concentration are reported as 31-37 g hemoglobin/dL (Fischbach 1992). Increased mean corpuscular hemoglobin concentrations often indicates spherocytosis, while decreased mean corpuscular hemoglobin concentrations may indicate macrocytic anemia, chronic blood loss anemia, or pyridoxine-responsive anemia (Fischbach 1992). None of the red blood cell end points were correlated with internal exposure as assessed by free urinary 2-butoxyacetic acid and the amount of conjugated 2-butoxyacetic acid is unknown. Serum creatinine and urinary retinol binding protein were not affected. No difference was observed for serum alanine transaminase or serum aspartate transaminase. No supporting chronic-duration inhalation studies in either animals or humans were identified.

No MRLs were derived specifically for 2-butoxyethanol acetate since human and animal data are limited. Only one acute-duration inhalation study was found in the literature (Truhaut et al. 1979). The study used only one concentration (400 ppm) administered to rats and rabbits for 4 hours. Hemoglobinuria and hematuria were observed in the rabbits but not in the rats. Similarly, only one study was found in the literature for intermediate-duration inhalation exposure (Truhaut et al. 1979). The study used only concentrations of 400 ppm for 1 month and 100 ppm for 10 months in rats and rabbits, and serious effects (hemoglobinuria, hematuria, or renal nephrosis) were observed at both concentrations. No chronic-duration inhalation exposure studies of 2-butoxyethanol acetate were found in the literature. Since 2-butoxyethanol acetateis metabolized to the same toxic metabolite as is 2-butoxyethanol, it seems likely that the MRLs for 2-butoxyethanol acetate would be similar to those of 2-butoxyethanol.

Oral MRLs

- An MRL of 0.4 mg/kg/day has been derived for acute-duration oral (14 days or less) exposure to 2-butoxyethanol.

  The MRL was derived from a LOAEL of 32 mg/kg for hematotoxicity in rats in a study by Ghanayem et al. (1987a). See Appendix A for details. In the study by Ghanayem et al. (1987a), young (4-5 weeks) and adult (9-13 weeks, 5-6 months, and 16 months) rats were dosed with 0,32, 63, 125, 250, or 500 mg/kg 2-butoxyethanol by gavage. Hematotoxicity, metabolism, and excretion of 2-butoxyethanol were monitored for 48 hours. Animals were killed at 48 hours after treatment, and spleen, liver, kidney, testes, and urinary bladder were weighed, fixed, and examined. 2-Butoxyethanol at 125 mg/kg caused severe acute hemolytic anemia in older rats, resulting in significant decreases in red blood cells, hemoglobin, and hematocrit, and increases in the concentration of free plasma hemoglobin. Secondary to the hemolytic effects, 2-butoxyethanol also caused hemoglobinuria in 16-month-old rats at ≥ 32 mg/kg, in 6-month-old rats at ≥ 63 mg/kg, and in younger rats (4-13 weeks) at ≥ 125 mg/kg. Other effects secondary to hemolysis consisted of histopathologic changes in the liver and kidney in adult rats, including focal disseminated coagulative necrosis of hepatocytes at 250 mg/kg and hemoglobin casts in the proximal tubules of the kidney at 125 mg/kg. Relative spleen weight was drastically increased (180-230% of control values). These effects of 2-butoxyethanol were dose- and time-dependent. Both the hemolytic effects and the secondary effects of 2-butoxyethanol were age-dependent, with older rats being more sensitive than younger rats. Since the severity of hemoglobinuria in the 16-month-old rats increased as the dose levels increased and since hemoglobinuria occurred in younger rats only at higher doses, the 32-mg/kg dose was considered to be a less serious LOAEL. The use of 32 mg/kg as a less serious LOAEL for the derivation of the acute oral MRL is supported by a study in pregnant rats in which no hematological effects were found at a similar dose of 30 mg/kg/day given for 3 days during gestation (NTP 1989).

  Hemolytic anemia is the characteristic toxic reaction to 2-butoxyethanol. Sperm-positive Fischer 344 rats (27-33 per dose group) were administered 0, 30, 100, and 200 mg/kg/day 2-butoxyethanol in distilled water by gavage on gestational days 9-l 1 (NTP 1989). The biological effects of 2-butoxyethanol on dams were assessed during treatment as well as 24 hours after treatment and on gestational day 20, and effects on fetuses were observed at sacrifice (gestational day 20); 22-24 females per group were confirmed pregnant. At sacrifice on gestational days 12 or 20, each pregnant female was examined by counting number of corpora lutea, and weighing body,

liver, right and left kidneys, spleen, and uterus. For females sacrificed on gestational day 20, the uterine contents were also evaluated (i.e., number of implantation sites, resorptions, and dead and live fetuses). The fetuses were sexed and examined for external and visceral malformations Maternal and fetal blood (pooled by litter) was analyzed for red blood cell and white blood cell counts, hemoglobin, hematocrit, mean corpuscular hemoglobin, mean corpuscular volume, mean corpuscular hemoglobin concentration, and platelet count. Red blood cell counts and corresponding measures of hematocrit and hemoglobin were significantly reduced in the 100- and 200-mg/kg/day dose groups at approximately 24 hours after the final dose. Mean corpuscular hemoglobin concentration was significantly decreased in the high-dose group. The blood dyscrasia produced at approximately 24 hours after the last treatment also included increases in reticulocytes, white blood cell count, platelet count, mean corpuscular volume, and mean corpuscular hemoglobin in the 100 and 200-mg/kg/day dose groups. On gestationall day 20, maternal hematological end points recovered toward control values, although a majority of values remained different from controls in the 100- and 200-mg/kg/day dose groups. Red blood cell counts measured on gestational day 20 in the 100- and 200-mg/kg/day dose groups of each treatment period were still lower than control values, but counts had increased since cessation of treatment (i.e., in the 200-mg/kg/day dose group, the red blood cell count increased to 91% of control levels by gestational day 20). Similarly, reticulocytes declined from values measured earlier in the 100- and 200-mg/kg/day dose groups but were still significantly above the control level. Corrected white blood cell counts measured on gestational day 20 after dam treatment were similar across dose groups. Platelet counts determined at the end of the study showed a significant decreasing trend with respect to dose which appeared to be determined by a nonsignificant decrease in the high dose compared to the control group. The mean corpuscular volume and mean corpuscular hemoglobin on gestational day 20 exhibited significant dose-related trends, with the values of the two highest dose groups greater than the control values. Calculated values of mean corpuscular hemoglobin concentration showed significant decreasing dose-related trends with the high-dose group values lower than controls. The changes in mean corpuscular hemoglobin concentration were significant in the high-dose group, but were relatively slight in magnitude. The maternal hematological profiles at 24 hours afterfinal treatment and at gestational day 20 indicate that 2-butoxyethanol-induced hemolysis triggered a compensatory hematopoietic response leading to reticulocytosis. Furthermore, recovery was more complete by gestational day 20 for dams treated on gestational days 9-l 1 than for those exposed on gestational days 1 l-13.

In a similar experiment conducted by NTP (1989), sperm-positive Fischer 344 rats (29-31 per dose group) were administered 0, 30, 100, and 300 mg/kg/day 2-butoxyethanol in distilled water by gavage on gestational days 11-13. The biological effects of 2-butoxyethanol on dams were assessed during treatment as well as 24 hours after treatment and on gestational day 20, and effects on fetuses were assessed at sacrifice (gestational day 20). Maternal and fetal blood (pooled by litter) was analyzed for red and white blood cell counts, hemoglobin, hematocrit, mean corpuscular hemoglobin, mean corpuscular volume, mean corpuscular hemoglobin concentration, and platelet count. Red blood cell count and corresponding measures of hematocrit and hemoglobin were significantly reduced in the 100- and 300-mg/kg/day dose groups at approximately 24 hours after the final dose. Mean corpuscular hemoglobin concentration was significantly decreased in the high-dose group. The blood dyscrasia produced at approximately 24 hours after the last treatment also included increases in reticulocytes, white blood cell count, platelet count, mean corpuscular volume, and mean corpuscular hemoglobin in the 100- and 300-mg/kg/day dose groups. On gestational day 20, maternal hematological end points recovered toward control values, although a majority of values remained different from controls in the 100- and 300-mg/kg/day dose groups. Red blood cell counts measured on gestational day 20 in the 100- and 300-mg/kg/day dose groups for each treatment period were still significantly lower than control values, but counts had increased after cessation of treatment (i.e., in the 300-mg/kg/day dose group, the red blood cell count increased from about 45% of control levels at 24 hours after the final treatment to 80% of control levels by gestational day 20). Similarly, reticulocytes declined from values measured earlier in the 100- and 300-mg/kg/day dose groups, but were still significantly above the control level. Corrected white blood cell count measured on gestational day 20 after dam treatment still exhibited a significant increasing trend with respect to dose, which appeared to be determined by a nonsignificant increase in the high-dose group. Platelet counts determined at the end of the study showed a significant increase at the high dose. The mean corpuscular volume and mean corpuscular Remoglobin on gestational day 20 exhibited significant dose-related trends with the values of the two highest dose groups significantly greater than the control values. Calculated values of mean corpuscular hemoglobin concentration showed significant decreasing dose-related trends, with the high-dose group values lower than controls. The changes in mean corpuscular hemoglobin concentration with treatment, although significant at the high-dose group, were relatively slight. The maternal hematological profiles at 24 hours after final treatment and at gestational day 20 indicate that 2-butoxyethanol-induced hemolysis triggered a compensatory hematopoietic response leading to reticulocytosis. Furthermore, recovery was more complete by gestational day 20 for dams treated on gestational days 9-l 1 than those exposed on

gestational days 11-13. Thus, 30 mg/kg/day was the NOAEL for hematological effects in both experiments in the NTP (1989) study, lending support for the use of the LOAEL of 32 mg/kg for hematological effects in the aged rats in the Ghanayem et al. (1987a) study for the derivation of the acute oral MRL.

Other studies show similar effects. In a study by Corley et al. (1994), which was conducted primarily to assist in the validation of a PBPK model (see Section 2.3.5), groups of two or three male Fischer 344 rats were given radiolabeled 2-butoxyethanol at doses of 8.6 or 126 mg/kg by gavage in water. Hemolysis and hemoglobinuria were observed in the two rats given the 126-mg/kg dose, but not in the three rats given 8.6 mg/kg.

In another study (Ghanayem et al. 1992), six male Fischer 344 rats received 125 mg/kg/day 2-butoxyethanol by gavage for 1, 2, 3, 6, or 12 consecutive days. Controls received 5 mL water/kg body weight daily for 12 consecutive days. Twenty-four hours after the last dose, blood samples were collected for hematological measurements and determination of blood ATP concentration in red blood cells in order to confirm previous observations (Ghanayem 1989; Ghanayem et al. 1990b) that hemolysis is preceded by cellular swelling, resulting in increased hematocrits and mean cell volume, and ATP depletion. Spleen and liver were removed and weighed. Organ weight/100 g body weight ratios were calculated. Treatment of rats with 2-butoxyethanol daily (125 mg/kg/day) for l-3 consecutive days resulted in a time-dependent increase in the hemolysis of erythrocytes. However, when daily treatment with 2-butoxyethanol continued beyond 3 days, the number of erythrocytes began to rebound and approached pretreatment levels within 12 days despite continued daily exposure, suggesting development of tolerance to the hemolytic effect of 2-butoxyethanol. Mean cell volumes, ATP concentration, and the number of reticulocytes increased to the 6th day and then decreased on the 12th day. Liver weight/body weight ratios were minimally affected by repeated dosing; they declined on days 3 and 6 but increased on day 12 compared to controls. Spleen weight increased in a time-dependent manner. In other studies, hematotoxic effects were observed at higher doses. For instance, hemolysis was observed in rats receiving one dose of 250 mg/kg/day; reduced red blood cell counts and hemoglobin, and increased mean corpuscular volume, reticulocytes, and mean corpuscular hemoglobin in the presence of increased liver and spleen weight were observed in rats at 500 mg/kg/day after 1 or 4 days (Ghanayem et al. 1987b; Grant et al. 1985). Hepatic and renal histopathology similar to that observed in Ghanayem et al. (1987a) at 125 mg/kg/day was also observed at 500 mg/kg/day after one dose (Ghanayem et al. 1987b).

- An MRL of 0.07 mg/kg/day has been derived for intermediate-duration oral (15-364 days) exposure to 2-butoxyethanol.

  The MRL was derived from a LOAEL of 69 mg/kg/day for hepatic effects in male rats in a study by NTP (1993). See Appendix A for details. In the NTP (1993) study, F344/N rats (10 males, 10 females) were given targeted concentrations of 0, 750, 1,500, 3,000, 4,500, or 6,000 ppm 2-butoxyethanol in drinking water daily for 13 weeks. Actual doses, determined by the study authors from drinking water consumption and body weight data, were as follows: 0, 69, 129, 281, 367, and 452 mg/kg/day for males and 0, 82, 151, 304,363, and 470 mg/kg/day for females. Animals were observed twice daily and weighed at the start of the studies, weekly thereafter, and at necropsy. Clinical observations were recorded weekly. Water consumption by cage was measured twice a week. Complete necropsies were performed on all animals in the base studies. The heart, liver, right kidney, lung, thymus, and right testis were weighed, examined for gross lesions, fixed or embedded in paraffin, and stained for microscopic examination. The protocol for the 13-week studies required that tissues be examined microscopically in all control animals, animals in the highest dose group with at least 60% survivors, and all animals in the higher-dose group (inclusive of early deaths and survivors). In the clinical pathology study, 20 males and 20 females per dose group were used. On days 5 and 21, blood samples were collected for the clinical pathology and hematology studies. Week-13 analyses were conducted on samples obtained from rats in the base studies. Urinalysis was done on week-13 samples collected overnigbt from the base study animals. In the sperm morphology and vaginal cytology evaluations, males were evaluated for necropsy body and reproductive tissue weights and spermatozoa1 data and females were evaluated for necropsy body weight, estrous cycle length, and percentage of cycle spent in the various stages. Hematological effects were noted at ≥82 mg/kg/day in females and at 281 mg/kg/day in males, but no hematological effects were found in males at 69 or 129 mg/kg/day. Hepatic effects were also noted at ≥82 mg/kg/day in females and at ≥69 mg/kg/day in males. The hepatic effects included significantly increased levels of serum alkaline phosphatase in high-dose males (452 mg/kg/day) and in females at 363 and 470 mg/kg/day, and of serum alanine aminotransferase in males at 452 mg/kg/day and in females at 363 and 470 mg/kg/day. Histological examination of livers revealed hepatocellular alterations (hepatocytes that stained more eosinophilic and lacked the amphophilic-to-basophilic granularity of the cytoplasm) at all doses (≥69 mg/kg/day for males and ≥ 82 mg/kg/day for females). The changes in staining may be indications of early degenerative changes in hepatocytes. Centrilobular hepatocellular degeneration was observed in males at ≥281 mg/kg/day and in females at ≥304 mg/kg/day; and brown-to-green granular pigment staining strongly positive for iron in Kupffer cell cytoplasm in

males at 452 mg/kg/day and in females at ≥151 mg/kg/day. Renal effects were also seen in males and females Renal effects included moderate increases in blood urea nitrogen in males at ≥69 mg/kg/day at week 3 and at ≥281 mg/kg/day at week 13 and in females at ≥304 mg/kg/day at week 13; increases in blood creatinine in females at ≥304 mg/kg/day at week 13; decreases in total blood protein in males at ≥281 mg/kg/day at week 13 and in females at ≥151 mg/kg/day at weeks 3 and 13; decreases in blood albumin in males at ≥281 mg/kg/day at week 13 and in females at ≥363 mg/kg/day at week 3 and at ≥304 mg/kg/day at week 13; decreased urine volume in females at ≥82 mg/kg/day at week 13; and increased specific gravity of the urine in males at ≥69 mg/kg/day and in females at ≥151 mg/kg/day at week 13. However, histological examination of the kidneys and urinary bladder revealed no pathological lesions. No hepatic or renal effects were found in $B6C3F_1$ mice in the same study at ≤694 mg/kg/day (males) and ≤1,306 mg/kg/day (females). The renal effects observed in rats may have reflected dehydration since drinking water consumption was decreased. Thus, the 69-mg/kg/day dose in male rats is a less-serious LOAEL for both hepatic and renal effects and therefore is an appropriate dose for the derivation of the intermediate oral MRL.

The only other comprehensive intermediate oral study was conducted in adult male albino rats (COBS CD (SD)BR) that were given undiluted 2-butoxyethanol by gavage in doses of 0 ,222, 443, or 885 mg/kg/day, 5 days per week over a 6-week period (Eastman Kodak 1983; Krasavage 1986). Body weights and feed consumption were recorded. Clinical signs of toxicity and deviations from normal appearance and behavior were observed. Animals were checked daily for mortality, and dead animals were necropsied; tissues (lung, heart, thymus, kidneys, liver, spleen, brain, salivary glands, stomach, cecum, colon, duodenum, jejunum, ileum, pancreas, esophagus, adrenal glands, pituitary, thyroid, parathyroid, trachea, mesenteric lymph nodes, testes, epididymides, prostate, seminal vesicles, coagulating gland, bone marrow, tongue, and nasal cavities) were collected for histologic examination. Eyes were fixed in Zenker's sohttion. Organ weights of liver, kidneys, heart, testes, brain, and spleen were determined. The LOAEL (serious) for hematological effects was 222 mg/kg/day, and there was no NOAEL for that effect. Hepatic and renal effects associated with the hemolytic effects occurred at 443 mg/kg/day (less serious LOAEL), but not at 222 mg/kg/day, and included hemosiderin disposition and an increase in alkaline phosphatase activity.

No MRL was derived for chronic-duration oral exposure because no data were found in the literature.

No oral MRLs were derived for 2-butoxyethanol acetate, specifically, because human and animal data are very scarce. Only one study of acute-duration oral exposure was found in the literature (Truhaut et al. 1979). The doses used were not specified. Thus, the data were not complete enough to derive an MRL. No studies were found for intermediate- or chronic-duration oral exposure. Since 2-butoxyethanol acetate is metabolized to the same toxic metabolite as is 2-butoxyethanol, it seems likely that the MRLs for 2-butoxyethanol acetate would be similar to those for 2-butoxyethanol.

Death. No data are available on death in humans following inhalation or dermal exposure to 2-butoxyethanol or inhalation, oral, or dermal exposure to 2-butoxyethanol acetate. An 87-year-old woman died of a cardiac arrest 3 days after ingesting an unknown amount of disinfectant cleaner containing 6.5% 2-butoxyethanol (Litovitz et al. 1991). Additional case reports of bumans who ingested 2-butoxyethanol at doses of 391-1,862 mg/kg suggest that death could easily occur without supportive therapy (Bauer et al. 1992; Dean and Krenzelok 1992; Gijsenbergh et al. 1989; Rambourg-Scbepens et al. 1988). Coma, respiratory depression, cardiovascular effects, hematuria, hemoglobirmria, metabolic acidosis, and oxaluria have been observed in humans after ingestion of 391-933 mg/kg 2-butoxyethanol (Bauer et al. 1992; Gijsenbergh et al. 1989; Rambourg-Schepens et al. 1988).

Acute- or intermediate-duration inhalation exposure to high concentrations (≥200 ppm) of 2-butoxyethanol (Carpenter et al. 1956; Dodd et al. 1983; Dow 1986; Nelson et al. 1984; Sabourin et al. 1992a; Tyl et al. 1984; Werner et al. 1934a) and 2-butoxyethanol acetate (Truhaut et al. 1979) has caused death in animals. Four-hour LC_{50} values for 2-butoxyethanol were 486 ppm for male rats and 450 ppm for female rats (Dodd et al. 1983). A 7-hour LC_{50} value of 700 ppm has been reported for mice (Werner et al. 1943a). Death in animals has been reported at concentrations of 2-butoxyethanol as low as 250 ppm for 7 hours in nonpregnant female rats (Nelson et al. 1984), 200 ppm for 6 hours per day during gestation for pregnant female rabbits (Tyl et al. 1984), and 400 ppm for 7 hours per day for 1 or 2 days in male rabbits (Dow 1986). Death was also reported in rats exposed to ≥314 ppm, and mice and guinea pigs exposed to ≥376 ppm for 30 days (Carpenter et al. 1956). For 2-butoxyethanol acetate, rabbits, but not rats, died after exposure to 400 ppm intermittently for 1 month (Truhaut et al. 1979).

