3. HEALTH EFFECTS

3.1 INTRODUCTION

The primary purpose of this chapter is to provide public health officials, physicians, toxicologists, and other interested individuals and groups with an overall perspective on the toxicology of xylene. 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.

Commercial xylene is a mixture of three isomers of xylene (*m*-, *o*-, and *p*-xylene) with <20% ethyl-benzene. In the following discussion of the health effects of xylene, the effects of both the mixture and the individual isomers are presented. Where possible, the effects of individual isomers will be identified and presented separately.

3.2 DISCUSSION OF HEALTH EFFECTS BY ROUTE OF EXPOSURE

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

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

believes that there is sufficient merit in this approach to warrant an attempt at distinguishing between "less serious" and "serious" effects. The distinction between "less serious" effects and "serious" effects is considered to be important because it helps the users of the profiles to identify levels of exposure at which major health effects start to appear. LOAELs or NOAELs should also help in determining whether or not the effects vary with dose and/or duration, and place into perspective the possible significance of these effects to human health.

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

A change from the last edition of this profile is that a single MRL is derived for each duration (acute, intermediate, or chronic) of inhalation or oral exposure that applies equally to mixed xylenes and to the individual isomers, rather than having specific MRLs for each chemical entity. This convention is in accordance with occupational exposure levels promulgated by agencies such as ACGIH, NIOSH, and OSHA (see Table 8-1) and is supported by several lines of evidence. The isomers have similar chemical properties such as log K_{ow} (see Table 4-2), resulting in similar absorption, distribution, and excretion patterns (see Section 3.4 and Table 3-6). The tissue:air partition coefficients (liver, fat, and muscle) and the blood:air partition coefficients, as well as the estimated hemoglobin binding constants, for the three isomers of xylene are almost identical or comparable (Adams et al. 2005; Poulin and Krishnan 1996a, 1996b). The xylene isomers are metabolized by the same enzymes, resulting in an isomer of methylhippuric acid as the predominant metabolite in each case (see Section 3.4.3). In addition, physiologically based pharmacokinetic (PBPK) models based on the characteristics of *m*-xylene have been shown to be able to simulate the kinetics of mixed xylenes (Tardif et al. 1993a, 1995). Toxicological data from comparative studies, as discussed by EPA (2003), demonstrate that, in some cases, the effects and effect levels of the isomers are similar; e.g., body weight findings in the acute oral study by Condie et al. (1988) or the alveolar concentration levels associated with anaesthetic effects as described by Fang et al. (1996). Other studies have indicated different orders of relative toxicity for the isomers, but there is no consistent pattern indicating that a particular isomer is the most potent for all end points, and the differences in effect levels among the isomers may be small. For example, the *ortho* isomer was most potent in assays on operant behavior (Moser et al. 1985) and motor coordination in rats

(Korsak et al. 1990), and in a developmental toxicity assay in rats in which mixed xylenes had the same effect levels (Saillenfait et al. 2003). On the other hand, the *para* isomer was most potent in a different test for motor performance, the inverted screen test (Moser et al. 1985), and in ototoxicity assays in rats (Gagnaire and Langlais 2005; Gagnaire et al. 2001). Given the lack of consistency among the different end points, the most sensitive effect by mixed xylenes or any isomer was chosen as the basis for the MRL for mixed xylenes and all isomers for that duration and route of exposure.

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

3.2.1 Inhalation Exposure

3.2.1.1 Death

One report was located regarding death in humans following acute inhalation exposure to xylene (composition unspecified) (Morley et al. 1970). One of three men died after breathing paint fumes for several hours that contained an estimated atmospheric concentration of 10,000 ppm xylene. Xylene comprised 90% of the solvent in the paint (small amounts of toluene were also present), with the total solvent comprising 34% of the paint by weight. An autopsy of the man who died showed severe pulmonary congestion, interalveolar hemorrhage, and pulmonary edema; the brain showed hemorrhaging and evidence of anoxic damage. Clinical signs noted in the two exposed men who survived included solvent odor of the breath, cyanosis of the extremities, and neurological impairment (temporary confusion, amnesia). Both men recovered completely. The authors hypothesized that anoxia did not contribute to the effects observed in the survivors because the flow of oxygen into the area in which the men were working should have been adequate. The study was inconclusive for evaluating the toxic effects of xylene because the subjects were concurrently exposed to other chemicals in the paint. No studies were located regarding mortality in humans after intermediate or chronic inhalation exposure to mixed xylene or xylene isomers.