Death after oral exposure to 2-butoxyethanol and 2-butoxyethanol acetate has been reported in animals. Acute oral LD_{50} values for 2-butoxyethanol range from 530 to 3,000 mg/kg for rats (Carpenter et al. 1956; Eastman Kodak 1988; Nelson et al. 1984; Olin 1976; Smyth et al. 1941; Union Carbide 1980b), from 1,230 to 1,5 19 mg/kg for mice (Carpenter et al. 1956; Eastman Kodak 1988), from 1,200 to 1,414 mg/kg for guinea

pigs (Carpenter et al. 1956; Shepard 1994b; Smytb et all. 194P), and from 320 to 370 mg/kg for rabbits (Carpenter et al. 1956). Death in acute oral studies of 2-butoxyethanol has occurred at doses as low as 200 mg/kg/day for 2 days in male rats (Smialowicz et al. l992), 150 mg/kg/day in pregnant rats (NTP 1989), 1,180 mg/kg/day in pregnant or nonpregnant mice (Hardin et al. 1987; Schuler et al. 1984), and 1,500 mg/kg in pregnant mice (Wier et al. 1987). The acute oral LD_{50} for 2-butoxyethanol acetate in rats is reported to be 2,400 mg/kg for females and 3,000 mg/kg for males (Truhaut et al. 1979). In intermediate oral studies of 2-butoxyethanol, deaths occurred in male rats at 885 mg/kg/day (Eastman Kodak 1983; Krasavage 1986) and in female mice at ≥1,300 mg/kg/day (Heindel et al. 1990; Nagano et al. 1979, 1984).

Death has also been reported in animals exposed dermally to 2-butoxyethanol and 2-butoxyethanol acetate. Acute dermal LD_{50} values in rabbits were reported as low as 99 mg/kg 2-butoxyethanol (Duprat and Gradiski 1979) or 220 mg/kg (Dow 1959), and as high as 406-1,804 mg/kg (Carpenter et al. 1956; Eastman Kodak 1988; Olin 1976; Union Carbide 1980a, 1980b). Death has been observed in rabbits (two of six) at a dose as low as 72 mg/kg 2-butoxyethanol (Duprat and Gradiski 1979). A 4-hour dermal LD_{50} of 2,273 mg/kg 2-butoxyethanol has been reported in females rats (Carpenter and Condra 1961). Rats exhibited 91% mortality after receiving undiluted 2-butoxyethanol on the skin four times daily (1.4 ml/day) during gestation (Hardin et al. 1984). Sixty-five percent of guinea pigs exposed to 2.0 mL 2-butoxyethanol in a single dose died within the 1 st week after exposure (Wahlberg and Boman 1979). The acute dermal LD_{50} for rabbits for 2-butoxy-ethanol acetate has been reported to fall between 4,766 and 5,957 mg/kg (Truhaut et al. 1979).

Exposure of humans to 2-butoxyethanol or 2-butoxyethanol acetate at levels found in the ambient environment or at hazardous waste sites does not seem likely to result in death. However, the existence of many household products containing 2-butoxyethanol or 2-butoxyethanol acetate makes accidental poisoning a potential problem, particularly for children.

Systemic Effects

Respiratory Effects. Respiratory effects have been observed in humans exposed experimentally to 2-butoxy-ethanol and in humans who intentionally ingested 2-butoxyethanol. No studies were located regarding respiratory effects in humans after dermal exposure to 2-butoxyethanol or after exposure by any route to 2-butoxyethanol acetate. While no adverse respiratory effects were reported after a 2-hour inhalation exposure of humans to 20 ppm 2-butoxyethanol (Johanson et al. 1986a), humans exposed for 4 or 8 hours experienced nasal irritation and increased nasal mucus discharge at 113 ppm, and nose and throat irritation at 195 ppm

(Carpenter et al. 1956). In humans who intentionally ingested 2-butoxyethanol, respiratory effects consisted of diffuse pulmonary edema at a dose of 650 mg/kg in a male patient who was an abuser of alcohol and had a history of trichloroethylene ingestion (Bauer et al. 1992), poor ventilation at 467-933 mg/kg in a woman (Rambourg-Schepens et al. 1988), and obstructive respiration at 391-469 mg/kg in another woman (Gijsenbergh et al. 1989).

Respiratory effects have also been observed in animals after acute- and intermediate-duration inhalation exposure; acute, but not intermediate, oral exposure; and acute, but not intermediate, dermal exposure to 2-butoxyethanol. No adverse respiratory effects have been reported in animals after inhalation, oral, or dermal exposure to 2-butoxyethanol acetate (Truhaut et al. 1979); bowever, the data are limited. In acute-duration inhalation studies on 2-butoxyethanol, respiratory effects consisted of rapid and shallow breathing prior to death in rats exposed to 867 ppm for 4 hours, audible respiration and nasal discharge in rats exposed intermittently to 245 ppm for 9 days (Dodd et al. 19839, perinasal encrustation in pregnant rats exposed to ≥100 ppm intermittently on gestational days 6-15 (Tyl et al. 1984), decreased respiratory rate in mice exposed to ≥153 ppm for 10-15 minutes (Kane et al. 198(a), congestion of lungs and nasal turbinates and nasal discharge in rabbits exposed to 400 ppm intermittently for l-2 days (Dow 1986), perinasal wetness in pregnant rabbits exposed to 200 ppm intermittently on gestational days 6-l 8 (Tyl et al. 1984), and congestion of lungs in dogs exposed to 617 ppm intermittently for 2 days and congestion and hemorrhage of the lungs in dogs exposed to 385 ppm for 8 days (Carpenter et al. 1956). In intermediate-duration inhalation studies of 2-butoxyethanol, respiratory effects consisted of congestion and hemorrhage of the lungs in rats at 432 ppm (males) and 314 ppm (females), in guinea pigs at 376 ppm, and in dogs at 200 or 385 ppm for 30 days (Carpenter et al. 1956). Slightly increased nasal secretions were observed in two dogs exposed to 415 ppm 2-butoxyethanol 7 hours per day, 5 days per week, for 12 weeks (Werner et al. 1943b).

In acute oral studies of 2-butoxyethanol, respiratory effects consisted of rapid, shallow, or difficult breathing in rats after a single gavage dose of 2,000 mg/kg/day, a dose that resulted in the death of two of three of the treated rats (Dow 1981), or a single gavage dose of 4,510-9,019 mg/kg (Union Carbide 1980b); congested or hemorrhaged lungs at death in rats after a gavage dose of 530 mg/kg (Carpenter et al. 1956); dyspnea in pregnant rats at 600 mg/kg/day on gestational days 9-11 or 11-1 3 (NTP 1989); and abnormal breathing in pregnant mice at gavage doses of ≥1,500 mg/kg/day on gestational days 8-14 (Wier et al. 1987). In acute dermal studies on 2-butoxyethanol, respiratory effects consisted of congestion and thickening of the alveolar walls in rabbits at ≥72 mg/kg (Duprat and Gradiski 1979), and orange-red lungs in rabbits given a single dose of 0.5 mL/kg 2-butoxyethanol that remained in contact with intact skin for 24 hours (Union Carbide 1980b).

Based on results observed in humans, it seems likely that exposures to high concentrations of 2-butoxyethanol in air of high doses of 2-butoxyethanol would cause respiratory irritation in humans. It also seems likely that oral exposures to high concentrations of 2-butoxyethanol would cause pulmonary edema, poor ventilation, and obstructive respiration. Based on results in dermal studies in animals, skin contact of humans with high doses of 2-butoxyethanol could also cause respiratory effects. Although studies on 2-butoxyethanol acetate were limited, 2-butoxyethanol acetate is readily metabolized to 2-butoxyethanol, indicating that respiratory irritation and other effects might occur in humans exposed to high doses by any route.

Cardiovascular Effects. No studies regarding cardiovascular effects were located in humans after dermal exposure to 2-butoxyethanol or after exposure to 2-butoxyethanol acetate by any route. While no cardiovascular effects, as evidenced by blood pressure and pulse rate measurements and/or electrocardiography, were observed in humans exposed by experimental inhalation exposure to 2-butoxy-ethanol at 98-195 ppm for 4-8 hours (Carpenter et al. 1956) or 20 ppm for 2 hours (Johanson et al. 1986a), case reports of intentional ingestion of household products containing 2-butoxyethanol have indicated tachycardia and low blood pressure in a patient with a history of alcohol and trichloroethylene abuse who ingested 650 mg/kg (Bauer et al. 1992) and low blood pressure in a patient who ingested 391 mg/kg (Gijsenbergh et al. 1989). An 87-year-old woman died from cardiac arrest 3 days after she ingested an unknown amount of a cleaner containing 6.5% 2-butoxyethanol (Litovitz et al. 1991). Before death she also experienced ventricular tachycardia and arrhythmias, and hypotension. Studies in animals that determined heart weights and/or that performed histological examination of the heart and/or the aorta after inhalation (Dodd et al. 1983), oral (Eastman Kodak 1983; Krasavage 1986; NTP 1993) or dermal (CMA 1983) exposure to 2-butoxyethanol or 2-butoxyethanol acetate (Truhaut et al. 1979) did not reveal any adverse effects.

Although information on cardiovascular effects in humans and animals exposed by inhalation to 2-butoxy-ethanol are limited, it is unlikely that exposure of humans to occupational and ambient environmental air levels and air levels at hazardous waste sites of 2-butoxyethanol or 2-butoxyethanol acetate would result in cardiovascular effects. Oral and dermal studies of animals exposed to 2-butoxyethanol and 2-butoxyethanol acetate also indicate little concern for human exposure in environmental and hazardous waste site scenarios. However, the case reports of human ingestion of household products containing 2-butoxyethanol indicate that caution should be taken in the cases of intentional or accidental ingestion of large quantities of 2-butoxyethanol.

Gastrointestinal Effects. The only information regarding gastrointestinal effects in humans after exposure to 2-butoxyethanol or 2-butoxyethanol acetate is the subjective report of occasional belching (eructation) of a male volunteer exposed to 113 ppm for 4 hours and emesis in a female volunteer exposed to 98 ppm 2-butoxyethanol for 8 hours (Carpenter et al. 1956). The subject who experienced emesis, however, believed that the emesis was due to the relatively high chamber temperature based on her personal historical experience that high temperature often caused emesis. However, emesis was observed in a dog exposed intermittently to 617 ppm 2-butoxyethanol for 2 days, a dog exposed to 385 ppm 2-butoxyethanol for 8 days, and a monkey exposed to 210 ppm for 30 days (Carpenter et al 1956). Hemorrhagic gastric ulcers were observed in rabbits exposed intermittently to 400 ppm 2-butoxyethanoI for l-2 days (Dow 1986), and congestion of the abdominal viscera was observed in rats exposed intermittently to 432 ppm (males) or 314 ppm (females) for 30 days (Carpenter et al. 1956).

In oral studies on 2-butoxyethanol in animals, gastrointestinal effects consisted of distended stomachs filled with liquid and gas, and bloody intestines in rats after single gavage doses of 2,255-9,019 mg/kg (Union Carbide 1980b); very red small intestines in rats at a single gavage dose of 2,560 mg/kg (Olin 1976); mild hyperkeratosis and acanthosis of the stomach in rats at gavage doses of 222-885 mg/kg/day for 6 weeks (Eastman Kodak 1983; Krasavage 1986); and diarrhea in rats at 452 mg/kg/day (males) and 470 mg/kg/day (females) in the drinking water for 13 weeks (NTP 1993). Moderate-to-mild necrosis and hemorrhage of the gastric mucosa was observed in guinea pigs treated with a single gavage dose of 1,000 mg/kg 2-butoxyethanol in water, a dose that also resulted in the death of one of each sex (Shepard 1994b). However, histological examination of the gastrointestinal tract revealed no pathological lesions in rats at ≤470 mg/kg/day or mice at ≤ 1,306 mg/kg/day in the 13-week drinking water study (NTP 1993).

In dermal studies on 2-butoxyethanol in animals, gastrointestinal effects consisted of reddened stomachs and intestines in rabbits that died at a dose of 902 mg/kg for 6 hours (Union Carbide 1980a), orange peritonea and intestines in rats that died at a dose of ≥451 mg/kg for 24 hours (Union Carbide 1980b), and very dark intestines in a rabbit dosed with 1,000 mg/kg 2-butoxyethanol on abraded skin (Olin 1976). Although one rabbit that died at 50 mg/kg/day on day 15 in a 90-day dermal study had a gastric ulcer, histological examination of gastrointestinal tracts of the rabbits at ≤150 mg/kg/day revealed no lesions (CMA 1983). Neither the gastric ulcer nor the death was related to 2-butoxyethanol exposure.

No studies were located regarding gastrointestinal effects in humans or animals after exposure to 2-butoxy-ethanol acetate.

The available data in humans and animals indicate that emesis, diarrhea, and ulcers might occur in humans exposed to very high levels of 2-butoxyethanol.

Hematological Effects. Hematological effects of 2-butoxyethanol exposure include hemolysis of red blood cells, and hemoglobinuria-the detection of hemoglobin in the urine resulting from lysed red blood cells (Jones and Hunt 1983). No adverse effect on erythrocyte osmotic fragility, which has been observed in animals, was found in humans experimentally exposed by inhalation to ≤195 ppm 2-butoxyethanol for 4-8 hours (Carpenter et al. 1956). Statistically significant changes in hematocrit (decrease) and mean corpuscular hemoglobin concentration (increase) were reported in men occupationally exposed to 0.46-0.75 ppm of 2-butoxyethanol (Haufroid et al. 1997). The changes were small and within normal biological variability, and so are considered a NOAEL. The effects are consistent with hemolysis observed in animal studies, and they may be indicators of potential adverse effects in humans Hematological effects in humans who ingested household cleaning agents that contain 2-butoxyethanol consisted of nonhemolytic anemia, low prothrombin time, and thrombopenia at 650 mg/kg (Bauer et al. 1992); progressive erythropenia and hemoglobinuria at 467-933 mg/kg (Rambourg-Schepens et al. 1988); and decreased blood hemoglobin at 391-469 mg/kg (Gijsenbergh et al. 1989). Hemodialysis, used in two cases (Bauer et al. 1992; Gijsenbergh et al. 1989), may have contributed to the hematological effects. No particularly sensitive population of humans has been identified; for a discussion of *in vitro* results with hypothetically sensitive human red blood cells see section 2.8 Populations That Are Unusually Susceptible.

Hematological effects of 2-butoxyethanol have been observed in animals after inhalation, oral, and dermal exposure. In inhalation studies, the hematological effects observed in animals include increased erythrocyte osmotic fragility in monkeys at 210 ppm for 30 days and at 100 ppm for 90 days, in rats at 62 ppm for 4 hours or ≥ 54 ppm for 30 days, in mice at 100 ppm for 7 hours and ≥100 ppm for 30-90 days, in rabbits at 125 ppm for 7 hours, and in dogs at 200-385 ppm for 8-30 days (Carpenter et al. 1956). Other hematological effects related to hemolysis included hemoglobinuria and hemin crystalluria in rats at 432 ppm for 2-8 hours, decreased red blood cell counts and hemoglobin levels in rats at 200 ppm for 7 hours per day for 9 days, hemoglobinuria in rats at 203 ppm for a single 7-hour exposure or for 7 hours per day for 30 days, at 250-800 ppm for 2-9 hours per day for l-6 days (Carpenter et al. 1956), and at 438 ppm for 6 hours (Sabourin et al. 1992a); hemoglobinuria, reduced red blood cells and mean corpuscular hemoglobin concentration, and increased mean corpuscular hemoglobin and mean corpuscular volume in pregnant rats at ≥100 ppm during gestation, and increased hemoglobin and hematocrit in pregnant rabbits at 100 ppm during gestation (Tyl et al. 1984); decreased hematocrit in dogs at 100 ppm for 90 days (Carpenter et al. 1956);

decreased hemoglobin, increased mean corpuscular volume, and decreased mean corpuscular hemoglobin concentration in rats at 2 86 ppm and depressed red blood cell counts and increased nucleated red blood cells and reticulocytes at 245 ppm for 6 hours per day for 9 days (Dodd et al. 1983), and decreased red blood cell counts, hemoglobin levels, and mean corpuscular hemoglobin concentration at 77 ppm for 13 weeks (Dodd et al. 1983). Decreased hemoglobin and hematocrit, and hypochromia, polychromatophilia, and microcytosis were observed in two dogs exposed to 415 ppm 2-butoxyetbanol 7 hours per day, 5 days per week, for 12 weeks (Werner et al. 1943b).

In oral studies on 2-butoxyethanol in animals, similar hematological effects have been observed, consisting of hemoglobinuria in rats at ≥ 1,500 mg/kg (Carpenter et al. 1956); hemoglobinuria, hemolysis, and increased concentration of free hemoglobin in plasma in rats at 500 mg/kg (Ghanayem et al. 1987b); and hemolysis, including decreased red blood cell counts, increased hematocrit, packed cell volumes, and mean cell volume in rats at ≥125 mg/kg (Corley et al. 1994; Ghanayem et al. 1987b, 1990,1992; Ghanayem and Sullivan 1993; Sivarao and Mehendale 1995). Similar effects related to erythrocyte hemolysis have been found in rats at 500 mg/kg/day for 4 days (Grant et al. 1985) and in pregnant rats at ≥ 100 mg/kg/day during gestation (NTP 1989). Hematological effects reflecting hemolysis have also been found in animals after intermediate-duration oral exposure to 2-butoxyethanol: decreased red blood cell counts and hemoglobin, increased mean corpuscular hemoglobin, and hemoglobinuria at ≥222 mg/kg/day for 6 weeks (Eastman Kodak 1983; Krasavage 1986); decreased red blood cell counts in mice at ≥ 500 mg/kg/day for 5 weeks (Nagano et al. 1979, 1984); and decreased red blood cell count, hematocrit, and hemoglobin in rats at ≥ 82 mg/kg/day in drinking water for 13 weeks (NTP 1993).

In dermal studies of 2-butoxyethanol in animals, hemolytic effects have also been found as follows: hemolysis and hemoglobinuria in rats at ≥ 260 mg/kg and increased mean corpuscular volume, decreased red blood cell count, and decreased hemoglobin in rats at 500 mg/kg applied to the skin for 6 hours (Bartnik et al. 1987); hemoglobinuria in pregnant rats at 1.4 mL/day applied during gestation (Hardin et al. 1984); increased erythrocyte osmotic fragility in rabbits after a 3minute skin contact at 505 mg/kg (Carpenter et al. 1956); and hemoglobinuria in rabbits at ≥ 72 mg/kg on the skin for 6-24 hours (Carpenter et al. 1956; Duprat and Gradiski 1979; Union Carbide 1980a) or hemoglobinuria with hemolytic effects on erythrocytes in rabbits at 361 mg/kg/day for 11 days (Union Carbide 1980a).

Similar hemolytic effects have been observed in animals after inhalation, oral, and dermal exposure to 2-butoxyethanol acetate (Truhaut et al. 1979). Hemoglobinuria was observed in rats and rabbits exposed by

inhalation to 400 ppm for 4 hours per day for 1 month- Rabbits also had decreased red blood cell counts and hemoglobin concentration. In an oral study, severe hemoglobinuria was observed in rats given an unspecified single dose. A decrease in hemoglobin and red blood cell count and hemoglobinuria occurred in rabbits after dermal application of ≥ 3,191 mg/kg 2-butoxyethanol acetate.

The hemolytic properties of 2-butoxyethanol have been found to be age-related in F344 rats, with greater sensitivity of rats 16 months old than of rats 5-6 months old, followed by rats 9-13 weeks old. Rats 4-6 weeks old were the least sensitive (Ghanayem et al. 1987a). Furthermore, *in vitro* experiments with human and animal erythrocytes indicate species-dependent sensitivity to the hemolytic effects of 2-butoxyethanol, with rodents, rabbits, and baboons more sensitive than pigs, dogs, cats, guinea pigs, and humans (Bartnik et al. 1987; Ghanayem and Sullivan 1993). In addition, an *in vitro* study showed that human erythrocytes were resistant to the hemolytic effects of 2-butoxyacetic acid, the hemolytic metabolite of 2-butoxyethanol (Udden 1994).

In addition to the hemolytic effects of 2-butoxyethanol, other hematological effects, which also appear to be age-related, were observed in young rats (4-5 weeks old) and adult rats (9-l 3 weeks old) at an oral dose of 500 mg/kg (Ghanayem et al. 1987a). Adult rats exhibited neutrophilic leukocytosis and mild lymphopenia within 4 hours, but not 48 hours. In young rats, an absolute increase in total leukocytes, band neutrophils, lymphocytes, and monocytes was observed at 48 hours.

In vitro data show that human red blood cells are much less sensitive to the hemolytic effects of the 2-butoxyethanol metabolite, 2-butoxyacetic acid. Concentrations of 2-butoxyacetic acid that caused hemolysis of rat red blood cells did not cause hemolysis of human red blood cells (Bartnik et al. 1987). For example, following a 60minute exposure of rat red blood cells to 7.5 mM 2-butoxyacetic acid 100% hemolysis was observed. Following a 60minute exposure of human red blood cells to 15 mM 2-butoxyacetic acid, no hemolysis was observed (Bartnik et al. 1987). An *in vitro* study measuring more sensitive end points of red blood cell effects (mean cell volume and hematocrit) has also shown rat red blood cells to be more sensitive to 2-butoxyacetic acid compared to human red blood cells (Ghanayem and Sullivan 1993). For example, following a l-hour exposure to 2 mM 2-butoxyacetic acid, mean corpuscular volume was about 150% of controls in rat red blood cells and 105% of controls in human red blood cells, and hematocrit was about 155% of controls in rat red blood cells and 105% of controls in human red blood cells. Red blood cells of rabbits, hamsters, mice, and baboons were also sensitive to the red blood cell effects of 2-butoxyacetic acid, while red blood cells of cats, pigs, dogs, and guinea pigs were not sensitive to the effects of 2-butoxyacetic acid

(Ghanayem and Sullivan 1993). The molecular mechanism for the resistance of red blood cells in some species but not others is not known. Until the mechanism is known, it has been suggested that 2-butoxyethanol be treated as a chemical with the potential for causing human toxicity (Ghanayem 1996).

The hematological and, in particular, the hemolytic effects of 2-butoxyethanol are the most consistent and most sensitive effects observed in animals. Although humans may be less sensitive than rats and rabbits, which are the most consistently tested animals, concern exists that humans exposed to high levels of 2-butoxyethanol may develop hemolytic effects.

Musculoskeletal Effects. No studies were located regarding musculoskeletal effects in humans after exposure to 2-butoxyethanol or 2-butoxyethanol acetate by any route, or in animals after exposure to 2-butoxyethanol acetate by any route. Muscular flaccidity, which may represent neurotoxicity rather than a direct effect on the muscle, was observed in rats that died at single gavage doses of ≥ 2,560 mg/kg and in rabbits that died after dermal doses of ≥1,000 mg/kg of 2-butoxyethanol (Olin 1976). In 13-week studies, histological examination of skeletal muscle and bone revealed no pathological lesions in rats exposed intermittently by inhalation to ≤77 ppm (Dodd et al. 1983), in rats receiving oral doses of ≤470 mg/kg/day and mice receiving oral doses of ≤ 1,306 mg/kg/day in drinking water (NTP 1993), or in rabbits exposed dermally to ≤ 150 mg/kg/day (CMA 1983).

Musculoskeletal effects are unlikely to occur in humans exposed to 2-butoxyethanol in environmental or hazardous waste site scenarios.

Hepatic Effects. No effects on serum alanine aminotransferase or aspartate aminotransferase were observed in 31 male workers exposed to 0.46-0.75 ppm 2-butoxyethanol for l-6 years (Haufroid et al. 1997). Case reports provide evidence that 2-butoxyethanol can result in hepatic effects following high oral doses. In a man who ingested 2 doses of a concentrated glass cleaner containing 2-butoxyethanol 12 days apart, increased serum alanine aminotransferase, aspartate aminotransferase, and bilirubin were observed following the first ingestion, but not following the second ingestion (Gualtieri et al. 1995). Hepatic failure was reported in an 87-year-old woman who died of cardiac arrest 3 days after ingesting an unknown volume of glass cleaner containing 6.5% 2-butoxyethanol (Litovitz et al. 1991). Abnormal liver function was noted in a man who had ingested 650 mg/kg 2-butoxyethanol (Bauer et al. 1992). The man was a known alcohol and trichloroethylene abuser, which probably contributed strongly to the abnormal liver function.

Hepatic effects have been observed in animals after inhalation, oral, and dermal exposure to 2-butoxyethanol. In inhalation studies in animals exposed to 2-butoxyethanol, hepatic effects consisted of increased liver weight in rats exposed intermittently for 9 days to ≥ 86 ppm (females) and 245 ppm (males) (Dodd et al. 1983) or intermittently to ≥ 107 ppm for 30 days (Carpenter et al. 1956); mottled livers in rabbits exposed to 400-411 ppm intermittently for 1 or 2 days (Dow 1986); congestion of the liver in dogs exposed for 8 or 28 days to 385 ppm (Carpenter et al. 1956); and cloudy swelling of the liver in rats exposed intermittently to ≥ 314 ppm for 30 days (Carpenter et al. 1956).

In oral studies in animals exposed to 2-butoxyethanol, hepatic effects consisted of mottled livers in rats that died at gavage doses ≥530 mg/kg (Carpenter et al. 1956); focal coagulative necrosis of hepatocytes accompanied by phagocytized hemoglobin in adult rats, but not young rats, at ≥250 mg/kg (Ghanayem et al. 1987a, 1987b); dark livers in rats at 1,310 mg/kg (Olin 1976) or 2,255 mg/kg (Union Carbide 1980b); increased liver weight in rats at 125 mg/kg/day for up to 12 days (Ghanayem et al. 1992), at 500 mg/kg/day 2-butoxyethanol for 4 days (Grant et al. 1985), at 180 mg/kg/day in the drinking water for 21 days (Exon et al. 199 l), at 1,540 mg/kg/day in the diet for 90 days (Carpenter et al. 1956), and at ≥ 222 mg/kg/day by gavage for 6 weeks (Eastman Kodak 1983; Krasavage 1986); decreased absolute liver weight in pregnant rats at ≥200 mg/kg/day during gestation (NTP 1989); and hemosiderin deposition in the liver and increased serum alkaline phosphatase levels at ≥ 443 mg/kg/day and hepatocytomegaly and increased serum alanine aminotransferase levels at 885 mg/kg/day in rats dosed for 6 weeks (Eastman Kodak 1983; Krasavage 1986). In rats treated with 2-butoxyethanol in the diet for 91-93 days, relative liver weights were increased by 25% in males at 919 mg/kg/day and by 27% in females at 976 mg/kg/day (Weil 1963). No effects on liver weight were noted at lower doses (188 mg/kg/day males, 222 mg/kg/day females), nor were any histopathological changes observed at any dose. Serum alkaline phosphatase and alanine aminotransferase levels were also increased in rats at ≥363 mg/kg in drinking water for 13 weeks (NTP 1993). In addition, hepatocellular alterations (hepatocytes that stained more eosinophilic and lacked the amphophilic to basophilic granularity of the cytoplasm) occurred at ≥ 69 mg/kg/day, centrilobular hepatocellular degeneration occurred at ≥ 281 mg/kg/day, and brown to green granular pigment staining strongly positive for iron in Kupffer's cell cytoplasm occurred at ≥ 151 mg/kg/day in the rats in the 13-week drinking water study. These lesions, particularly in the females, displayed both increased dose-related incidence and increased dose-related severity. None of these effects occurred in mice similarly exposed (NTP 1993).

In dermal studies on 2-butoxyethanol in animals, hepatic effects consisted of pale livers in rabbits that died after 24-hour skin contact with ≥406 mg/kg (Carpenter et al 1956), congestion in the liver with necrotic foci

and inconstant (i.e., not always present) steatosis in rabbits that died at ≥ 72 mg/kg 2-butoxyethanol (Duprat and Gradiski 1979), mottled livers with pocked surfaces in rabbits at ≥ 451 mg/kg for 6 hours (Union Carbide 1980a) or 24 hours (Union Carbide 1980b), and discolored or pale liver in rabbits dosed with 250-1,000 mg/kg on abraded skin (Olin 1976). However, histological examination of liver and gall bladder revealed no lesions in rabbits at ≤150 mg/kg/day 2-butoxyethanol for 90 days (CMA 1983). In addition, no effects on liver weight were observed.

Limited data are available on hepatic effects in animals exposed to 2-butoxyethanol acetate or 2-butoxyacetic acid. Histological examination of the livers revealed no pathological lesions in rats given unspecified acute oral doses of 2-butoxyethanol acetate or in rabbits given dermal applications of ≤ 10,000 mg/kg of 2-butoxy-ethanol acetate (Truhaut et al. 1979). Likewise, no histopathological lesions were found in the livers of rats dosed by gavage with ≤ 868 mg/kg 2-butoxyacetic acid (Foster et al. 1987).

The data in animals indicate that hepatic effects might occur in humans exposed to very high levels of 2-butoxyethanol.

Renal Effects- No effects on serum creatinine or urinary retinol binding protein were observed in 3 1 male workers exposed to 0.46-0.75 ppm 2-butoxyethanol for l-6 years (Haufroid et al. 1997). Renal effects observed in persons who intentionally ingested bousehold cleaning agents containing 2-butoxyethanol include albuminuria in a known alcohol and trichloroethylene abuser who ingested 650 mg/kg 2-butoxyethanol (Bauer et al. 1992); oxaluria and increased serum creatinine in a patient who ingested 467-933 mg/kg (Rambourg-Schepens et al. 1988); and hematuria in a patient who ingested 391-469 mg/kg (Gijsenbergh et al. 1989), and renal failure in an 87-year-old woman who died after ingesting an unknown amount of 2-butoxyethanol (Litovitz et al. 1991).

Similar renal effects have been observed in animals after inhalation, oral, and dermal exposure to 2-butoxy-ethanol, with hematuria, the detection of red blood cells in the urine (Jones and Hunt 1983) being a prevalent finding. In inhalation studies of 2-butoxyethanol in animals, renal effects consisted of evidence of hematuria (red-stained urine) and enlarged kidneys in rats exposed to ≥523 ppm for 4 hours or to 245 ppm, 6 hours per day for 9 days (Dodd et al. 1983); hematuria in rats exposed to 800 ppm for 3 hours (Doe 1984), to 250 ppm for 7 hours (Nelson et al. 1984), or to 100 ppm for 6 or 7 hours per day during gestation (Nelson et al. 1984; Tyl et al. 1984); evidence of hematuria in rabbits at ≥100 ppm for 6 hours per day during gestation (Tyl et al. 1984) or 400 ppm for 7 hours per day for l-2 days (Dow 1986); cloudy swelling of the renal convoluted

tubules in rats at 432 ppm for 2 hours and in guinea pigs at ≥376 ppm for 7 hours per day for 30 days (Carpenter et al. 1956); congestion of the kidneys in dogs at 617 ppm for 2 days and 385 ppm for 8 days (Carpenter et al. 1956); and increased kidney weight in rats at ≥ 107 ppm and in guinea pigs at ≥203 ppm, 7 hours per day for 30 days (Carpenter et al. 1956).

In oral studies of 2-butoxyethanol in animals, renal effects consisted of severely congested kidneys in rats at gavage doses ≥500 mg/kg (Carpenter et al. 1956; Sivarao and Mehendale 1995); dark and/or enlarged kidneys in rats at 1,310 mg/kg (Olin 1976); hematuria in rats at ≥ 500 mg/kg (Dow 1959; Olin 1976; Union Carbide 1980b) and in pregnant rats at 600 mg/kg/day during gestation (NTP 1989); and increased kidney weights in rats at ≥ 500 mg/kg/day for 4 days (Grant et al. 1985) and at 1,540 mg/kg/day in the diet for 90 days (Carpenter et al. 1956), and in mice at ≥ 700 mg/kg/day in the drinking water in a continuous breeding study (Heindel et al. 1990). In a 13-week drinking water study, renal effects were found in rats, but not in mice (NTP 1993). The effects in rats consisted of increased blood urea nitrogen at ≥ 69 mg/kg/day, increased blood creatinine at ≥304 mg/kg/day, decreased total blood protein at ≥ 151 mg/kg/day, decreased blood albumin at ≥ 281 mg/kg/day, decreased urine volume at ≥ 82 mg/kg/day, and increased specific gravity of the urine at ≥ 69 mg/kg/day. However, histological examination of the kidneys and urinary bladders of the rats revealed no pathological lesions. In rats treated with 2-butoxyethanol in the diet for 91-93 days, relative kidney weights were increased in by 18% in males at 919 mg/kg/day, and 23% in females at 976 mg/kg/day (Weil and Carpenter 1963). No effects on kidney weight were noted at lower doses (188 mg/kg/day males, 222 mg/kg/day females), nor were any histopathological changes observed at any dose.

Renal effects that are secondary to the hemolytic effects of 2-butoxyethanol were found in rats after oral exposure, as evidenced by hemosiderin accumulation in the proximal tubules at ≥443 mg/kg/day by gavage for 6 weeks (Eastman Kodak 1983; Krasavage 1986) and hemoglobin casts in the proximal tubules at ≥ 125 mg/kg by gavage (Ghanayem et al. 1987a). The finding of hemoglobin casts in the proximal tubules was found to be age-related, with adult rats (≥ 9 weeks old) being more sensitive than immature rats (Ghanayem et al. 1987a). This is consistent with the age-related hemolytic effects of 2-butoxyethanol, as discussed above under Hematological Effects.

In dermal studies of 2-butoxyethanol in animals, renal effects consisted of congested kidneys in rabbits after 24-hour skin contact with ≥ 406 mg/kg (Carpenter et al. 1956); enlarged kidneys with hemoglobinuric nephrosis and interstitial reactions (secondary to hemolysis) in rabbits at ≥72 mg/kg for 8-hour dermal contact (Duprat and Gradiski 1979); hematuria, enlarged dark kidneys with pocked surfaces, or discolored kidneys in

rabbits at ≥451 mg/kg for 6 hours (Union Carbide 1980a) or 24 hours (Olin 1976; Union Carbide 1980b); and hematuria at ≥180 mg/kg/day for 9 dermal6-hour applications (Union Carbide 1980a). In the rabbits given 9 dermal applications of 2-butoxyethanol, other renal effects consisted of increased urinary protein at 361 mg/kg/day and dimpling of the renal surface, tubular vacuolization, tubular degeneration, tubular cell hyperplasia, glomerular adhesions, and interstitial nephritis at ≥ 271 mg/kg/day (Union Carbide 1980a).

Renal effects have also been observed in animals after inhalation, oral, and dermal exposure to 2-butoxyethanol acetate and after oral exposure to 2-butoxyacetic acid. In an inhalation study of 2-butoxyethanol acetate, renal effects consisted of slight hematuria in rabbits, but not rats, exposed to 400 ppm for 4 hours; hematuria and tubular nephrosis in rats and rabbits at 400 ppm, 4 hours per day for 1 month; and tubular nephritis with tubular enlargement and Henle's loop dilation, cortical atrophy, and inflammatory fibrosis in rats and rabbits at 100 ppm, 4 hours per day for 10 months (Truhaut et al. 1979). Similar effects were seen in rats given unspecified single oral doses of 2-butoxyethanol acetate by gavage and in rabbits exposed dermally to ≥ 3,191 mg/kg of 2-butoxyethanol acetate. Hematuria was also found in rats given a single gavage dose of 2-butoxyacetic acid at 868 mg/kg (Foster et al. 1987).

The finding of hematuria in a patient who ingested 2-butoxyethanol and the consistent finding of hematuria in animals after inhalation, oral, and dermal exposure to 2-butoxyethanol and 2-butoxyethanol acetate and after oral exposure to 2-butoxyacetic acid strongly suggests that hematuria is a concern for humans exposed to high levels of 2-butoxyethanol.

Endocrine Effects. No studies were located regarding endocrine effects in humans after inhalation, oral, or dermal exposure to 2-butoxyethanol or 2-butoxyethanol acetate. Severely hemorrhaged adrenal glands were found in a dog that died after exposure to 385 ppm for 8 days (Carpenter et al. 1956). Histological examination of the adrenal, parathyroid, thyroid, or pituitary glands showed no lesions in rats at ≤ 77 ppm, 6 hours per day, 5 days per week for 13 weeks (Dodd et al. 1983). In oral studies on 2-butoxyethanol in animals, red adrenal glands, which may be related to the hemolytic effects, were seen in rats at 2,255-9,019 mg/kg (Union Carbide 1980b). Histological examination of the pancreas, adrenal glands, pituitary, thyroid, or parathyroid revealed no lesions in rats at ≤885 mg/kg/day for 6 weeks, 5 days per week (Eastman Kodak 1983; Krasavage 1986) or in rats at ≥470 mg/kglday and mice at ≤ 1,306 mg/kg/day in drinking water for 13 weeks (NTP 1993). Likewise, in dermal studies, reddened adrenal glands, probably related to hemolysis, were seen in rabbits at ≥ 451 mg/kg for 6 hours (Union Carbide 1980a), but histological

examination of adrenals, pancreas, pituitary, thyroid, or parathyroid glands revealed no lesions in rabbits at ≤ 150 mg/kg/day 2-butoxyethano16 hours per day, 5 days per week for 90 days (CMA 1983).

Similarly, in studies on 2-butoxyethanol acetate, no gross or histologically observed lesions were noted in the pancreas or adrenal glands of rats and rabbits exposed by inhalation for 4 hours per day, 5 days per week to 400 ppm for 1 month, or to 100 ppm for 10 months in rats at an unspecified single gavage dose, or in rabbits at dermal doses ≤ 10,000 mg/kg for 24 hours (Truhaut et al. 1979).

Since the only endocrine effects were hemorrhaged or reddened adrenal glands in animals exposed to high inhalation concentrations and high oral or dermal doses of 2-butoxyethanol, it is unlikely that humans exposed to 2-butoxyethanol or 2-butoxyethanol acetate in environmental, or hazardous waste site scenarios would experience endocrine effects.

Dermal Effects. No studies were located regarding dermal effects in humans after inhalation or oral exposure to 2-butoxyethanol, but dermal effects in humans after direct dermal exposure to neat 2-butoxyethanol included rigidity of the skin and decreased skin vohrme and skinfold thickness (Johanson et al. 1988). Slight erythema was noted during skin patch tests with 2-butoxyethanol acetate in humans (Jacobs et al. 1989), although a human sensitivity study in 201 persons with 10% 2-butoxyethanol showed no overt effect (CMA 1992; Greenspan et al. 1995). The 10% concentration was tested because it is the highest concentration found in cosmetics. Concentrations as high as 50% 2-butoxyethanol can be found in cleaning products which are intended to be diluted before use (OECD 1997).

In studies of 2-butoxyethanol in animals, dermal effects included necrosis of the tail tip in nonpregnant female rats at ≥250 ppm for up to 7 hours (Nelson et al. 1984) and in pregnant rats at 200 ppm for 6 hours per day during gestation (Tyl et al. 1984) in inhalation studies; in rats after a single oral dose of 1,000 mg/kg (Dow 1981); and in pregnant rats after dermal contact with 1.4 mL/day during gestation (Hardin et al. 1984). The cause of necrosis of the tail tip was not clearly stated, but possibly it is associated with the effects of 2-butoxy-ethanol on the red blood cells and subsequent vascular response.

In other oral studies of 2-butoxyethanol in animals, dermal effects consisted of rough coat in rats that died at ≥ 530 mg/kg (Carpenter et al. 1956), piloerection in rats at >1,3 10 mg/kg (Olin 1976), and pale coloration of the skin in rats at 600 mg/kg/day during gestation (NTP 1989). The rough coats and piloerection may be

related to neurotoxicity. Histological examination of skin revealed no dermal lesions in rats at ≤470 mg/kg/day or in mice at ≤ 1,306 mg/kg/day (females) in drinking water for 13 weeks (NTP 1993).

In other dermal studies of 2-butoxyethanol in animals, dermal effects consisted of strong skin irritation in guinea pigs after application of unspecified doses (Eastman Kodak 1988); necrosis of epidermis and dermis in rabbits at ≥ 72 mg/kg for 8 hours (Duprat and Gradiski 1979); hyperemia, edema, slight exfoliation, and/or slight to moderate irritation in rabbits after dermal contact with undiluted or unspecified concentrations of 2-butoxyethanol (Dow 1958, 1959,1981; Eastman Kodak 1988; Rohm and Haas 1983); necrosis and erythema in rabbits at ≥541 mg/kg for 6 hours (Union Carbide 1980a), at 902 mg/kg for 24 hours (Union Carbide 1980b), and at ≥18 mg/kg (for erythema) and ≥271 mg/kg (for necrosis) after nine applications (Union Carbide 1980a); slight to moderate erythema in rabbits at ≥ 10 mg/kg/day for 6 hours per day for 90 days (CMA 1983); and capillary congestion in rabbits after dermal contact with 0.01 mL of undiluted 2-butoxyethanol (Union Carbide 1980b).

The dermal irritation of 2-butoxyethanol and 2-butoxyethanol acetate have been studied in New Zealand rabbits using both the Draize protocol (24-hour occluded exposure) and the European Economic Communities protocol (4-hour occluded exposure) (Zissu 1995). For both protocols, 0.5 mL of undiluted compound was placed on the skin. 2-Butoxyethanol was considered a severe irritant by the Draize protocol, and an irritant by the European Economic Communities protocol. 2-Butoxyethanol acetate was considered a moderate irritant by the Draize protocol, and non-irritating by the European Economic Communities protocol.

In dermal studies of 2-butoxyethanol acetate in animals, mild to moderate erythema, indicating mild irritation, was observed in rabbits after dermal contact with undihtted 2-butoxyethanol acetate (Jacobs et al. 1989; Truhaut et al. 1979).

The data indicate that skin contact of humans with high concentrations of 2-butoxyethanol or 2-butoxyethanol acetate may result in skin irritation.