Acute inhalation LC_{50} values have been determined in animals for xylene and its isomers (Bonnet et al. 1979; Carpenter et al. 1975a; Harper et al. 1975; Hine and Zuidema 1970; Ungvary et al. 1980b). The 4-hour LC_{50} value for mixed xylene in rats ranged from 6,350 ppm (Hine and Zuidema 1970) to 6,700 ppm (Carpenter et al. 1975a). The 4-hour LC_{50} value for *p*-xylene in rats was reported to be 4,740 ppm (Harper et al. 1975). In mice, the 6-hour LC_{50} values for *m*-, *o*-, and *p*-xylene were determined to be 5,267, 4,595, and 3,907 ppm, respectively (Bonnet et al. 1979). These data suggest that *p*-xylene

may be slightly more toxic than the other xylene isomers. According to the toxicity classification system of Hodge and Sterner (1949), these values indicate that mixed xylene and its isomers are slightly toxic by acute inhalation.

Mice appear to be more sensitive than rats to the lethal effects of the *m*- and *o*-isomers of xylene (Cameron et al. 1938). While no rats died following a 24-hour exposure to 2,010 ppm *m*-xylene, 6 of 10 mice died as a result of a similar exposure. Similarly, a 24-hour exposure of rats to 3,062 ppm *o*-xylene resulted in a death rate of only 1 in 10, whereas in mice, 4 of 10 died. It is unclear whether differential sensitivities exist for the *p*-isomer of xylene in mice and rats (Cameron et al. 1938).

Information regarding lethality following intermediate-duration exposures is limited to the results of a single study examining mortality in rats, guinea pigs, monkeys, and dogs following intermittent and continuous exposure to *o*-xylene (Jenkins et al. 1970). Continuous exposure to 78 ppm *o*-xylene for 90–127 days resulted in the death of only 1 of 15 rats. Intermittent exposure to 780 ppm *o*-xylene resulted in deaths of 3 of 15 rats; none of the 15 guinea pigs, 3 monkeys, or 2 dogs died. No data were located regarding death following chronic-duration exposure to mixed xylene or its isomers.

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

3.2.1.2 Systemic Effects

No human or animal data were available regarding dermal effects following inhalation exposure to mixed xylene or xylene isomers. The systemic effects observed after inhalation exposure to xylene are discussed below. The highest NOAEL value and all LOAEL values from each reliable study for systemic effects in each species and duration category are recorded in Table 3-1, and are plotted in Figure 3-1.

Respiratory Effects. Self-reported symptoms of respiratory irritation and impaired performance in tests of pulmonary function have been observed in studies of volunteers exposed to xylene for short periods of time under controlled conditions. In humans, nose and throat irritation has been reported following exposure to mixed xylene at 200 ppm for 3–5 minutes (Nelson et al. 1943), to *m*-xylene at 50 ppm for 2 hours (Ernstgard et al. 2002), and to *p*-xylene at 100 ppm for 1–7.5 hours/day for 5 days (NIOSH 1981). However, no increase in reports of nose and throat irritation and no change in respiratory rate were seen in a study of subjects exposed to mixed xylene at a concentration of 396 ppm for

Table 3-1 Levels of Significant Exposure to Xylene - Inhalation

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (ppm)	LOAEL Less Serious (ppm)	LOAEL Serious (ppm)	Reference Chemical Form	Comments
ACUTE EXPOSURE								
Death								
1	Rat Wistar	24 hr				3062 (1/10 died)	Cameron et al. 1938 ortho	
2	Rat (Wistar)	12 hr				19650 (8/10 died)	Cameron et al. 1938 para	
3	Rat Harlan-Wistar	4 hr				6700 M (LC50)	Carpenter et al. 1975a mixed	
4	Rat CD	4 hr				4740 F (LC50)	Harper et al. 1975 para	
5	Rat Long- Evans	4 hr				6350 M (LC50)	Hine and Zuidema 1970 mixed	
6	Rat CFY	7 d 24 hr/d				700 F (4/30 died)	Ungvary et al. 1980b meta	
7	Mouse SPF-Of1	6 hr				3907 F (LC50)	Bonnet et al. 1979 para	
8	Mouse SPF-Of1	6 hr				5267 F (LC50)	Bonnet et al. 1979 meta	
9	Mouse SPF-Of1	6 hr				4595 F (LC50)	Bonnet et al. 1979 ortho	
10	Mouse NS	24 hr				3062 (4/10 died)	Cameron et al. 1938 ortho	

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

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (ppm)	LOAEL Less Serious (ppm)		LOAEL Serious (ppm)		Reference Chemical Form	Comments
11	Mouse NS	12 hr					19650	(9/10 died)	Cameron et al. 1938 para	
12	Mouse NS	24 hr					2010	(6/10 died)	Cameron et al. 1938 meta	
Systemic										
13	Human	0.25 hr	Resp	460	690	(throat irritation)			Carpenter et al. 1975a mixed	
			Ocular	230	460	(eye irritation)				
14	Human	2 hr	Resp		50	(decreased forced vital capacity; increased severity score for throat/airway discomfort, breathing difficulty, nose irritation)			Ernstgard et al. 2002 meta	
			Ocular		50	(slight eye irritation)				
15	Human	2 or 3 d 70 min/d	Cardio	299 M					Gamberale et al. 1978 mixed	
16	Human	30 min	Resp	396 M					Hastings et al. 1986 mixed	
			Ocular	396 M						

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

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (ppm)	LOAEL Less Serious (ppm)	LOAEL Serious (ppm)	Reference Chemical Form	Comments
17	Human	2-6 d 5-5.5 hr/d	Resp	200 M			Laine et al. 1993 meta	
			Cardio	200 M				
			Hemato	200 M				
18	Human	3-5 min	Resp		200 (nose and throat irritation)		Nelson et al. 1943 mixed	
			Ocular		200 (eye irritation)			
19	Human	5 d 1-7.5 hr/d	Resp		100 F (nose and throat irritation)		NIOSH 1981 para	
			Cardio	100 F				
			Hemato	100 F				
			Renal	100 F				
			Ocular		100 F (eye irritation)			
20	Human	7 hr	Cardio	100 M			Ogata et al. 1970 para	
21	Human	7 hr	Cardio	200 M			Ogata et al. 1970 meta	

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

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (ppm)	LOAEL Less Serious (ppm)	LOAEL Serious (ppm)	Reference Chemical Form	Comments
22	Human	4 d 3.67 hr/d	Resp	200 M			Seppalainen et al. 1989 meta	
			Cardio	200 M				
23	Rat Harlan-Wistar	0.75 hr	Hemato	15000 M			Carpenter et al. 1975a mixed	
24	Rat Wistar	1 or 2 wk 5 d/wk 6 hr/d	Hepatic	750 M			Elovaara 1982 meta	
25	Rat NS	24 hr	Resp		75 M (decrease in P-450 and 7-ethoxycoumarin O-deethylase activity)		Elovaara et al. 1987 meta	
26	Rat Sprague-Dawley	4 d 4 hr/d	Resp		1000 F (decreased pulmonary microsomal activity)		Patel et al. 1978 para	
27	Rat Sprague-Dawley	4 hr	Resp		1000 F (decreased pulmonary microsomal activity)		Patel et al. 1978 para	
28	Rat NS	1, 3, or 5 d 6 hr/d	Resp		300 M (transiently decreased surfactant levels)		Silverman and Schatz 1991 para	