Ocular Effects. Eye irritation was experienced by humans exposed experimentally by inhalation to 2-butoxy-ethanol at ≥ 113 ppm for 4-8 hours (Carpenter et al. 1956), probably due to direct contact of the eyes with the 2-butoxyethanol vapor. A woman who ingested 391-469 mg/kg 2-butoxyethanol exhibited isocoric light reactive mydriasis (Gijsenbergh et al. 1989).

Ocular effects have been observed in animals after inhalation, oral, dermal, and ocular exposure to 2-butoxyethanol. Periocular wetness occurred during inhalation exposure of pregnant rats at ≥ 25 ppm and in pregnant rabbits at ≥ 100 ppm during gestation (Tyl et al. 1984). The periocular wetness was probably due to direct contact of the eyes with 2-butoxyethanol vapor. Slightly increased ocular secretions were observed in 2 dogs exposed to 415 ppm 2-butoxyethanol intermittently for 12 weeks (Werner et al. 1943b). A reddish ocular discharge accompanied by yellow discoloration of the sclera occurred in rabbits exposed to 400-411 ppm for 7 hours per day for 1-2 days (Dow 1986); these effects may have been secondary to the hemolytic property of 2-butoxyethanol. In other inhalation studies of 2-butoxyethanol in animals, ophthalmological examination revealed no effects in dogs exposed to 200 ppm for 31 days (Carpenter et al. 1956) or in rats exposed for 6 hours per day to ≤245 ppm for 9 days or to ≤77 ppm for 13 weeks (Dodd et al. 1983).

In oral studies of 2-butoxyethanol in animals, ocular effects consisted of palpebral closure in rats at 2,000 mg/kg (Dow 1981) and chromodacryorrhea in pregnant rats at 600 mg/kg/day during gestation (NTP 1989). However, histological examination of the eyes revealed no lesions in rats treated with ≤885 mg/kg/day by gavage for 6 weeks (Eastman Kodak 1983; Krasavage 1986), or in rats at ≤470 mg/kg/day, or mice at ≤ 1,306 mg/kg/day in the drinking water for 13 weeks (NTP 1993).

In studies of 2-butoxyethanol instilled into the eyes of rabbits, severe irritation, moderate to extensive conjunctivitis, moderate cornea1 damage, and/or slight iritis were observed (Dow 1958, 1959,198 1; Kennah et al. 1989a; Rohm and Haas 1983; Union Carbide 1980b). In dermal studies, grey iris was observed in rabbits at 902 mg/kg for 6 hours (Union Carbide 1980a) or for 24 hours (Union Carbide 1980b); lacrimation at 2,000 mg/kg and a yellow cornea at 500 mg/kg occurred in rabbits (Olin 1976). For 2-butoxyethanol acetate, slight conjunctival redness and discharge were observed in rabbits after ocular instillation (Truhaut et al. 1979).

In a study designed to evaluate the use of *in vitro* cytotoxicity data for predicting the ocular irritancy potential of chemicals, BALB/c 3T3 cells were grown overnight, then exposed for 30 minutes to 10-100% (volume in culture) 2-butoxyethanol (Kennah et al. 1989b). Growth of the cells with 2-butoxyethanol compared to a control culture was recorded. The concentration at which the growth of the cells was inhibited by 50% compared to controls was calculated and compared to *in vivo* eye irritancy tests (Kennah et al. 1989a). A linear relationship was observed, although the *in vivo* tests indicated greater irritancy than would have been predicted from the cytotoxicity tests. Other *in vitro* test systems have been investigated using 2-butoxyethanol, with less than satisfactory results (Bagley et al. 1994; Booman et al. 1988).

The data indicate that exposure to vapors of 2-butoxyethanol or direct ocular contact with liquid 2-butoxy-ethanol would be irritating to the eyes of humans.

Body Weight Effects. No studies were located regarding body weight effects in humans after inhalation, oral, or dermal exposure to 2-butoxyethanol or 2-butoxyethanol acetate. However, effects on body weight have been observed in animals after inhalation, oral, and dermal exposure to 2-butoxyethanol or after oral exposure to 2-butoxyacetic acid.

In inhalation studies on 2-butoxyethanol in animals, weight loss, or decreased body weight gain was observed in rats at ≥523 ppm for 4 hours and at ≥ 83 ppm for 9 days (Dodd et al. 1983); in pregnant rats at 2 100 ppm and pregnant rabbits at 200 ppm for 6 hours per day during gestation (Tyl et al. 1984); in dogs at 385 ppm for 8 or 25 days (Carpenter et al. 1956); and in mice at 400 ppm for 60 days (Carpenter et al. 1956). Two dogs exposed to 415 ppm 2-butoxyethano17 hours per day 5 days per week for 12 weeks gradually lost 6-9% of their body weights (Werner et al. 1943b). No control body weight data were provided, although the study authors indicate that the other dogs in the study gained weight or remained constant throughout the study period.

In oral studies of 2-butoxyethanol in animals, weight loss, or reduced body weight gain was observed in rats at a gavage dose of 1,000 mg/kg/day for 4 days (Grant et aI 1985), at 885 mg/kg/day by gavage for 6 weeks (with decreased food consumption) (Eastman Kodak 1983; Krasavage 1983), at 265 mgIkg/day in drinking water for 2 weeks (with decreased water consumption), at 443 mg/kg/day in drinking water for 60 days (NTP 1993) (with decreased water consumption), and at ≥367 mg/kg/day for 13 weeks (with decreased water consumption in 1 of these studies) (Carpenter et al. 1956; NTP 1993), and in mice at 12,750 mg/kg/day in drinking water for 2 weeks (Heindel et al. 1990). In rats treated with 2-butoxyethanol in the diet for 91-93 days, body weight gain was decreased by 53% in males at 919 mg/kg/day and by 45% in females at 976 mg/kg/day (Weil and Carpenter 1963). Body weight gain was decreased by as much as 91% at day 1 of the study in males at 188 mg/kg/day but was only 9% lower than controls by the end of the study. The 188-mg/kg/day dose in males was also associated with about an 18% decrease in food intake. No effects on body weight gain were noted at 28 mg/kg/day in males and at 222 mg/kg/day in females. Pregnant rats exhibited decreased gestational weight gain (as well as decreased food and water intake) at ≥150 mg/kg/day during gestation (NTP 1989), as did pregnant mice at ≥ 1,000 mg/kg/day during gestation (Hardin et al. 1987; Schuler et al. 1984; Wier et al. 1987) and at 1,300 mg/kg/day m drinking water (Heindel et al. 1990) in a reproductive study.

In dermal studies of 2-butoxyethanol, decreased body weight gain was observed in pregnant rats at 0.48 mL/day during gestation (Hardin et al. 1984) and in rats at 200 mg/kg for 24 hours (Dow 1959).

For 2-butoxyethanol acetate, no effect on body weight was noted in rats or rabbits exposed by inhalation intermittently to 400 ppm for 1 month or to 100 ppm for 10 months (Truhaut et al. 1979). For 2-butoxyacetic acid, rats given single gavage dose ≥174 mg/kg had decreased weight gain during the first 2 days after treatment at all doses, but the weight gain returned to normal by day 14 (Foster et al. 1987).

Effects on body weight do not appear to be a concern for humans exposed to 2-butoxyethanol, 2-butoxyethanol acetate, or 2-butoxyacetic acid in environmental or hazardous waste site scenarios.

Metabolic Effects. The only information regarding metabolic effects of 2-butoxyethanol comes from case reports of humans who ingested household cleaning agents containing 2-butoxyethanol. The metabolic effects consisted of metabolic acidosis and hypoxemia with lactic acidosis in the known alcohol and trichloroethylene abuser who ingested 650 mg/kg (Bauer et al. I992), metabolic acidosis and hypokalemia in a patient who ingested 467-933 mg/kg (Rambourg-Schepens et al. 1988), and metabolic acidosis in another patient who ingested 391-469 mg/kg (Gijsenbergh et al. 1989). Significant acid-base disturbance was observed in a man following ingestion of 1,006-l ,341 mg/kg 2-butoxyethanol in a concentrated glass cleaner (Gualtieri et al. 1995). A second ingestion of the glass cleaner (1,341 mg/kg 2-butoxyethanol) 12 days later did not result in metabolic disturbances. After the second ingestion, hemodialysis and ethanol treatment were initiated within 8 hours after exposure, but it is unclear how much time elapsed before treatment of first injection. It is to be expected that metabolism of significant quantities of 2-butoxyethanol to 2-butoxyacetic acid would cause metabolic acidosis.

Whether or not humans exposed to 2-butoxyethanol in environmental or hazardous waste site scenarios would have metabolic effects cannot be determined based on the limited available data.

Other Systemic Effects. No studies were located regarding other systemic effects in humans after inhalation, oral, or dermal exposure to 2-butoxyethanol. In animal studies on 2-butoxyethanol, other systemic effects consisted of reduced food consumption at ≥ 100 ppm and reduced water consumption at 200 ppm in pregnant rats exposed to 2-butoxyethanol vapors for 6 hours per day during gestation (Tyl et al. 1984); decreased water consumption in rats at ≥ 152 mg/kg/day and in mice at ≥150 mg/kg/day in drinking water for 2 weeks (Heindel et al. 1990; NTP 1993); dehydration in mice at ≥376 mg/kg/day in drinking water for 2 weeks (NTP 1993);

decreased food and water intake at ≥150 mg/kg/day, and coldness to the touch at 600 mg/kg/day in pregnant rats given gavage doses during gestation (NTP 1989); decreased water consumption in rats at ≥ 444 mg/kg/day in drinking water for 21 days (Exon et al. 1991); decreased food consumption in rats at 885 mg/kg/day by gavage for 6 weeks (Eastman Kodak 1983; Krasavage 1986); decreased water consumption in rats at ≥ 128 mg/kg/day in drinking water for 13 weeks (NTP 1993); decreased food intake in rats fed 2-butoxyethanol in the diet at ≥188 mg/kg/day for males and 976 mg/kg/day for females (Weil and Carpenter 1963); and decreased water consumption in mice at 700 mg/kg/day in drinking water for up to 25 weeks (Heindel et al. 1990).

Although decreased food and water consumption may have been due to unpalatability of 2-butoxyethanol in the dietary and drinking water studies, these effects were also observed in animals after inhalation or gavage treatment. Furthermore, the decreased body weight gain observed in animals in some studies cannot be completely attributed to decreased food or water intake. Nevertheless, decreased water consumption and decreased food intake do not appear to be effects of concern in humans exposed to 2-butoxyethanol under any scenario.

Immunological and Lymphoreticular Effects. No studies were located regarding immunological and lymphoreticular effects in humans after inhalation or oral exposure to 2-butoxyethanol or 2-butoxyethanol acetate. Slight erythema, but no allergic reaction, was noted on skin-patch tests with 2-butoxyethanol acetate in humans (Jacobs et al. 1989). A human sensitivity study in 201 individuals with 10% 2-butoxyethanol showed no overt effect (CMA 1992; Greenspan et al. 1995). The 10% concentration was tested because it is the highest concentration found in cosmetics. Concentrations as high as 50% 2-butoxyethanol can be found in cleaning products which are intended to be diluted before use (OECD 1997).

The information on immunological effects of 2-butoxyethanol in animals consists of the following findings: no effect on the immune system was found in rats at gavage doses of ≤400 mg/kg/day for 2 days following immunization with trinitrophenyl-lipopolysaccharide (TNP-EPS) (Smialowicz et al. 1992); and no IgG change compared with controls in antibody production or delayed-type hypersensitivity response after-challenge with anti-keyhole limpet hemocyanin; also, no adverse effects on natural killer cell function or splenocyte production of cytokines, or thymus and spleen weights were observed. No effect on thymus or spleen weight, and no histopathological lesions in the thymus were observed in rats given ≤ 506 mg/kg/day for 21 days (Exon et al. 199 1). 2-Butoxyethanol has also tested negative for dermal sensitization in guinea pigs using the maximized Magnusson and Kliman test (Zissu 1995).

Several studies in animals describe effects on the spleen, all of which are probably related to the hematological effect of 2-butoxyethanol and the resultant accumulation of lysed red blood cells in the spleen.

In inhalation studies on 2-butoxyethanol in animals, splenic effects consisted of increased spleen weight in pregnant rats at 200 ppm for 6 hours per day during gestation (Tyl et al. 1984). However, histological examination of the spleen, mediastinal lymph nodes, and thymus revealed no lesions in rats exposed intermittently to ≤177 ppm for 13 weeks (Dodd et al. 1983).

In oral studies on 2-butoxyethanol in animals, effects on lymphoreticular organs consisted of increased spleen weight in pregnant rats at ≤100 mg/kg/day during gestation (NTP 1989), in young and adult rats at ≥ 125 mg/kg by gavage (Ghanayem et al. 1987a, 1987b) or 125 mg/kg/day for up to 12 days (Ghanayem et al. 1992), and increased spleen weight accompanied by extramedullary hematopoiesis at ≥ 500 mg/kg/day for 4 days (Grant et al. 1985). In addition, splenic congestion, secondary to hematological effects, at ≥ 222 mg/kg/day and increased spleen weight and enlarged dark spleens at ≥ 443 mg/kg/day were observed in rats treated by gavage for 6 weeks, but no histological lesions were seen in any lymphoreticular organs (Eastman Kodak 1983; Krasavage 1986). Decreased thymus weight occurred in mice at ≥ 370 mg/kg/day in drinking water for 2 weeks (NTP 1993). Bone marrow hyperplasia at ≥281 mg/kg/day, hematopoietic cell proliferation at ≥ 363 mg/kg/day, increased hemosiderin pigmentation in the spleen at ≥ 129 mg/kg/day, and decreased thymus weight at ≥ 367 mg/kg/day were found in rats given 2-butoxyethanol in drinking water for 13 weeks (NTP 1993). None of these effects occurred in mice similarly exposed at ≤ 1,306 mg/kg/day.

In dermal studies on 2-butoxyethanol in animals, effects on Iymphoreticular organs consisted of congestion in the spleen, erythrocytic infiltration, and white atrophic pulp in rabbits at ≥ 72 mg/kg/day for 8 hours (Duprat and Gradiski 1979); engorged spleen in rabbits at ≥ 406 mg/kg (Carpenter et al. 1956); and enlarged spleen in rabbits at 902 mg/kg for 6 hours (Union Carbide 1980a) and dark red spleens at ≥451 mg/kg for a 24-hour exposure (Union Carbide 1980b).

For 2-butoxyethanol acetate, no gross or histopathological lesions were found in the spleen of rats or rabbits exposed by inhalation intermittently to 400 ppm for 1 month or to 100 ppm for 10 months, in rats at unspecified single gavage doses, or in rabbits exposed dermally at ≤10,000 mg/kg for 24 hours (Truhaut et al. 1979).

The limited available data in humans and animals indicate that exposure to 2-butoxyethanol or 2-butoxy-ethanol acetate under any scenario would not result in immunological effects; however, lymphoreticular effects in the spleen and bone marrow, which are probably attributable to the hemolytic effects of 2-butoxyethanol, may occur in humans exposed to high levels of 2-butoxyethanol.

Neurological Effects. Humans exposed experimentally by inhalation to 2-butoxyethanol for 4-8 hours experienced headache at 98 ppm and disturbed taste sensation at 113 and 195 ppm (Carpenter et al. 1956). Case reports of intentional poisoning indicate that coma can result from the ingestion of ≥467 mg/kg 2-butoxyethanol (Bauer et al. 1992; Gijsenbergh et al. 1989; Litovitz et al. 1991; Rambourg-Schepens et al. 1988).

Neurological effects have been observed in animals after inhalation, oral, and dermal exposure to 2-butoxyethanol. In inhalation studies of 2-butoxyethanol in animals, neurological effects consisted of loss of coordination in rats at ≥523 ppm for 4 hours (Dodd et al. 1983), extreme weakness in dogs at 617 ppm for 2 days, weakness and apathy in dogs at 385 ppm for 8 or 28 days (Carpenter et al. 1956), excessive salivation in dogs at 400 ppm for 5 days (Dow 1972, 1986), and poor coordination of the extremities and loss of equilibrium in rabbits at 400 ppm 2-butoxyethanol for 7 hours per day on l-2 days (Dow 1986).

In oral studies of 2-butoxyethanol in animals, neurological effects consisted of sluggishness, prostration, and narcosis in rats that died after gavage doses ≥530 mg/kg (Carpenter et al. 1956) and drowsiness, sluggishness, lethargy, muscle flaccidity, and/or ataxia in rats at 252-9,000 mg/kg/day (Dow 1959, 1981; Eastman Kodak 1983; Krasavage 1986; Olin 1976; Union Carbide 1980b). Ataxia was reported in rats following a single gavage dose of 500 mg/kg (Sivarao and Mehendale 1995). Moderate to severe weakness and prostration was observed directly after dosing in guinea pigs given a single gavage dose of 1,000 mg/kg 2-butoxyethanol, a dose that resulted in the death of one of five males, and one of five females (Shepard 1994b). Slight weakness was observed directly after dosing at 500 mg/kg, a dose at which all treated guinea pigs survived. Lethargy also was observed in pregnant rats treated at 600 mg/kg/day by gavage during gestation (NTP 1989) and in rats at ≥443 mg/kg/day for 6 weeks (Eastman Kodak 1983; Krasavage 1986), and lethargy and failure to right were observed in pregnant mice treated at ≥1,500 mg/kg/day by gavage during gestation (Wier et al. 1987).

In dermal studies of 2-butoxyethanol in animals, neurological effects consisted of prostration and narcosis prior to death in rabbits at ≥72 mg/kg for 8 hours (Duprat and Gradiski 1979); muscle flaccidity at 1,000 and 2,000 mg/kg and anorexia and no spontaneous movement at 2,000 mg/kg prior to death in rabbits treated with

2-butoxyethanol for 24 hours on abraded skin (Olin 1976); ataxia progressing to moderate-to-marked inactivity prior to death in pregnant rats at 1.4 mL/day during gestation (Hardin et al. 1984); and nystagmus and convulsion prior to death in rabbits at 902 mg/kg (Union Carbide 1980a).

For 2-butoxyethanol acetate, no clinical signs of neurotoxicity and no histopathological lesions in the brain were found in rats or rabbits exposed by inhalation intermittently to 400 ppm for 1 month or to 100 ppm for 10 months, in rats at unspecified single gavage doses, or in rabbits exposed dermally at ≤ 10,000 mg/kg for 24 hours (Truhaut et al. 1979).

The data indicate that humans exposed to sufficient quantities of 2-butoxyethanol by any route may experience lethargy, but more serious neurological effects would not be expected to occur in humans exposed to 2-butoxyethanol or 2-butoxyethanol acetate under environmental or hazardous waste site scenarios.

Reproductive Effects. No studies were located regarding reproductive effects in humans after inhalation, oral, or dermal exposure to 2-butoxyethanol or 2-butoxyethanol acetate.

Reproductive effects have been observed in animals after inhalation, oral, and dermal exposure to 2-butoxyethanol. In inhalation studies, reproductive effects consisted of reduced maternal gravid uterine weight, decreased percentages of viable implants and in live fetuses per litter, and an increase in non-viable implants in pregnant rats, and decreased total implants and implant viability in pregnant rabbits at 200 ppm during gestation (Tyl et al. 1984). This concentration also resulted in the death of 4 of 24 exposed rabbits, and maternal hematological effects.

In oral studies on 2-butoxyethanol in animals, reproductive effects consisted of decreased sperm concentration (with no effects on spermatid heads, spermatid counts, or percent mobile sperm) in male rats at ≥ 281 mg/kg/day in drinking water for 13 weeks (NTP 1993); testicular atrophy and decreased body weight gain were observed in rats fed 2-butoxyethanol in the diet at 188 mg/kg/day for about 90 days (Weil and Carpenter 1963); altered estrous cycles (more time spent in diestrus) with no change in total cycle length in female rats at ≥363 mg/kg/day in the drinking water for 13 weeks (NTP 1993); vaginal bleeding at 200-600 mg/kg/day and increased resorptions and implantation loss at ≥ 200 mg/kg/day in rats during gestation (NTP 1989); decreased incidence of viable litters in pregnant mice at 1,180 mg/kg/day by gavage during gestation (Hardin et al. 1987; Schuler et al. 1984); a green or red vaginal discharge at ≥ 1,500 mg/kg/day and increased resorptions at ≥ 1,000 mg/kg/day in mice during gestation (Wier et al. 1987);

and a decrease in the number of litters produced per breeding pair and litter size in mating pairs of mice at ≥ 1,300 mg/kg/day in drinking water for 21 weeks (Heindel et al. 1990). Cross-breeding experiments indicated that these effects were due to effects on the females (Heindel et al. 1990).

In dermal studies on 2-butoxyethanol in animals, the only reproductive effect consisted of a slight increase in testicular weight of male rabbits at 150 mg/kg/day for 6 hours per day for 90 days (CMA 1983). Histological examination of reproductive organs of males and females revealed no lesions.