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

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (ppm)	LOAEL Less Serious (ppm)	LOAEL Serious (ppm)	Reference Chemical Form	Comments
29	Rat Fischer- 344	1 or 3 d 6 hr/d	Hepatic	1600 M			Simmons et al. 1991 para	
30	Rat Sprague-Dawley	3 d 6 hr/d	Resp		2000 M (decreased cytochrome P-450)		Toftgard and Nilsen 1982 para	
			Renal		2000 M (decreased relative kidney weight)			
31	Rat Sprague-Dawley	3 d 6 hr/d	Resp		2000 M (decreased cytochrome P-450)		Toftgard and Nilsen 1982 meta	
32	Rat Sprague-Dawley	3 d 6 hr/d	Resp		2000 M (decreased cytochrome P-450)		Toftgard and Nilsen 1982 ortho	
			Renal		2000 M (decreased relative kidney weight)			
33	Rat Sprague-Dawley	3 d 6 hr/d	Resp		2000 M (decreased cytochrome P-450)		Toftgard and Nilsen 1982 mixed	
34	Rat CFY	7 d 24 hr/d Gd 7-14	Hepatic	700 F			Ungvary et al. 1980b meta	
			Bd Wt	350 F	700 F (16% decrease in body weight gain)			

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

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (ppm)	LOAEL Less Serious (ppm)	LOAEL Serious (ppm)	Reference Chemical Form	Comments
35	Rat CFY	7 d 24 hr/d Gd 7-14	Hepatic	700 F			Ungvary et al. 1980b	ortho
			Bd Wt	700 F				
36	Rat CFY	7 d 24 hr/d Gd 7-14	Hepatic	700 F			Ungvary et al. 1980b	para
			Bd Wt	700 F				
37	Rat Wistar	9 d 5 hr/d	Hemato	2764			Wronska-Nofer et al. 1991	mixed
38	Mouse Swiss-Webster	1 min	Resp	460 M		1300 M (50% decrease in respiratory rate)	Carpenter et al. 1975a	mixed
39	Mouse Swiss Of1	5 min	Resp			1467 M (50% decrease in respiratory rate)	De Ceaurriz et al. 1981	ortho
40	Mouse	6 min	Resp			2440 M (50% decrease in respiratory rate)	Korsak et al. 1988	mixed
41	Mouse Balb/C	6 min	Resp			2513 M (32% decrease in respiratory rate)	Korsak et al. 1990	ortho

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

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (ppm)	LOAEL Less Serious (ppm)	LOAEL Serious (ppm)	Reference Chemical Form	Comments
42	Mouse Balb/C	6 min	Resp		2626 M (transient 46% decrease in respiratory rate)		Korsak et al. 1990 para	
43	Mouse Balb/C	6 min	Resp		2700 M (transient 57% decrease in respiratory rate)		Korsak et al. 1990 meta	
44	Mouse Balb/c	once 6 min	Resp			1361 M (50% decrease in respiratory rate)	Korsak et al. 1993 meta	
45	Mouse C3H/H3J	4 d 6 hr/d	Hepatic	1208 F			Selgrade et al. 1993 para	
			Bd Wt	1208 F				
46	Rabbit New Zealand	2 d 4 hr/d	Resp		1000 M (decreased pulmonary microsomal activity)		Patel et al. 1978 para	
Neurological								
47	Human	0.25 hr		460	690 (dizziness)		Carpenter et al. 1975a mixed	
48	Human	4 hr			100 M (increased reaction time)		Dudek et al. 1990 mixed	

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

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (ppm)	LOAEL Less Serious (ppm)	LOAEL Serious (ppm)	Reference Chemical Form	Comments
49	Human	2 hr once			50[b] (increased severity scores for headache, dizziness in males; intoxication in males and females)		Ernstgard et al. 2002 meta	
50	Human	2 d 70 min/d		299 M			Gamberale et al. 1978 mixed	
51	Human	1 d 70 min/d			299 M (impairment in reaction time and short-term memory after exercising; not without exercising)		Gamberale et al. 1978 mixed	
52	Human	30 min		396 M			Hastings et al. 1986 mixed	
53	Human	2-6 d 5-5.5 hr/d		200 M			Laine et al. 1993 meta	
54	Human	5 d 1-7.5 hr/d			100 F (dizziness)		NIOSH 1981 para	
55	Human	7 hr		200 M			Ogata et al. 1970 meta	
56	Human	4 hr		69 M			Olson et al. 1985 para	