For 2-butoxyethanol acetate, no histopathological lesions were found in testes or ovaries in rats or rabbits exposed by inhalation intermittently to 400 ppm for 1 month or to 100 ppm for 10 months, in rats exposed orally at unspecified single gavage doses, or in rabbits exposed dermally to ≤10,000 mg/kg for 24 hours (Truhaut et al. 1979).

The data indicate that reproductive effects might be a concern for pregnant women exposed to high levels of 2-butoxyethanol. Based on the findings in male animals, little concern is indicated for reproductive effects in men.

Developmental Effects. No studies were located regarding developmental effects in humans after inhalation, oral, or dermal exposure to 2-butoxyethanol or 2-butoxyethanol acetate. No studies were located regarding developmental effects in animals after exposure to 2-butoxyethanol acetate by any route. However, developmental effects have been observed in animals after inhalation, oral, and dermal exposure to 2-butoxyethanol.

In inhalation studies on 2-butoxyethanol in animals, the developmental effects consisted of retarded skeletal ossification in the offspring of pregnant rats at ≥ 100 ppm and in pregnant rabbits at 200 ppm for 6 hours per day during gestation (Tyl et al. 1984). In oral studies, the developmental effects consisted of decreased fetal body weight of the offspring of pregnant rats at ≥300 mg/kg/day and decreased gravid uterine weight of rat dams at 600 mg/kg/day during gestation (NTP 1989), and some evidence of cleft palate in the-fetuses of pregnant mice given ≥1,000 mg/kg/day during gestation (Wier et al. 1987). In a reproductive toxicity study in male and female mice, decreased pup weight was observed in all mating pairs at ≥700 mg/kg/day in drinking water for 21 weeks (Heindel et al. 1990). A crossover mating trial indicated that the reproductive effects could be attributed primarily to an effect on the female. In dermal studies on 2-butoxyethanol, no developmental effects were observed in the offspring of pregnant rats at 0.48 mL/day during gestation (Hardin et al. 1984).

With the exception of decreased pup weight observed in mice at 700 mg/kg/day (Heindel et al. 1990), developmental effects reported occurred at 2-butoxyethanol doses that resulted in moderate to severe maternal toxicity (Hardin et al. 1987; NTP 1989; Tyl et al. 1984; Wier et al. 1987).

2-Butoxyethanol has been shown to have developmental toxicity in an *in vitro* assay using the fresh water coelenterate *Hydra attenuatu* (Bowden et al. 1995; Johnson et al. 1984), whereas both 2-butoxyethanol (Bowden et al. 1995; Giavini et al. 1993) and 2-butoxyacetic acid (Giavini et al. 1993; Rawlings et al. 1985) have been shown to be embryotoxic in rat embryo culture. A significant increase in malformations (anomalous encephalon, hypoplasia of the branchial arches, unrotated) was observed only at the highest concentration of 2-butoxyethanol (l2.5 mM) used in this study, but not at 6.25 mM (Giavini et al. 1993). Malformations noted following exposure to 2-butoxyacetic acid at concentrations of 0.8 mM and greater were anomalous encephalon and irregular disposition of the somites (Giavini et al. 1993); however, another study found abnormalities at 5 mM 2-butoxyacetic acid (no statistics done), but not at 2 mM (Rawlings et al. 1985). Based on the *in vitro* data, Bowden et al. (1995) suggested that 2-butoxyethanol "presents a teratogenic potential that *in vivo* is masked by maternal toxicity."

The available data indicate that low birth weights and delayed ossification might be concerns for the offspring of pregnant women exposed to high levels of 2-butoxyethanol.

Genotoxic Effects. No increases in micronuclei or sister chromatid exchanges were observed in varnish production workers exposed to both 2-ethoxyethanol and 2-butoxyethanol (Sohnlein et al. 1993). Postshift biological monitoring for the acetic acids in urine indicated that urinary levels of 2-ethoxyacetic acid (53.8 mg/L) were higher than urinary levels of 2-butoxyacetic acid (16.4 mg/L). One report of a 2-butoxyethanol test in a rat primary hepatocyte test was located (McGregor 1984). [^{14}C]2-Butoxyethanol was tested for 2 hours at concentrations up to 0.1% in the culture medium. Significant increases in nuclear and DNA-associated radioactivity at two low-dose levels were observed, with the maximum response 1.3 times the control levels. 2-Butoxyethanol was reported not to induce sister chromatid exchange in Chinese hamster ovary cells exposed to concentrations up to 0.25% for 2 hours in the presence of metabolizing enzymes (S9) and for 5 hours in the absence of S9 (Tyler 1982). 2-Butoxyethanol and 2-butoxyacetic acid did not result in mutagenic effects in Chinese hamster ovary cells exposed for 5 hours (Chiewchanwit and Au 1995). The maximum non-toxic concentrations studied were 1% for 2-butoxyethanol and 6.5% for 2-butoxyacetic acid.

The mutagenic effect of 2-butoxyethanol was investigated using bacteriophage T4D with *Escherichia coli* B, CR63, and K12 (Kvelland 1988). No mutagenic activity was observed, but 2-butoxyethanol had a severe toxic effect on phage yield. 2-Butoxyethanol was negative for mutation in *Salmonella typhimurium* strains TA98, TAlOO, and TA102 both with and without rat S-9 activation (Hoflack et al. 1995). *In S. typhimurium* strain TA97a, 2-butoxyethanol tested positive for mutation both with and without S-9 activation. The mutation assay of 2-butoxyethanol in strain TA97a was repeated by Gollapudi et al. (1996). In this study, 2-butoxyethanol was negative for mutations both with and without rat S-9 metabolic activation, and it was negative in *E. coli WP2uvr* A strain. Gollapudi et al. (1996) suggested that Hoflack et al. (1995) found positive results because they used 2-butoxyethanol contaminated with higher levels of peroxides, which test positive in strain TA97a. The 2-butoxyethanol used by Gollapudi et al. (1996) contained 23 ppm of peroxides which was measured just before the tests. The 2-butoxyethanol used by Hoflack et al. (1995) contained less than 50 ppm peroxides according to the manufacturer, but the actual concentrations of peroxides may have been higher since measurements were not made immediately prior to testing. Gollapudi et al. (1996) concluded that peroxides and/or experimental variables probably account for the difference in results observed in their study and in the study by Hoflack et al. (1995). *In vitro* genotoxic effects are shown in Table 2-13.

Cancer. No studies were located regarding carcinogenic effects in humans or animals after exposure to 2-butoxyethanol, 2-butoxyethanol acetate, or 2-butoxyacetic acid by any route. However, structure-activity studies with 2-butoxyethanol and eight other glycol ethers were conducted with a cellular leukemia transplant model in male Fischer 344 rats to determine the chemotherapeutic potential of these glycol ethers (Dieter et al. 1990). 2-Butoxyethanol was administered *ad libitum* in the drinking water to male rats at doses of 0, 3, or 6 mg/mL. Treatment was begun after the rats were injected with the leukemic transplant. Untreated transplant control rats and untreated non-transplant controls were also included. Treatment continued for 55-65 days, at which time the animals were killed and blood was analyzed. 2-Butoxyethanol had no effect on the progression of the leukemic transplant.

2-Butoxyethanol and 2-butoxyethanol acetate have not been classified for carcinogenic effects by the Department of Health and Human Services (DHHS), the National Toxicology Program (NTP), the International Agency for Research on Cancer (IARC), or EPA.

Table 2-13. Genotoxicity of 2-Butoxyethanol *In Vitro*

Species (test system)	End point	Result		Reference
		With activation	Without activation	
Prokaryotic organisms				
Escherichia coli (T4D phage)	Mutagenesis	–	–	Kvelland 1988
CR63 (T4D phage)	Mutagenesis	–	–	Kvelland 1988
K12 (T4D phage)	Mutagenesis	–	–	Kvelland 1988
Salmonella typhimurium TA98, TA100, TA102	Mutagenesis	–	–	Hoflack et al. 1995
S. typhimurium TA97a	Mutagenesis	+	+	Hoflack et al. 1995
S. typhimurium TA97a, TA100	Mutagenesis	–	–	Gollapudi et al. 1996
E. coli WP2 *ovr*	Mutagenesis	–	–	Gollapudi et al. 1996
Mammalian cells				
Rat (primary hepatocyte)	DNA-associated radioactivity	No data	+	McGregor 1984
Chinese hamster (ovary cells)	Sister chromatid exchange	–	–	Tyler 1982
Chinese hamster (ovary cells)	Mutagenesis	No data	–	

– = negative results; + = positive results

2.6 BIOMARKERS OF EXPOSURE AND EFFECT

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

Due to a nascent understanding of the use and interpretation of biomarkers, implementation of biomarkers as tools of exposure in the general population is very limited. A biomarker of exposure is a xenobiotic substance or its metabolite(s), or the product of an interaction between a xenobiotic agent and some target molecule(s) or cell(s) that is measured within a compartment of an organism (NAWNRC 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 2-butoxyethanol and 2-butoxyethanol acetate are discussed in Section 2.6.1.

Biomarkers of effect are defined as any measurable biochemical, physiologic, or other alteration within an organism that, depending on magnitude, can be recognized as an established or potential health impairment or disease (NASKNRC 1989). This definition encompasses biochemical or cellular signals of tissue dysfunction (e.g., increased liver enzyme activity or pathologic changes in female genital epithelial cells), as well as physiologic signs of dysfunction such as increased blood pressure or decreased lung capacity. Note that these markers are not often substance specific. They also may not be directly adverse, but can indicate potential health impairment (e.g., DNA adducts). Biomarkers of effects caused by 2-butoxyethanol and 2-butoxyethanol acetate are discussed in Section 2.6.2.

A biomarker of susceptibility is an indicator of an inherent or acquired limitation of an organism’s ability to respond to the challenge of exposure to a specific xenobiotic substance. It can be an intrinsic genetic or other characteristic or a preexisting disease that results in an increase in absorbed dose, a decrease in the biologically

effective dose, or a target tissue response. If biomarkers of susceptibility exist, they are discussed -in Section 2.8, Populations That Are Unusually Susceptible.

2.6.1 Biomarkers Used to Identify or Quantify Exposure to 2-butoxyethanol

The presence of 2-butoxyacetic acid in the urine is the accepted biomarker of exposure for both 2-butoxyethanol and 2-butoxyethanol acetate (Johanson et al. 1989) and has been used in surveys of occupational exposure, although 2-butoxyethanol has also been detected in blood and urine (Angerer et al. 1990; Haufroid et al. 1997; Johanson 1988; Johanson and Boman 1991; Johanson and Femstrom 1986; Johanson and Johnsson 1991; Johanson et al. 1986a, 19881989; Jiinsson and Steen 1978; Rettenmeier et al. 1993; Sakai et al. 1994; Vincent et al. 1993). These have also been used as biomarkers of exposure in animal experiments (Ghanayem et al. 1987a, 1987b, 1987c, 1992; Medinsky et al. 1990; Romer et al. 1985; Sabourin et al. 1992a, 1992b, 1993). More recently, analytical methods have been able to detect 2-butoxyacetic acid in human blood (Johanson and Johnsson 199 1) Urinalysis for 2-butoxyacetic acid should include detection of glutamine conjugates since the amount of conjugate detected in urine can be equal to or greater than the amount of free 2-butoxyacetic acid (Rettenmeier et al. 1993; Sakai et al. 1994)

For practical purposes, estimates of exposure can best be made from 2-butoxyacetic acid levels in the urine. Based on the work of Johanson (1988), 2-butoxyacetic acid is the appropriate biomarker of exposure for the following reasons: it is not normally in human urine, it can be reliably assayed with current methods, levels in the urine are not affected by hemolytic activity, the elimination half-life (4-7 hours) reflects exposure over the workday, and urine collection is non-invasive. Possible sources of variation and error inherent in the biological monitoring of 2-butoxyacetic acid in the urine include variable uptake during inhalation exposure caused by changes in workload, or during dermal exposure caused by changes in temperature and humidity, and changes in excretion caused by alcohol consumption or other personal habits (e.g., smoking) (Johanson and Boman 199 1; Johanson et al. 198 8). Inter-individual variation in the rates of absorption, distribution, metabolism, and excretion may also influence the evaluation of 2-butoxyacetic acid in the urine of exposed individuals. For example, a genetic polymorphism in the CYP 2El gene may possibly result in differences in the metabolism of 2-butoxyethanol; this speculation is based on the results of one study in which urinary levels of free butoxyacetic acid (conjugates were not measured) were measured in 30 c1/c1 homozygotes and one c1/c2 heterozygote (Haufroid et al. 1997). Persons heterozygous for the c2 allele may produce less 2-butoxyacetic acid, and more ethylene glycol than persons homozygous for the c1 allele; however, ethylene glycol and its metabolites were not measured in the urine in this study. This hypothesis merits further

investigation. Studies indicate that the concentration of 2-butoxyacetic acid in the urine may not be linearly correlated with the absorbed dose; however, for individuals, it is related to absorbed dose and thus is a useful measure (Johanson 1988; Johanson et al. 1986a). Since these studies examined only free 2-butoxyacetic acid, it is unknown how much conjugated 2-butoxyacetic acid was in the urine. Because some (15-27% worst case) dermal absorption may occur from vapor exposure, 2-butoxyacetic acid in the urine resulting from ambient air concentrations of 2-butoxyethanol may represent both pulmonary and dermal absorption (Corley et al. 1997; Johanson and Boman 1991). Individual monitoring throughout the work day (urine samples prior to and after the workshift) may be a good choice for widescale biological monitoring in the occupational setting.

2-Butoxyethanol has been shown to increase human plasma osmolality *in vitro* (Browning and Curry 1992). The osmolal gap is frequently used in the evaluation of a patient who has accidentally or intentionally ingested a glycol ether or related compound (Gijsenbergh et al. 1989). The presence of an elevated osmolal gap indicates the presence of significant blood levels of the toxic compound. This is not an indicator that is specific to 2-butoxyethanol or 2-butoxyethanol acetate and should be used in conjunction with other biomarker assays.

2.6.2 Biomarkers Used to Characterize Effects Caused by 2-Butoxyethanol

Hematotoxic effects are the characteristic biomarker of effect for 2-butoxyethanol exposure of animals (Bartnik et al. 1987; Ghanayem et al. 1987a, 1987b). In animals 2-butoxyethanol-induced hemolytic anemia is accompanied by a dose- and time-dependent decrease in the number of circulating red blood cells and decreases in hemoglobin concentration and hematocrit (Ghanayem et al. 1987a, 1987b). Hemolysis has been shown to be preceded by erythrocyte swelling (Ghanayem et al. 1989). Thus, hematological evaluations could be useful in monitoring effects of 2-butoxyethanol exposure. Work by Ghanayem et al. (1990b) indicates that using an impedance-based hematology analyzer allows for the detection of the erythrocytic swelling early after exposure that is undetectable by laser-based analyzer. Both analyzers were able to detect the subsequent decrease in red blood cell concentration signaling hemolytic effects (Ghanayem et al. 1990b). Thus, routine hematology using an appropriate hematology analyzer could be useful in detecting early hemolytic effects after 2-butoxyethanol exposure.

2-Butoxyethanol has been shown to increase human plasma osmolality *in vitro* (Browning and Curry 1992). The osmolal gap is frequently used in the evaluation of a patient who has accidentally or intentionally ingested a glycol ether or related compound (Gijsenbergh et al. 1989). The presence of an elevated osmolal gap is

associated with metabolic acidosis, which has been observed in humans after ingestion of 2-butoxyethanol (Bauer et al. 1992; Gijsenbergh et al. 1989; Rambourg-Schepens et al. 1988). This is not an indicator that is specific to 2-butoxyethanol or 2-butoxyethanol acetate and should be used in conjunction with other biomarker assays.

The use of urinary D-glucaric acid as a biomarker of effect following exposure to 2-butoxyethanol has been examined in 12 or 13 foundry workers exposed to the compound in paints and 18 unexposed controls (Collinot et al. 1996). Increased urinary D-glucaric acid is a nonspecific indicator of an increase in the activity of the glucuronic acid enzyme pathway. Urinary levels of D-glucaric acid were measured both in the summer and winter, and exposure concentrations were measured only in winter when they were expected to be higher because of reduced ventilation. Urinary concentrations of D-glucaric acid in nmol/hour were 2,095±991 in controls, 3,888±2,284 in the exposed workers during summer, and 5,792±2,387 in the exposed workers during winter. The values were significantly different from controls at levels of $p<0.05$ during summer and $p<0.01$ during winter. When the groups were divided into smokers and nonsmokers, the smokers showed a higher level of D-glucaric acid than the nonsmokers The exposure concentrations never exceeded 7.5 ppm, and the study authors indicated that other substances were undetectable (detection limits not stated). Although measurement of D-glucaric acid is not an effect specific to 2-butoxyethanol exposure, the study authors concluded that it was more sensitive than a standard blood count.

2.7 INTERACTIONS WITH OTHER CHEMICALS

Data describing the interaction of 2-butoxyethanol or 2-butoxyethanol acetate with other chemicals are scarce. Simulations of human pharmacokinetics of co-exposure to ethanol (0.1% in the blood) during an 8-hour exposure to 20 ppm 2-butoxyethanol with no exercise predict that the arterial levels of 2-butoxyethanol will be elevated as a result of a decrease in elimination rate (Johanson 1986, 1991a; Johanson and Naslund 1988). Because the increase in 2-butoxyethanol is due to decreased elimination and not increased uptake, the rise and fall of blood concentrations of 2-butoxyethanol are slower in the ethanol co-administration model than in the increased workload model. A study in rats indicated that co-administration of ethanol and 2-butoxyethanol resulted in a higher blood level and a prolonged blood elimination time of 2-butoxyethanol than administration of 2-butoxyethanol alone (Romer et al. 1985). The prolonged retention of 2-butoxyethanol was considered to be due to inhibition of metabolism of 2-butoxyethanol to 2-butoxyacetaldehyde via competition with ethanol for alcohol dehydrogenase, the enzyme common to the metabolism of both ethanol and 2-butoxyethanol. Based on these results in rats, the study authors concluded that the elimination of 2-butoxyethanol could be

inhibited in humans after consumption of alcoholic beverages and simultaneous exposure to 2-butoxyethanol. As discussed in Section 2.9.3, administration of ethanol after 2-butoxyethanol exposure can be used to control the production of toxic metabolites and to promote conjugation of the parent compound to nontoxic products of elimination such as the glucuronide and sulfate conjugates of 2-butoxyethanol.

Hemolysis is a critical effect of exposure to 2-butoxyetbanol, but it is unknown whether 2-butoxyethanol interacts with other hemolytic agents, such as naphthalene, the toxicity of which is reviewed in the ATSDR *Toxicological Profile for Naphthalene* (ATSDR 1995b).

2.8 POPULATIONS THAT ARE UNUSUALLY SUSCEPTIBLE

A susceptible population will exhibit a different or enhanced response to 2-butoxyethanol than will most persons exposed to the same level of 2-butoxyethanol in the environment. Reasons may include genetic makeup, age, health and nutritional status, and exposure to other toxic substances (e.g., alcohol and cigarette smoke). These parameters may result in reduced detoxification or excretion of 2-butoxyethanol, or compromised function of target organs affected by 2-butoxyethanol. Populations who are at greater risk due to their unusually high exposure to 2-butoxyethanol are discussed in Section 5.6, Populations With Potentially High Exposure.

Since the hemolytic effect of 2-butoxyethanol is caused by the action of the toxic metabolite 2-butoxyacetic acid, presumably on the red cell membrane, populations that are unusually susceptible to the toxic effects of 2-butoxyethanol would be those that have increased metabolism to 2-butoxyacetic acid and decreased excretion of 2-butoxyacetic acid. Persons whose red blood cell walls are less resistant to the effects of 2-butoxyacetic acid would also be more sensitive to 2-butoxyethanol. However, erythrocytes from normal, aged, sickle-cell anemia, and hereditary spherocytosis patients were resistant to the hemolytic effects of 2-butoxyacetic acid *in vitro* (Udden 1996; Udden and Patton 1994). Since erythrocytic swelling has been shown to precede hemolysis in rats after 2-butoxyethanol exposure, Udden and Patton (1994) investigated the possibility that increased deformability of erythrocytes in humans could predispose the individual to 2-butoxy-ethanol toxicity. They exposed erythrocytes from normal, aged, sickle-cell anemia, and hereditary spherocy-tosis patients to 2-butoxyacetic acid, the hemolytic metabolite of 2-butoxyethanol, *in vitro* under conditions that cause hemolysis of rat erythrocytes. No increase in hemolysis, changes in mean cellular volume or morphology, or deformability were found in any of the human erythrocytes. However, further definition of the metabolic or structural differences that render human erythrocytes more resistant to the effects of 2-butoxy-

ethanol compared to rat erythrocytes may also point to characteristics in the human population that may indicate increased susceptibility. It is unknown whether people with a genetic predisposition to hemolytic anemia from other causes (such as glucose-6-phosphate dehydrogenase mutations) will be more susceptible to 2-butoxyethanol-induced hemolysis.

In rats, Ghanayem et al. (1987a, 1990a) have shown that susceptibility to the hematotoxic effects of 2-butoxyethanol increases with increasing age. Investigation of the respiratory excretion and urinary metabolites of young (4-5 weeks old) and adult (9-13 weeks old) rats indicated that adult rats excreted a significantly lower proportion of the administered dose as respiratory CO_2 and eliminated less in the urine as the 2-butoxyethanol-glucuronide; more was eliminated as 2-butoxyacetic acid. No studies of this kind have been done in humans. However, it is possible that people of advanced age or with altered hepatic conjugating enzymes, reduced respiratory capacity, or compromised renal status could be at greater risk for toxic reactions to 2-butoxyethanol exposure.

Some incomplete and very tentative results on the influence of a CYP2El Polymorphism on the metabolism of 2-butoxyethanol are discussed in Section 2.3.3 Metabolism.

2.9 METHODS FOR REDUCING TOXIC EFFECTS

This section will describe clinical practice and research concerning methods for reducing toxic effects of exposure to 2-butoxyetbanol. 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 2-butoxyethanol. When specific exposures have occurred, poison control centers and medical toxicologists should be consulted for medical advice. The following text provides specific information about treatment following exposures to 2-butoxyethanol: Ellenhom and Barceloux 1988.

2.9.1 Reducing Peak Absorption Following Exposure

Human exposure to 2-butoxyethanol or 2-butoxyethanol acetate can occur by inhalation, oral, or dermal contact. General recommendations for reducing absorption of 2-butoxyethanol or 2-butoxyethanol acetate following acute high-level inhalation exposure have included moving the patient to fresh air and monitoring for respiratory distress (Ellenhorn and Barceloux 1988); washing exposed skin after high level air exposure is probably a good idea since there can be some dermal absorption from 2-butoxyethanol vapors as well (Corley

et al. 1997). In the case of eye exposure, irrigation with copious amounts of water or saline have been recommended (Ellenhorn and Barceloux 1988). The removal of contaminated clothing and a thorough washing of exposed areas with soap and water have also been recommended (Ellenhorn and Barceloux 1988). Induction of emesis may be indicated following recent substantial ingestion of 2-butoxyethanol or 2-butoxyethanol acetate unless the patient is or could rapidly become comatose or convulsive (Ellenhorn and Barceloux 1988). Furthermore, the use of emetics may result in aspiration pneumonitis. Gastric lavage has been recommended, but charcoal and cathartics may be ineffective (Ellenhorn and Barceloux 1988). Gastric lavage and induced emesis using syrup of ipecac have been used in the treatment of accidental poisoning of children (Dean and Krenzelok 1992).

2.9.2 Reducing Body Burden

As discussed in Section 2.3 on Toxicokinetics, following absorption into the blood, 2-butoxyethanol is rapidly distributed throughout the body. Since 2-butoxyethanol is slightly hydrophilic, it is not preferentially distributed to lipid-rich tissues. 2-Butoxyethanol toxicity results from the formation of 2-butoxyacetic acid in the liver, through alcohoValdehyde dehydrogenases. Dealkylation to ethylene glycol may also account for some toxicity. Detoxification pathways generally involve the formation of the sulfate and glucuronide conjugates of the parent compound and conjugation and excretion of the active metabolite 2-butoxyacetic acid. Exhalation is a minor route for excretion of CO_2 from 2-butoxyacetic acid and ethylene glycol, but metabolites are primarily excreted in the urine. Studies in humans and animals indicate that both exhalation and urinary excretion occur in several phases, with half-lives of minutes to hours. Hence, 2-butoxyethanol and its metabolites have relatively short half-lives in the body, and while some of these metabolites are toxic, substantial body burdens are not expected.

It is possible that methods could be developed to enhance detoxification and elimination pathways for the parent compound, in preference to oxidation pathways leading to 2-butoxyacetic acid, to aid in ridding the body of what has been absorbed prior to the production of toxic metabolites.

2.9.3 Interfering with the Mechanism of Action for Toxic Effects

The toxicity of 2-butoxyethanol has been shown to be dependent on the production of 2-butoxyacetic acid via alcohoYaldehyde dehydrogenases through the metabolic intermediate butoxyacetaldehyde. Methods successful in interfering with the mechanism of action for toxic effects are those that interfere with the metabolic

processes responsible for producing 2-butoxyacetic acid. For instance, treatment of rats with pyrazole, an alcohol dehydrogenase inhibitor, provided protection against the hematotoxic effect of 2-butoxyethanol (Ghanayem et al. 1987b). Inhibition of the metabolism of 2-butoxyethanol to butoxyacetaldehyde increased conjugation of the parent compound and urinary excretion of the sulfate or glucuronide detoxification products. Similar results were observed with cyanamide, an aldehyde dehydrogenase inhibitor. Based on observations that 2-butoxyethanol-induced hemolytic anemia was age-dependent, with older rats being more susceptible than younger rats, Ghanayem et al. (1987a) speculated that increased susceptibility of older rats may have been related, in part, to compromised elimination of 2-butoxyacetic acid. Inhibition of renal clearance by the pretreatment of rats with probenecid, an inhibitor of organic acid transport in the proximal renal tubules, significantly increased the half-life of 2-butoxyacetic acid, indicating the importance of renal transport in 2-butoxyacetic acid detoxification (Ghanayem et al. 1990a). Thus, increasing renal transport in exposed individuals might reduce the residence time of the toxic metabolite in the body, consequently reducing the opportunity for toxic reactions.

Ethanol has been shown to block the metabolism of 2-butoxyethanol to 2-butoxyacetic acid through competitive inhibition of alcohol dehydrogenase (Romer et al. 1985). Administration of ethanol after 2-butoxyethanol exposure can be used to control the production of toxic metabolites and to promote conjugation of the parent compound to nontoxic products of elimination such as the glucuronide and sulfate conjugates. This approach has been recommended for clinical therapy for 2-butoxyethanol poisoning (Buckley et al. 1993).

2.10 ADEQUACY OF THE DATABASE

Section 104(I)(5) of CERCLA, as amended, directs the Administrator of ATSDR (in consultation with the Administrator of EPA and agencies and programs of the Public Health Service) to assess whether adequate information on the health effects of 2-butoxyethanol and 2-butoxyethanol acetate is available. Where adequate information is not available, ATSDR, in conjunction with the National Toxicology Program (NTP), is required to assure the initiation of a program of research designed to determine the health effects (and techniques for developing methods to determine such health effects) of 2-butoxyethanol and 2-butoxyethanol acetate.

The following categories of possible data needs have been identified by a joint team of scientists from ATSDR, NTP, and EPA. They are defined as substance-specific informational needs that if met would reduce

the uncertainties of human health assessment. This definition should not be interpreted to mean that all data needs discussed in this section must be filled. In the future, the identified data needs will be evaluated and prioritized, and a substance-specific research agenda will be proposed.

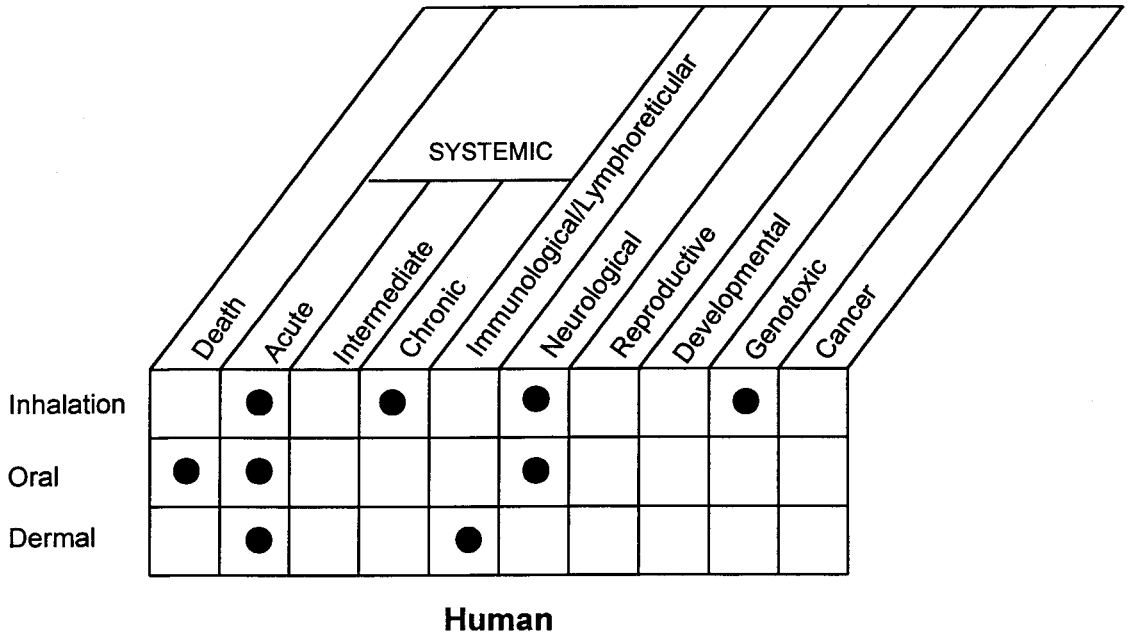
2.10.1 Existing Information on Health Effects of 2-Butoxyethanol

The existing data on health effects of inhalation, oral, and dermal exposure of humans and animals to 2-butoxyethanol and 2-butox yethanol acetate are summarized in Figures 2- 12 and 2- 13, respectively. The purpose of these figures is to illustrate the existing information concerning the health effects of 2-butoxyethanol and 2-butoxyethanol acetate. Each dot in the figure indicates that one or more studies provide information associated with that particular effect. The dot does not necessarily imply anything about the quality of the study or studies, nor should missing information in this figure be interpreted as a "data need." A data need, as defined in ATSDR's *Decision Guide for Identifying Substance-Specific Data Needs Related to Toxicological Profiles* (ATSDR 1989), is substance-specific information necessary to conduct comprehensive public health assessments. Generally, ATSDR defines a data gap more broadly as any substance-specific information missing from the scientific literature.

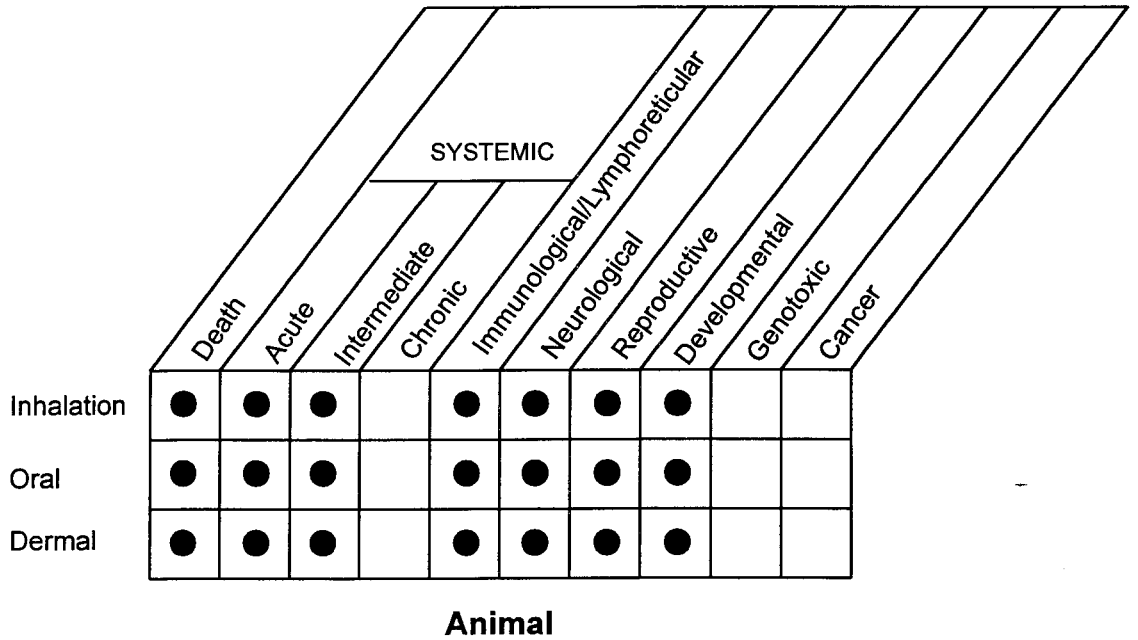
Virtually all of the information regarding health effects in humans comes from experimental studies of volunteers exposed by inhalation or dermally to 2-butoxyethanol, or case reports of accidental or intentional poisoning in humans who ingested household products containing 2-butoxyethanol. Exposures to 2-butoxyethanol and 2-butoxyethanol acetate occur in minting and silk screening shops, at furniture manufacturers, in auto body shops, or anywhere that large-scale painting, inking, furniture stripping, or metal stripping or cleaning occurs. In addition, since 2-butoxyethanol and 2-butoxyethanol acetate are contained in many all-purpose cleaning agents, exposure occurs through window, car and floor washing. Consumer products containing 2-butoxyethanol consist primarily of all-purpose cleaners, but some paints, inks, furniture staining and stripping products, and cosmetics also contain these chemicals. Inhalation and dermal absorption are the main routes of exposure, although accidental or intentional poisoning also occurs.

As seen in Figure 2-l 2 for 2-butoxyethanol, inhalation and oral information for humans is available describing acute-duration systemic and neurological effects. A case report describing the death of a woman following ingestion of a cleaner containing 2-butoxyethanol is also available. A study report of genotoxic effects in humans following occupational exposure to 2-butoxyethanol and the structurally similar 2-ethoxyethanol is

FIGURE 2-12. Existing Information on Health Effects of 2-Butoxyethanol



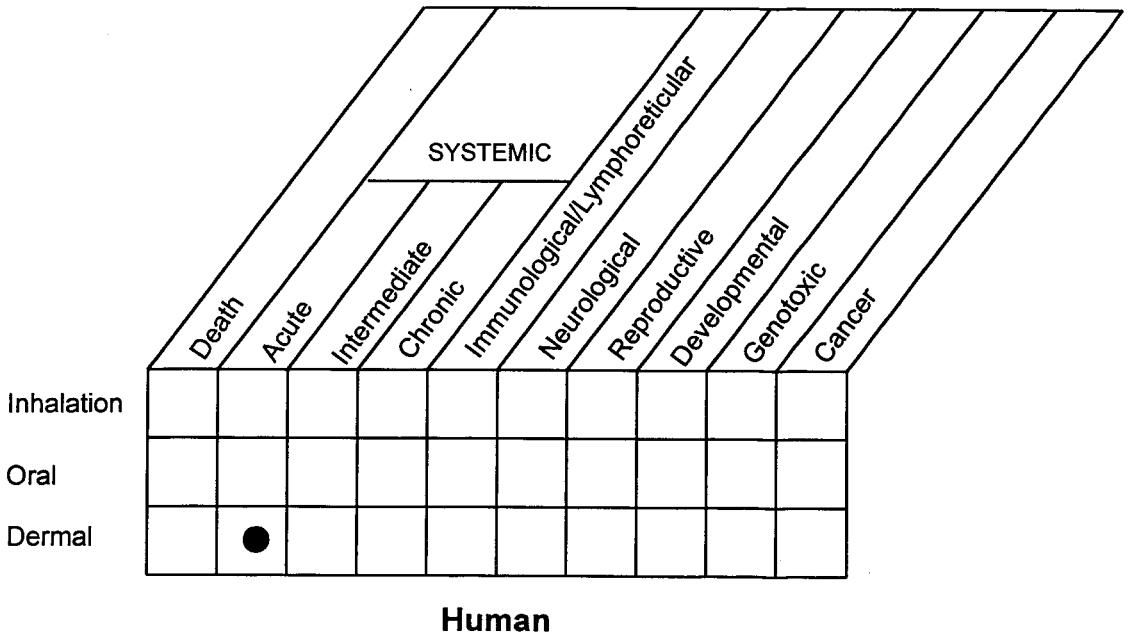
Human



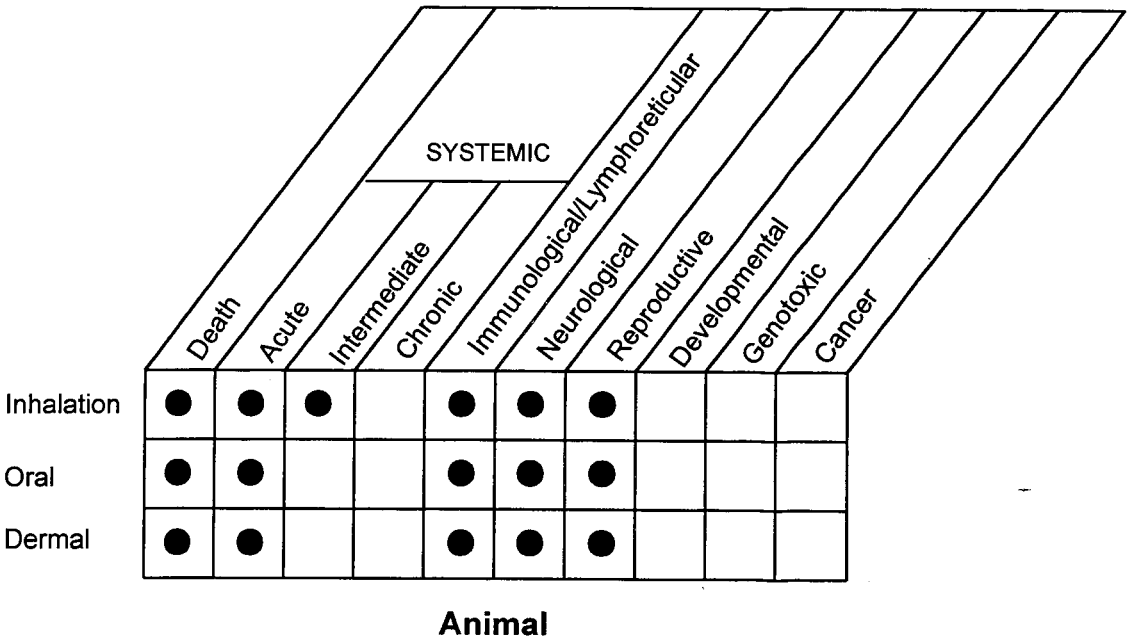
Animal

● Existing Studies

FIGURE 2-13. Existing Information on Health Effects of 2-Butoxyethanol Acetate



Human



Animal

● Existing Studies

available. Information describing the effects of human dermal exposure is limited, consisting of acuteduration systemic and immunological effects. Inhalation, oral, and dermal studies in animals have provided information on death, acute- and intermediate-duration systemic effects, and immunological and lymphoreticular, neurological, reproductive, and developmental effects.

For 2-butoxyethanol acetate, data describing health effects in humans are limited to a dermal irritancy study (Figure 2- 13). Animal studies with 2-butoxyethanol acetate are extremely limited but provide some information on death; acute-duration systemic effects; and immunological and lymphoreticular, neurological, reproductive, and developmental effects after inhalation and oral exposure. In addition, information exists on intermediate-duration systemic effects after inhalation exposure. Dermal studies in animals provide information on death; acute-duration systemic effects; and immunological and lymphoreticular, neurologic, and reproductive effects. It should be noted that the information for 2-butoxyethanol acetate is contained in only two studies.

2.10.2 Identification of Data Needs

Acute-Duration Exposure. There are reports on the health effects resulting from acute exposure in a limited number of studies of humans, and more from animals exposed to 2-butoxyethanol via the inhalation, oral, and dermal routes. In humans exposed by inhalation to 2-butoxyethanol, nasal, throat, and eye irritation, and possibly emesis have been identified as effects (Carpenter et al. 1956). In humans who ingested 2-butoxy-ethanol, respiratory, cardiovascular, hematological, hepatic, renal, ocular, and metabolic effects have been identified (Bauer et al. 1992; Gijsenbergh et al. 1989; Gualtieri et al. 1995; Litovitz et al. 1991; Rambourg-Schepens et al. 1988). Death also occurred in one of these cases (Litovitz et al. 1991). Very little information exists for health effects resulting from dermal exposure of humans, and the data indicate minor local dermal effects and no dermal sensitivity (CMA 1992; Greenspan et al. 1990; Johanson et al. 1988). The primary target organs for acute exposure are the hematological, hepatic, renal, and lymphoreticular (spleen) systems.