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

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (ppm)	LOAEL Less Serious (ppm)	LOAEL Serious (ppm)	Reference Chemical Form	Comments
57	Human	2 x/dose 1 x/wk 4 hr/x		281 M			Savolainen 1980 meta	
58	Human	4 hr			400 M (impaired body balance and reaction times)		Savolainen et al. 1984 meta	
59	Human	4 d 3.67 hr/d			200 M (altered visual evoked potentials)		Seppalainen et al. 1989 meta	
60	Rat Sprague-Dawley	3 d 6 hr/d			2000 M (increased brain levels of catecholamine)		Andersson et al. 1981 para	
61	Rat Sprague-Dawley	3 d 6 hr/d			2000 M (increased dopamine and catecholamine in brain)		Andersson et al. 1981 mixed	
62	Rat Sprague-Dawley	3 d 6 hr/d			2000 M (increased brain levels of catecholamine)		Andersson et al. 1981 meta	
63	Rat Sprague-Dawley	3 d 6 hr/d			2000 M (increased brain levels of catecholamine)		Andersson et al. 1981 ortho	

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

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (ppm)	LOAEL Less Serious (ppm)	LOAEL Serious (ppm)	Reference Chemical Form	Comments
64	Rat NS	4 hr		580 M		1300 M (incoordination)	Carpenter et al. 1975a mixed	
65	Rat (Long- Evans)	5 d 8 hr/d				1800 M (18-30 dB increased in mid-range auditory thresholds)	Crofton et al. 1994 mixed	
66	Rat Long- Evans	4 hr		800 M	1600 M (altered visual evoked potentials)		Dyer et al. 1988 para	
67	Rat F344	1 d 3 x/d 2 hr/x			113 M (transiently decreased operant responding)		Ghosh et al. 1987 mixed	
68	Rat F344	5 hr		99 M			Ghosh et al. 1987 mixed	
69	Rat F344	3 d 6 hr/d			114 M (transiently decreased operant responding)		Ghosh et al. 1987 mixed	
70	Rat NS	4 hr		2010 M		2870 M (impaired rotarod performance)	Korsak et al. 1988 mixed	

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

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (ppm)	LOAEL Less Serious (ppm)	LOAEL Serious (ppm)	Reference Chemical Form	Comments
71	Rat	6 hr				3000 M (impaired rotarod performance)	Korsak et al. 1990 ortho	
72	Rat NS	6 hr				3000 M (impaired rotarod performance)	Korsak et al. 1990 para	
73	Rat	6 hr				3000 M (impaired rotarod performance)	Korsak et al. 1990 meta	
74	Rat Wistar Imp:DAK	once 4 hr				1982 M (EC50 for decreased rotarod performance)	Korsak et al. 1993 meta	
75	Rat NS	4 hr				1940 M (narcosis)	Molnar et al. 1986 para	
76	Rat	4 hr				2180 M (narcosis)	Molnar et al. 1986 ortho	
77	Rat	4 hr				2100 M (narcosis)	Molnar et al. 1986 meta	
78	Rat NS	1.5 wk 5 d/wk 6 hr/d			800 M (decreased axonal transport)		Padilla and Lyerly 1989 mixed	