In acute-duration inhalation studies in animals, systemic effects have been observed as follow; respiratory effects (Carpenter et al. 1956; Dodd et al. 1983; Dow 1986; Kane et al. 1980; Tyl et al. 1984), gastrointestinal effects (Carpenter et al. 1956; Dow 1986), hematological effects (Carpenter et al. 1956; Dodd et al. 1983; Sabourin et al. 1992a; Tyl et al. 1984; Werner et al 1943a), hepatic effects (Carpenter et al. 1956; Dodd et al. 1983; Dow 1986), renal effects (Carpenter et al. 1956; Dodd et al. 1983; Doe 1984; Dow 1986; Nelson et al. 1984; Tyl et al. 1984; Werner et al. 1943a), ocular effects (Dow 1986; Tyl et al. 1984), and body weight

effects (Carpenter et al. 1956; Dodd et al. 1983; Dow 1972; Tyl et al. 1984). The acute inhalation data were sufficient to derive an acute inhalation MRL of 6 ppm based on a NOAEL of 50 ppm for hematological effects in rats in the study by Tyl et al. (1984).

In acute-duration oral studies in animals, systemic effects have been observed as follows: respiratory effects (Carpenter et al. 1956; Dow 1981; NTP 1989; Union Carbide 1980b; Wier et al. 1987), gastrointestinal effects (Olin 1976; Shepard 1994b; Union Carbide 1980b), hematological effects (Carpenter et al. 1956; Corley et al. 1994; Ghanayem and Sullivan 1993; Ghanayem et al. 1987a, 1987b, 1990b, 1992; Grant et al. 1985; NTP 1989; Sivarao and Mehendale 1995), muscular effects secondary to neurotoxicity (Olin 1976), hepatic effects (Carpenter et al. 1956; Ghanayem et al. 1987b, 1992; Grant et al. 1985; NTP 1989; Olin 1976; Union Carbide 1980b), renal effects (Carpenter et al. 1956; Dow 1959; Ghanayem et al. 1987a, 1987b Grant et al. 1985; NTP 1989; Olin 1976; Sivarao and Mehendale 1995; Union Carbide 1980b), possible dermal effects (Dow 1981; NTP 1989; Olin 1976), ocular effects (Dow 1981; NTP 1989), and body weight effects (Grant et al. 1985; Hat-din et al. 1987; Heindel et al. 1990; NTP 1989, 1993; Union Carbide 1980b; Schuler et al. 1984; Wier et al. 1987). The acute oral data were sufficient to derive an acute oral MRL of 0.4 mg/kg/day based on a LOAEL of 32 mg/kg for hematological effects in rats in the study by Ghanayem et al. (1987b).

In acute-duration dermal studies in animals, systemic effects have been observed as follows: respiratory effects (Duprat and Gradiski 1979; Union Carbide 1980b), gastrointestinal effects (Union Carbide 1980b), hematological effects (Bartnik et al. 1987; Carpenter et al. 1956; Duprat and Gradiski 1979; Hardin et al. 1984; Union Carbide 1980a), muscular effects secondary to neurotoxicity (Olin 1976), hepatic and renal effects (Carpenter et al. 1956; Duprat and Gradiski 1979; Olin 1976; Union Carbide 1980a, 1980b), dermal effects (Dow 1959; Duprat and Gradiski 1979; Hardin et al. 1984; Rohm and Haas 1983; Union Carbide 1980a, 1980b; Zissu 1995), ocular effects (Dow 1959; Kennah et al. 1989a; Olin 1976; Rohm and Haas 1983; Union Carbide 1980a, 1980b), and body weight effects (Dow 1959; Hardin et al. 1984; Union Carbide 1980a).

Information is also available for levels that cause death in a number of animals species following inhalation exposure (Carpenter et al. 1956; Dodd et al. 1983; Dow 1986; Nelson et al. 1984; Sabourin et al. 1992a; Tyl et al. 1984; Werner et al. 1943a), oral exposure (Carpenter et al. 1956; Dow 1959, 1981; Eastman Kodak 1983,1988; Hardin et al. 1987; Heindel et al. 1990; Kennedy and Graepel 1991; Krasavage 1986; NTP 1989; Olin 1976; Sivarao and Mehendale 1995; Smialowicz et al. 1992; Union Carbide 1980b; Schuler et al. 1984;

Shepard 1994b; Smyth et al. 194 l), and dermal exposure (Carpenter and Condra 196 1; Carpenter et al. 1956; Dow 1959; Duprat and Gradiski 1979; Eastman Kodak 1988; Olin 1976; Union Carbide 1980a, 1980b) to 2-butoxyethanol. Studies have shown that rats are particularly sensitive to the hematotoxic effects of 2-butoxyethanol, compared to humans (Bartnik et al. 1987; Ghanayem and Sullivan 1993; Udden 1994). However, since numerous data are available regarding systemic effects in animals, more studies on species-specific systemic effects, particularly hematotoxicity, after acute-duration exposure by inhalation, oral, and dermal routes in animals would not be particularly useful. Doses or concentrations that would result in hematological effects in humans are not known, and it would be unethical to find out by direct experiments on humans; however, there may be a place for more in *in vitro* experiments on fresh human blood.

The only information regarding health effects in bumans resulting from acute exposure to 2-butoxyethanol acetate is a study by Jacobs et al. (1989), who found mild erythema in humans after acute dermal exposure. Very few data are available describing health effects in animals following inhalation, oral, or dermal exposure for acute-duration exposure. The data found are principally contained in three articles (Jacobs et al. 1989; Truhaut et al. 1979; Zissu 1995). Although hematology was performed in the acute-duration inhalation study conducted in rats and rabbits by Truhaut et al. (1979), only one exposure concentration was used, and selected organs were examined only for gross pathology. No effects were observed in rats, and hemoglobinuria and/or hematuria were the only effects noted for rabbits. For an acute-duration oral study on 2-butoxyethanol acetate in rats conducted by Truhaut et al. (1979), LD_{50} data were provided, gross and histological examinations were performed, and hemoglobinuria and renal effects were observed, but the doses used were not specified. In the acute dermal study in rabbits conducted by Truhaut et al. (1979), an LD_{50} value was determined, gross and histological examinations were performed, and hematological and renal effects were observed at $\geq$ 3,191 mg/kg. In the study by Jacobs et al. (1989), the only information provided was that dermal exposure of rabbits to 2-butoxyethanol acetate resulted in slight erythema. 2-Butoxyethanol acetate was considered a moderate irritant by the Draize protocol and non-irritating by the European Economic Communities protocol (Zissu 1995).

Although 2-butoxyethanol acetate is readily metabolized to 2-butoxyethanol and both compounds are metabolized by the same pathways, the limited data indicate that dose-response data for the acetate may be different from that for 2-butoxyethanol. For example, the oral LD_{50} values for male Wistar rats were reported to be 1,590 mg/kg for 2-butoxyethanol (Olin 1976) and 3,000 mg/kg for 2-butoxyethanol acetate in olive oil (Truhaut et al. 1979). The contribution of a vehicle effect on the difference in toxicity between 2-butoxyethanol and the acetate is not known; it is also unknown whether there are differences in absorption between

the two compounds. Other toxicological data for 2-butoxyethanol acetate are too limited to permit comparison with the toxicological data for 2-butoxyethanol for the same species, strains, and exposure conditions. Additional animal or human data on the toxic effects of 2-butoxyethanol acetate by all routes would be useful in assessing the human risk of exposure to 2-butoxyethanol acetate. Characterization of the relative toxicity of 2-butoxyethanol acetate compared to 2-butoxyethanol would aid in the use of data already generated for 2-butoxyethanol in predicting risk factors of exposure to the acetate form. In addition, clarification of species-specific toxicity, similar to that observed for 2-butoxyethanol, would aid in understanding the risk associated with exposure to 2-butoxyethanol acetate.

Intermediate-Duration Exposure. No studies were located regarding health effects in humans following intermediate-duration exposure to 2-butoxyethanol by inhalation, oral, or dermal routes. Studies were located regarding systemic effects in animals following intermediate-duration exposure to 2-butoxyethanol by inhalation, oral, and dermal routes. In intermediate-duration inhalation studies in animals, systemic effects have been observed as follows: respiratory effects (Carpenter et al. 1986; Werner et al. 1943b), gastrointestinal effects, renal effects, body weight effects (Carpenter et al. 1956), and hematological effects (Carpenter et al. 1956; Dodd et al. 1983; Werner et al. 1943b). The intermediate inhalation data were sufficient to derive an intermediate inhalation MRL of 3 ppm based on a NO&L of 25 ppm for hematological effects in rats in the study by Dodd et al. (1983). In intermediate-duration oral studies in animals, systemic effects have been observed as follows: gastrointestinal effects (NTP 1993), hematological effects (Eastman Kodak 1983; Krasavage 1986; Nagano et al. 1979,1984; NTP 1993), hepatic effects and renal effects (Carpenter et al. 1956; Eastman Kodak 1983; Heindel et al. 1990; Krasavage 1986; NTP 1993; Weil and Carpenter 1963), and body weight effects (Carpenter et al. 1956; NTP 1993; Weil and Carpenter 1963). The intermediate oral data were sufficient to derive an intermediate oral MRL of 0.07 mg/kg/day for hepatic effects in rats based on the study by NTP (1993). Only one intermediate-duration dermal study was located. In this study (CMA 1983), rabbits were treated dermally for 90 days at ≤ 150 mg/kg, and comprehensive hematology and histological examinations were performed for all relevant systemic organs. The only treatment-related effects were slight to moderate erythema and edema at ≥ 10 mg/kg/day.

The hematological system is the primary target for 2-butoxyethanol toxicity. Therefore, further animal studies designed to confirm and extend the data of Dodd et al. (1983) for inhalation exposure, and animal studies for intermediate-duration exposure by oral administration would be helpful in defining the NOAEL for hematological, hepatic, and renal effects, since these effects occurred at the lowest doses tested (the intermediate oral MRL is based on a LOAEL). Additionally, although some data exist that describe health

effects of 2-butoxyethanol in rabbits after intermediate dermal exposure (CMA 1983), studies using higher doses might define target organs and doses that cause bematological effects. These data are important, since dermal exposure to 2-butoxyethanol vapor and aqueous solutions is a primary route of human exposure. Furthermore, there are populations surrounding hazardous waste sites that might be exposed to 2-butoxy-ethanol for intermediate durations.

No studies were located regarding health effects in humans after inhalation, oral, or dermal intermediate-duration exposure to 2-butoxyethanol acetate. No studies were located regarding health effects in animals after intermediate-duration oral or dermal exposure to 2-butoxyethanol acetate. Limited data are available describing health effects in animals following intermediate-duration inhalation exposure. The data found are contained in one article (Truhaut et al. 1979). In this study, in which rats and rabbits were exposed to only one concentration of 2-butoxyethanol acetate (400 ppm for II month or 100 ppm for 10 months), histological examinations of several organs identified serious hematological (hemoglobinuria) and renal effects in rats and rabbits. Additional studies defining dose-response data for these and other possible effects of 2-butoxyethanol acetate after intermediate-duration exposure and clarifying how it relates to 2-butoxyethanol would be useful in predicting human toxicity to 2-butoxyethanol acetate exposure.

Chronic-Duration Exposure and Cancer. Statistically significant changes in hematocrit (decrease) and mean corpuscular hemoglobin concentration (increase) were reported in men occupationally exposed to 0.46-0.75 ppm 2-butoxyethanol for l-6 years (Haufroid et al. 1997). The changes were small, showed no relationship to exposure concentration and were still within normal biological variability. Although the effects may be early indicators of potential adverse effects in humans, and they are consistent with effects in animals, ATSDR considers the average exposure (0.6 ppm) to be a NOAEL. This study serves as the basis of the chronic-duration inhalation MRL of 0.2 ppm. No effects on serum creatinine, urinary retinol binding protein, or serum transaminase levels were noted.

No studies were located regarding health effects in humans or animals after chronic inhalation, oral, or dermal exposure or in animals after chronic inhalation exposure to 2-butoxyethanol or 2-butoxyethanol acetate. The primary targets for adverse systemic effects of 2-butoxyethanol following acute- and intermediate-duration exposure are the hematological and renal systems. Since chronic low-level exposure to 2-butoxyethanol and the acetate form by the inhalation, oral, or dermal routes is possible for populations living near waste sites containing these chemicals, inhalation, oral, and dermal studies on health effects of chronic exposure to

2-butoxyethanol and 2-butoxyethanol acetate in animals would be useful in defining potential adverse health effects.

No studies were located regarding carcinogenic effects in humans or animals after exposure to 2-butoxyethanol and 2-butoxyethanol acetate by any route. These compounds have not been classified for carcinogenic effects by the Department of Health and Human Services (DHHS), the National Toxicology Program (NTP), the International Agency for Research on Cancer (IARC), or the Environmental Protection Agency (EPA). Histological examination of comprehensive organs and tissues in animals in chronic studies performed by any route of exposure would be useful in determining potential carcinogenic effects of 2-butoxyethanol or 2-butoxyethanol acetate; however, the genotoxicity data available for 2-butoxyethanol suggest that it is unlikely to cause cancer by damaging DNA.

Genotoxicity. The only *in vivo* study was in workers exposed to both 2-butoxyethanol and the structurally related 2-ethoxyethanol (Sohnlein et al. 1993). No increases in micronuclei or sister chromatid exchanges were observed in these workers. Several reports of *in vitro* studies were found (Chiewchanwit and Au 1995; Gollapudi et al. 1996; Hoflack et al. 1995; Kvelland 1988; McGregor 1984; Tyler 1982). [^{14}C]2-Butoxy-ethanol was positive in a test that monitored increases in DNA-associated radioactivity in rat primary hepatocytes (McGregor 1984). However, 2-butoxyethanol was negative in tests of mutagenicity with bacteria (Kvelland 1988) and sister chromatid exchange (Tyler 1982) or mutations (Chiewchanwit and Au 1995) in Chinese hamster ovary cells, with or without metabolic activation. 2-Butoxyethanol was negative for mutation in *Salmonella typhimurium* strains TA98, TA100, and TA102 both with and without rat S-9 activation (Hoflack et al. 1995). In *S. typhimurium* strain TA97a 2-butoxyethanol tested positive for mutation both with and without S-9 activation. The mutation assay of 2-butoxyethanol in strain TA97a was repeated by Gollapudi et al. (1996). In this study, 2-butoxyethanol was negative for mutations both with and without rat S-9 metabolic activation, and it was negative in *E. coli WP2uvr*A strain. Gollapudi et al. (1996) suggested that Hoflack et al. (1995) found positive results because they used 2-butoxyethanol contaminated with higher levels of peroxides, which test positive in strain TA97a. The 2-butoxyethanol used by Gollapudi et al. (1996) contained 23 ppm of peroxides which was measured just before the tests. The 2-butoxyethanol used by Hoflack et al. (1995) contained less than 50 ppm peroxides; the information was provided by the manufacturer, so the actual concentrations of peroxides may have been higher. Gollapudi et al. (1996) concluded that peroxides and/or experimental variables probably account for the difference in results observed in their study and the study by Hoflack et al. (1995). The available data suggest that 2-butoxyethanol is not genotoxic. The exception may be when it is contaminated with peroxides. No studies were located regarding

genotoxicity of 2-butoxyethanol acetate. Further studies of 2-butoxyethanol and 2-butoxyethanol acetate in *vivo* and *in vitro* would more fully determine the genotoxic potential of these compounds.

Reproductive Toxicity. No studies were located regarding reproductive effects in humans after inhalation, oral, or dermal exposure to 2-butoxyethanol or 2-butoxyethanol acetate.

For 2-butoxyethanol, two inhalation (800 ppm for 3 hours; ≤ 867 ppm for 4 hours; ≤245 ppm for 9 days; ≤77 ppm for 13 weeks) studies (Dodd et al. 1983; Doe 1984), several oral (≤500 mg/kg administered once; 500 or 1,000 mg/kg for 4 days; ≤346 or ≤627 mg/kg/day for 2 weeks) studies (Eastman Kodak 1983; Exon et al. 1991; Ghanayem et al. 1987; Grant et al. 1985; Heindel et al. 1990; Krasavage 1986; Nagano et al. 1979, 1984; NTP 1993), and two dermal (doses up to 150 mg/kglday for 90 days or 18-361 mg/kg/day for 11 days) studies (CM4 1983; Union Carbide 1980a) in animals measured testicular weight and/or performed histological examination of male reproductive organs and found no effect. However, one intermediateduration oral (≤443 mg/kg/day for 60 days) study found decreased sperm concentration in male rats (NTP 1993), and a go-day study reported testicular atrophy in rats (Weil and Carpenter 1963). In the go-day dermal study (doses of up to 150 mg/kg/day) in rabbits (CMA 1983), there was only a slight increase in testicular weight of rabbits. Reproductive effects (implantation loss, decreases in live fetuses, and/or increase in nonviable implants) have been found in pregnant rats and rabbits exposed to 2-butoxyethanol(200 ppm) by inhalation during gestation (Tyl et al. 1984). In oral (≤ 363 mg/kg/day for 13 weeks) studies, female rats, but not female mice, had altered estrous cycles (NTP 19939, although the total duration of the cycle was not altered. Pregnant rats (treated with 600 mg/kg/day on gestation days 9-l 1 or 1 l-13) had vaginal bleeding, increased resorptions, and implantation loss (NTP 1989), and pregnant mice (treated with 1,180 mg/kg/day on gestation days 6-13) had decreases in viable litters (Hardin et al. 1987; Schuler et al. 1984) and increased resorptions (Wier et al. 1987). In a breeding-pair study in male and female mice (treated with 1,300 or 2,100 mg/kg/day for 21 weeks), there was a decrease in the number of litters produced per pair that was found to be a result of effects on female reproduction in cross-mating experiments (Heindel et al. 1990). However, in the NTP (1993) drinking water study, histological examination of female rat and mouse reproductive organs (NTP 1993), and vaginal cytology evaluations of rats and/or mice revealed no pathology. Histological examination of the reproductive organs of female rabbits in the go-day dermal study by CMA (1983) also revealed no pathology. Since several studies that performed histological examination of reproductive organs of animals exposed by all routes were negative, additional studies of this kind do not appear to be useful. One oral continuous breeding study was performed in mice, a drinking water study (Heindel et al. 1990) that has also been summarized in Morrissey et al. (1988, 1989). An oral reproductive study in rats and reproductive

studies in two species conducted by the inhalation and dermal routes (which are the two routes most relevant for humans) would help to complete the picture of reproductive effects of 2-butoxyethanol in animals and help to determine whether 2-butoxyethanol exposure presents a reproductive hazard to humans.

For 2-butoxyethanol acetate, no gross pathological lesions were found in the testes or ovaries of rats or rabbits exposed to 400 ppm for 4 hours and no gross or histopathological lesions were found in the testes or ovaries of rats or rabbits exposed by inhalation intermittently to 400 ppm for 1 month or 100 ppm for 10 months, in rats at unspecified single gavage doses, or in rabbits exposed dermally at ≤ 10,000 mg/kg for 24 hours (Truhaut et al. 1979). However, no reproductive function studies were located.

The results of both 2-butoxyethanol and 2-butoxyethanol acetate studies indicate that further histological examination of male and female reproductive organs would not be particularly useful, but reproductive studies in mated pairs of two species exposed to 2-butoxyethanol acetate by all routes would be useful to determine the reproductive toxicity potential of this compound.

Developmental Toxicity. No studies were located regarding developmental effects in humans after inhalation, oral, or dermal exposure to 2-butoxyethanol or 2-butoxyethanol acetate.

Developmental effects have been observed in animals after inhalation, oral, and dermal exposure to 2-butoxyethanol. In inhalation studies on 2-butoxyethanol in animals, the developmental effects consisted of retarded skeletal ossification in the offspring of pregnant rats and pregnant rabbits (Tyl et al. 1984). In oral studies, the developmental effects consisted of decreased fetal body weight of the rats and decreased gravid uterine weight of the rat dams (NTP 1989), some evidence of cleft palate in the fetuses of pregnant mice (Wier et al. 1987), and decreased pup weight in the offspring of mating pairs of rats (Heindel et al. 1990). In dermal studies, no developmental effects were observed in the offspring of pregnant rats exposed at 0.48 mL/day during gestation (Hardin et al. 1984). A higher dermal dose (1.4 mL/day) caused mortality of the dams. With the exception of decreased pup weight observed in mice (Heindel et al. 1984), developmental effects reported occurred at 2-butoxyethanol doses that resulted in moderate to severe maternal toxicity (Hardin et al. 1987; NTP 1989; Tyl et al. 1984; Wier et al. 1987). An additional developmental study on 2-butoxyethanol by the dermal route in another species (mouse or rabbit) would be useful, since dermal exposure to 2-butoxyethanol in cleaning solutions is a primary route of exposure in the general population.

No studies were located regarding developmental effects in animals after inhalation, oral, or dermal exposure to 2-butoxyethanol acetate. Studies of the developmental toxicity of this compound by inhalation, oral, and dermal routes would be useful in predicting potential developmental toxicity, and relating the toxicity of 2-butoxyethanol acetate to 2-butoxyethanol.

Immunotoxicity. No studies were located regarding immunological and lymphoreticular effects in humans after inhalation or oral exposure to 2-butoxyethanol or 2-butoxyethanol acetate. Slight erythema, but no allergic reaction, was noted on skin-patch tests with 2-butoxyethanol acetate in humans (Jacobs et al. 1989). A limited human dermal sensitivity study in 201 persons with 10% 2-butoxyethanol showed no overt effect (CMA 1992). The 10% concentration was tested because it is the highest concentration found in cosmetics. 2-Butoxyethanol has also tested negative for dermal sensitization in guinea pigs using the maximized Magnusson and Kliman test (Zissu 1995).

In two oral studies, the information on immunological effects of 2-butoxyethanol in animals consists of the findings that no effect on the immune system was found in rats (Exon et al. 1991; Smialowicz et al. 1992), and no effect on thymus and spleen weight and no histopathological lesions in the thymus were observed in rats (Exon et al. 1991).

Studies in animals describe effects on the spleen, many of which are related to the hematological effect of 2-butoxyethanol. In inhalation studies, splenic effects consisted of increased spleen weight of pregnant rats (Tyl et al. 1984). In oral studies, splenic and other lymphoreticular effects in animals consisted of increased spleen weight (Eastman Kodak 1983; Ghanayem et al. 1987a, 1987b, 1992; Grant et al. 1985; Krasavage 1986; NTP 1989), extramedullary splenic hematopoiesis (Grant et al. 1985), splenic congestion and enlarged dark spleens (Eastman Kodak 1983; Krasavage 1986), hemosiderosis in the spleen, decreased thymus weight, and bone marrow hyperplasia and increased hematopoiesis (NTP 1993). In dermal studies on 2-butoxyethanol in animals, effects on lymphoreticular organs consisted of congestion in the spleen, erythrocytic infiltration, and white atrophic pulp (Duprat and Gradiski 1979); engorged spleen (Carpenter et al. 1956); and enlarged and dark red spleen (Union Carbide 1980a, 1980b).

One reasonably thorough study examined immune function in rats orally dosed at up to 400 mg/kg/day 2-butoxyethanol twice after immunization with trinitrophenyl-lipopolysaccharide (TNP-LPS) (Smialowicz et al 1992). Compared with controls, the 2-butoxyethanol had no adverse effect on antibody production, delayed B type hypersensitivity, natural killer cell function, or splenocyte production of cytokines.

Since the effects on the spleen and bone marrow are secondary to the hemolytic effects of 2-butoxyethanol, and since a number of studies performed histological examination of lymphoreticular organs, additional studies on lymphoreticular effects do not appear to be useful. However, the potential for immunotoxicity of 2-butoxyethanol has not been sufficiently studied. A battery of immunotoxicity tests conducted by all routes, but particularly by the inhalation and dermal routes (the most relevant routes for humans) would help to determine whether 2-butoxyethanol is immunotoxic.

For 2-butoxyethanol acetate, no gross or histopathological lesions were found in the spleen of rats or rabbits exposed by inhalation intermittently to 400 ppm for 1 month or to 100 ppm for 10 months, in rats at unspecified single gavage doses, or in rabbits exposed dermally at ≤ 10,000 mg/kg for 24 hours (Truhaut et al. 1979). Studies such as those suggested for 2-butoxyethanol would be helpful not only in defining the effect of 2-butoxyethanol acetate on the immune system, but also in relating it to the toxicity of 2-butoxyethanol.

Neurotoxicity. The only information on neurological effects in humans exposed to 2-butoxyethanol is that humans exposed experimentally by inhalation experienced headache and disturbed taste sensation (Carpenter et al. 1956), and that people who intentionally ingested household cleaning agents containing 2-butoxyethanol became comatose (Bauer et al. 1992; Gijsenbergh et al. 1989; Eitovitz et al. 1991; Rambourg-Schepens et al. 1988).

Neurological effects have been observed in animals after inhalation, oral, and dermal exposure to 2-butoxyethanol. In inhalation studies of 2-butoxyethanol in animals, neurological effects consisted of loss of coordination (Dodd et al. 1983), extreme weakness and apathy (Carpenter et al. 1956), excessive salivation (Dow 1972, 1986), and poor coordination of the extremities and loss of equilibrium (Dow 1986). In oral studies of 2-butoxyethanol in animals, neurological effects consisted of sluggishness, prostration, narcosis, lethargy, failure to right, and/or ataxia (Carpenter et al. 1956; Dow 1959, 198 1; Eastman Kodak 1983; Krasavage 1986; NTP 1989; Olin 1976; Sivarao and Mehendale 1995; Shepard 1994b; Union Carbide 1980b; Wier et al. 1987). In dermal studies of 2-butoxyethanol in animals, neurological effects consisted of prostration, narcosis, convulsion, nystagmus, anorexia, lack of spontaneous movement, and/oFinactivity (Duprat and Gradiski 1979; Hardin et al. 1984; Olin 1976; Union Carbide 1980a). Most of these effects occurred in animals prior to death at high lethal doses. In intermediate-duration inhalation (Dodd et al. 1983) oral (Eastman Kodak 1983; Krasavage 1986; NTP 1993), and dermal (CMA 1983) studies, comprehensive histological examinations of the brain, spinal cord, and/or sciatic nerves revealed no lesions.

For 2-butoxyethanol acetate, no clinical signs of neurotoxicity and no histopathological lesions in the brain were found in rats or rabbits exposed by inhalation intermittently to 400 ppm for 1 month or to 100 ppm for 10 months, in rats at unspecified single gavage doses, or in rabbits exposed dermally at ≤ 10,000 mg/kg for 24 hours (Truhaut et al. 1979).

A battery of neurological and behavioral tests in animals after inhalation, oral, and dermal exposure would help to determine whether 2-butoxyethanol and 2-butoxyethanol have more subtle neurobehavioral effects.

Epidemiological and Human Dosimetry Studies. Studies of human populations exposed to 2-butoxyethanol or 2-butoxyethanol acetate primarily involve evaluations of kinetic parameters and biomonitoring for occupational exposure after inhalation or dermal exposure (Angerer et al. 1990; Haufroid et al. 1997; Johanson and Boman 1991; Johanson and Johnsson 1991; Johanson et al. 1986a, 1988; Rettenmeier et al. 1993; Sakai et al. 1994; Vincent et al. 1993). Hematological effects have been examined in a small group of men occupationally exposed to 2-butoxyethanol and methyl ethyl ketone (Haufroid et al. 1997). Small but statistically significant changes in hematocrit (decrease) and mean corpuscular hemoglobin concentration (increase) that were still within normal biological variability were reported. Although the effects may be early indicators of potential adverse effects in humans, and they are consistent with effects in animals, ATSDR considers the average exposure (0.6 ppm) to be a NOAEL. This study serves as the basis of the chronic-duration inhalation MRL of 0.2 ppm. The study authors indicated that the hematological effects should be confirmed in additional populations exposed to 2-butoxyethanol. One study regarding genotoxicity and exposure assessment in workers exposed to both 2-butoxyethanol and 2-ethoxyethanol (Sohnlein et al. 1993) is available. Occupational exposure to 2-butoxyethanol and 2-butoxyethanol acetate by the inhalation or dermal route comprises a large proportion of the potential exposure in the population. Thus, epidemiological studies of humans exposed occupationally, or at or near hazardous waste sites would provide valuable information on the potential toxicity of the compound. This information would be useful, especially in light of the apparent species differences in relative toxicity.

Biomarkers of Exposure and Effect

Exposure. The presence of 2-butoxyacetic acid in the urine is the accepted biomarker of exposure for both 2-butoxyethanol and 2-butoxyethanol acetate (Johanson et al. 1989), and it has been used as a biomarker in surveys of occupational exposure; 2-butoxyethanol in the blood and urine or 2-butoxyacetic acid and its glutamine conjugate in urine have also been used as biomarkers (Angerer et al. 1990; Haufroid et al. 1997;

Johanson and Boman 199 1; Johanson and Fernstrom 1986; Johanson and Johnsson 199 1; Johanson et al. 1986a, 1988, 1989; Jiinsson and Steen 1978; Rettenmeier et al. 1993; Sakai et al. 1994; Vincent et al. 1993). These have also been used as biomarkers of exposure in animal experiments (Ghanayem et al. 1987a, 1987b, 1987c, 1992; Medinsky et al. 1990; Romer et al. 1985; Sabourin et al. 1992a, 1992b, 1993). More recently, analytical methods have been able to detect 2-butoxyacetic acid in human blood (Johanson and Johnsson 1991). These biomarkers are unique, relatively short-lived, and serve well for monitoring exposure. However, the exact relationship between blood or urine levels of these compounds and exposure by the inhalation, oral, or dermal routes is still not clear; some of this may be due to measurement of only free 2-butoxyacetic acid in the urine rather than the total of free and conjugates. Additional studies of exposure and blood and urine levels of these compounds would be helpful in predicting the health effects of exposure in the occupational setting, and in instances of exposure of populations near waste sites, or those using consumer products containing 2-butoxyethanol or the acetate form. An exploratory study of the role of genetic polymorphism of CYP 2El on 2-butoxyethanol metabolism by humans would also be useful. Haufroid et al. (1997) looked at urinary metabohtes in a small sample of humans exposed to 2-butoxyethanol; the subject included 30 c1/c1 homozygotes and one c1/c2 heterozygote. Individuals beterozygous for the c2 allele may produce less 2-butoxyethanol acid, and more ethylene glycol than individual homozygous for the c1 allele; however, ethylene glycol and its metabolites were not measured in the urine in this study and only free butoxyacetic acid, not conjugates, was measured.

Effect. Monitoring of hematological parameters may be useful as a biomarker of effect of 2-butoxyethanol and 2-butoxyethanol acetate. Toxic effects occur in the red blood cells and are either from 2-butoxyethanol or its metabolites (Bartnik et al. 1987; Ghanayem et al. 1987a, 1987b). However, hematological effects are not specific to 2-butoxyethanol. The tests cannot be relied upon to find preclinical disease, but may be useful in identifying changes that are early indicators of effects Furthermore, these markers of effect may not be useful for long periods following cessation of exposure, nor do they distinguish between acute and chronic exposure. The use of urinary D-glucaric acid as a biomarker of effect following exposure to 2-butoxyethanol has been examined in foundry workers exposed to the compound in paints (Collinot et al. 1996). Although measurement of D-glucaric acid is not an effect specific to 2-butoxyethanol exposure, the study authors concluded that it was more sensitive than a standard blood count. Further investigation of possible biomarkers of 2-butoxyethanol or 2-butoxyethanol acetate exposure would be helpful in monitoring the health effects of these chemicals.

Absorption, Distribution, Metabolism, and Excretion. Data from both humans (Angerer et al. 1990; Johanson and Boman 199 1; Johanson and Johnsson 1991; Johanson et al. 1986a, 1989) and rats (Johanson 1994; Sabourin et al. 1992a) indicate that 2-butoxyethanol is rapidly and extensively absorbed through the lungs. No studies were located regarding pulmonary absorption in other animal species. Although no experimentally acquired data were available on gastrointestinal absorption of 2-butoxyethanol in humans, some data are available from case reports of oral poisonings, which suggest that absorption from the gastrointestinal system is rapid (Bauer et al. 1992; Gijsenbergh et al. 1989; Gualtieri et al. 1995; Litovitz et al. 1991; Rambourg-Schepens et al. 1988). Data from rat studies corroborate these results (Corley et al. 1994; Ghanayem et al. 1987c; Medinsky et al. 1990), but no data were available for other animal species. Studies in humans exposed experimentally (Corley et al. B 997; Johanson and Boman 199 1; Johanson et al. 1988) or occupationally (Vincent et al. 1993) indicate that 2-butoxyethanol can be absorbed through the skin, both from the vapor and from the liquid form. Results suggest that persons that expose large portions of their skin to the liquid form of 2-butoxyethanol are at risk of absorbing acutely toxic doses. *In vitro* studies using human skin also indicate dermal absorption (Bartnik et al. 1987; Dugard et al. 1984). Data from rats (Bartnik et al. 1987; Sabourin et al. 1992b, 1993) and guinea pigs (Johanson and Fernstrom 1986) exposed dermally support this conclusion. An *in vitro* study using skin from humans, rats, and pigs shows that the absorption of neat 2-butoxyethanol was initially slower than from aqueous solutions, such as all purpose cleaners (Bartnik et al. 1987).

No information on distribution of 2-butoxyethanol to tissues in humans was located. Following absorption into the body, 2-butoxyethanol was widely distributed from the blood to tissues of rats after inhalation (Johanson 1994), oral (Ghanayem et al. 1987b, 1987c), and dermal (Bartnik et al. 1987) exposure.

There is no evidence to suggest that the route of admiuistration has any substantial effect on the subsequent metabolism of 2-butoxyethanol in animals, although the route of administration and the dose may alter the relative amounts of individual metabolites (Medinsky et al. 1990; Sabourin et al. 1992a, 1992b, 1993). 2-Butoxyacetic acid (free or glycine or glutamine conjugates) is the major blood and urinary metabolite of 2-butoxyethanol in humans after inhalation exposure (Johanson and Johnsson 199 1; Johansouet al. 1986a), while oxalate, formed from metabolism of ethylene glycol (a hydrolysis product of 2-butoxyethanol), was found in the urine of a patient after ingestion (Rambourg-Schepens et al. 1988). 2-Butoxyacetic acid is also the major urinary metabolite in rats, guinea pigs, dogs, rabbits, dogs, and monkeys exposed to 2-butoxyethanol by inhalation (Carpenter et al. 1956). Other metabolites in rats after inhalation, oral, or dermal exposure are expired CO_2, ethylene glycol, and glucuronide and sulfate conjugates of 2-butoxyethanol in the

urine, and 2-butoxyacetic acid and 2-butoxyethanol-glucuronide conjugate in the bile (Corley et al. 1994; Ghanayem et al. 1987c; Sabourin et al. 1992a, 1992b, 1993). Based on the rat data, metabolic pathways have been elucidated. 2-Butoxyethanol is oxidized via alcohol dehydrogenase to the aldehyde, then further oxidized to 2-butoxyacetic acid by aldehyde dehydrogenase. 2-Butoxyacetic acid can then be detoxified by conjugation with glycine or glutamine, or it can be broken down to CO_2. 2-Butoxyethanol can also be metabolized to ethylene glycol, another toxic glycol ether, or it can be directly detoxified by conjugation and excreted. Biotransformation of 2-butoxyethanol to 2-butoxyacetic acid is necessary for 2-butoxyethanol-induced toxicity. It is clear, however, that the metabolic pathway can be influenced, thus altering the amount of toxic metabolites and subsequent toxicity. Additional studies on conditions that favor detoxification of the parent compound through conjugation over metabolism of the parent compound to its toxic metabolite 2-butoxyacetic acid would be useful in not only understanding the toxic reactions after exposure, but may also suggest ways to prevent or minimize toxicity after exposure. These studies might include the examination of the effects of polymorphism of CYP 2El in humans on the metabolism of 2-butoxyethanol to follow up one study in which a person with one alternate allele had less free 2-butoxyacetic acid in urine than 30 with 2 copies of a common allele (Haufroid et al. 1997).

Respiratory excretion of 2-butoxyethanol consists of expired CO_2 and unchanged parent compound (Bartnik et al. 1987; Ghanayem et al. 1987a; Medinsky et al. 1990; Sabourin et al. 1992a, 1992b, 1993). However, urinary excretion of 2-butoxyethanol, 2-butoxyacetic acid, ethylene glycol, and the conjugates is the primary route in both humans and animals (Bartnik et al. 1987; Carpenter et al. 1956; Ghanayem et al. 1987a, 1987b, 1987c; Gijsenbergh et al. 1989; Johanson 1994; Johanson and Johnsson 1991; Johanson et al. 1986a, 1988; Jijnsson and Steen 1978; Rambourg-Schepens et al. 1988; Rettenmeier et al. 1993; Sabourin et al. 1992a, 1992b, 1993; Sakai et al. 1994; Vincent et al. 1993). 2-Butoxyethanol, 2-butoxyacetic acid, and the glucuronic conjugate of 2-butoxyethanol are also excreted in the bile (Ghanayem et al. 1987a, 1987b, 1987c). Fecal excretion is a minor route (Bartnik et al. 1987; Ghanayem et al. 1987b, 1987c; Medinsky et al. 1990; Sabourin et al. 1992a, 1992b, 1993).

The absorption of 2-butoxyethanol acetate has not been extensively studied, although in humans occupationally exposed to the acetate, 2-butoxyacetic acid was detected in the urine (Johanson et al. 1989), and absorbed doses of the acetate were calculated for rabbits exposed dermally (Truhaut et al. 1979). No studies regarding distribution of 2-butoxyethanol acetate were located. The metabolism of 2-butoxyethanol acetate has not been extensively studied, presumably because 2-butoxyethanol acetate is assumed to be metabolized to 2-butoxyethanol in the body, and then to follow the same pathway as 2-butoxyethanol, since

2-butoxyacetic acid has been detected in human urine after 2-butoxyethanol acetate exposure (Johanson et al. 1989). Other than detection of 2-butoxyacetic acid in urine in humans occupationally exposed to 2-butoxy-ethanol acetate, no studies were located regarding excretion of the acetate form.

The absorption, distribution, metabolism, and excretion of 2-butoxyethanol are relatively well understood, leading to a number of PBPK models (Corley 1996; CorIey et al. 1994; Johanson 1986,199 1 a; Johanson and Naslund 1988; Shyr et al. 1993). These models have been validated with acute- and intermediate-duration pharmacokinetic data. Chronic pharmacokinetic data would be useful so that the models could be validated for chronic exposure. Additional studies designed to define the differences and similarities between 2-butoxyethanol and its acetate with regard to absorption, distribution, metabolism, and excretion would be useful to establish these parameters for to 2-butoxyethanol acetate.

Comparative Toxicokinetics. Qualitatively, absorption, metabolism, and excretion appear to be similar in humans and laboratory animals (see discussion above). However, quantitative variations in absorption, metabolism, and excretion of 2-butoxyethanol have been observed with respect to age in rats (Ghanayem et al. 1987a). In addition, the efficiency of absorption of 2-butoxyethanol and excretion of 2-butoxyacetic acid after inhalation exposure differs for several animal species (Carpenter et al. 1956). However, the absorption, distribution, metabolism, and excretion of 2-butoxyetbanol after inhalation, oral, and dermal exposure have been studied extensively only in rats. Further studies that focus on the age-related and species-related differences and their implications for human health would be useful. Additionally, *in vitro* studies using human tissue and further research into PBPK modeling in animals would contribute significantly to the understanding of the kinetics of 2-butoxyethanol and would aid in the development of pharmacokinetic models of exposure in humans.

Methods for Reducing Toxic Effects. Development of methods and practices that are specific for 2-butoxyethanol or 2-butoxyethanol acetate would be useful for reducing peak absorption, and body burden, and for interfering with the mechanism of action following 2-butoxyethanol or 2-butoxyethanol acetate exposure. With regard to accidental or intentional poisoning, hemodialysis (Gijsenbergh et al.-1989), gastric lavage, and emesis have been used in the prevention of toxicity (Dean and Krenzelok 1992), and administration of ethanol has been suggested as appropriate therapy (Buckley et al. 1993). Based on animal studies, the administration of inhibitors of alcohol dehydrogenase and aldehyde dehydrogenase may also be appropriate (Ghanayem et al. 1987b). Since 2-butoxyethanol metabolites are thought to play the major role in the toxicity, more information is needed about their interaction with cell membranes and cellular

macromolecules. This information would help the development of methods for possible prevention of 2-butoxyethanol-induced toxicity. Special attention should be paid to the production of toxic metabolites, and preferential metabolism to detoxification products.

2.10.3 Ongoing Studies

The open literature contained preliminary reports of ongoing studies in two areas. In an abstract, Raymond et al. (1994) reported immediate and late effects of acute exposure to 2-butoxyethanol in seven clerical workers. Severe, immediate eye and upper respiratory irritation, presyncope, and nausea were observed. Exposure resulted from floor stripping with undiluted 2-butoxyethanol in a small room with little ventilation. The symptoms suggest exposure at 100-300 ppm. All seven workers recovered within several days but noticed skin lesions (cherry angiomas) and recurrent upper respiratory irritation beginning 3 months later. In another preliminary report, the flow properties of rat red blood cells were investigated after oral exposure to 50-500 mg/kg 2-butoxyethanol (Kurantsin-Mills et al. 1992). The physical characteristics of the red blood cells were determined at 0.5,2, and 4 hours after treatment. Membrane and cellular changes were noted that compromise the flow properties of the cells, thus possibly affecting microcirculation, possibly resulting in hemolysis.

The Chemical Manufacturer's Association is working with the EPA to develop an RfC and an RfD for 2-butoxyethanol. A draft IRIS support document is available (CMA 1997b).

NTP has nearly completed lifetime bioassays of 2-butoxyethanol in mice and rats. The study is undergoing peer review and should provide information on both carcinogenic potential and systemic effects.

No studies regarding the health effects of 2-butoxyethanol or 2-butoxyethanol acetate were reported in the CRISP Data Base of the National Institutes of Health.