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

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (ppm)	LOAEL Less Serious (ppm)	LOAEL Serious (ppm)	Reference Chemical Form	Comments
79	Rat NS	1, 3, 8, 13 d 5 d/wk 6 hr/d		400 M		800 M (decreased axonal transport)	Padilla and Lyerly 1989 para	
80	Rat NS	4 hr		1700 M			Pryor et al. 1987 mixed	
81	Rat NS	8 hr				1450 M (hearing loss)	Pryor et al. 1987 mixed	
82	Rat (Long- Evans)	5 d 8 hr/d		1700 M		2000 M (50% decreased integrated amplitude of brainstem auditory evoked potentials at 16 kHz)	Rebert et al. 1995 mixed	
83	Rat (albino)	4 hr once			230 M (18% inhibition of electrically evoked seizure discharge)		Vodickova et al. 1995 ortho	
84	Rat F344	2 hr		102 M	192 M (decreased self-stimulation behavior)		Wimolwattanapun et al. 1987 mixed	
85	Mouse (Swiss-Webster)	5 min		250 M	500 M (decreased response rate for schedule-controlled operant behavior)		Bowen et al. 1998 meta	

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

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (ppm)	LOAEL Less Serious (ppm)	LOAEL Serious (ppm)	Reference Chemical Form	Comments
86	Mouse Swiss Of1	4 hr			1010 M (altered behavior in swimming test)		De Ceaurriz et al. 1983 ortho	
87	Mouse (albino)	4 hr once			320 F (11% decreased duration of response to electric shock)		Vodickova et al. 1995 ortho	
88	Cat NS	2 hr				9500 M (salivation, ataxia, seizures, anesthesia)	Carpenter et al. 1975a mixed	
Reproductive								
89	Rat (CFY)	8 d 24 hr/d Gd 7-15				775 (8% decreased fertility; increased resorptions)	Balogh et al. 1982 mixed	
Developmental								
90	Rat (CFY)	8 d 24 hr/d Gd 7-14				775 (postimplantation loss)	Balogh et al. 1982 mixed	
91	Rat (Wistar)	Gd 7-20 6 hr/d				500 F (delayed air righting reflex, impaired motor coordination on Rotarod; impaired memory in Morris water maze)	Hass et al. 1995 mixed	

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

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (ppm)	LOAEL Less Serious (ppm)	LOAEL Serious (ppm)	Reference Chemical Form	Comments
92	Rat (Wistar)	Gd 7-20 6 hr/d				500 F (increased latency in Morris water maze persisting to 28 weeks)	Hass et al. 1997 mixed	
93	Rat Sprague-Dawley	10 d 6 hr/d Gd 7-16		1612 F			Rosen et al. 1986 para	
94	Rat (Sprague-Dawley)	Gd 6-20 6 hr/d		500	1000 (6% decrease in fetal body weight)		Saillenfait et al. 2003 meta	
95	Rat (Sprague-Dawley)	Gd 6-20 6 hr/d		100	500 (5% decrease in fetal body weight)		Saillenfait et al. 2003 ortho	
96	Rat (Sprague-Dawley)	Gd 6-20 6 hr/d		500	1000 (5-6% decrease in fetal body weight)		Saillenfait et al. 2003 para	
97	Rat (Sprague-Dawley)	Gd 6-20 6 hr/d		100	500 (4% decrease in fetal body weight)		Saillenfait et al. 2003 mixed	
98	Rat CFY	9 d 24 hr/d Gd 7-15		438 F	784 F (increased fetal death and resorption)		Ungvary and Tatrai 1985 mixed	

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

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (ppm)	LOAEL Less Serious (ppm)	LOAEL Serious (ppm)	Reference Chemical Form	Comments
99	Rat CFY	8 d Gd 7-14 24 hr/d		35	350 (9% decrease in fetal weight)		Ungvary et al. 1980b ortho	
100	Rat	8 d 24 hr/d Gd 7-14		350 F	700 F (fetal and maternal weight decreased, decreased implantation)		Ungvary et al. 1980b meta	
101	Rat CFY	8 d 24 hr/d Gd 7-14		350 F		700 F (postimplantation loss)	Ungvary et al. 1980b para	
102	Rat CFY	24-48 hr Gd 9 and 10				691 (27% decrease in fetal weight)	Ungvary et al. 1981 para	
INTERMEDIATE EXPOSURE								
Death								
103	Monkey Squirrel	6 wk 5 d/wk 8 hr/d				780 M (1/2 died)	Jenkins et al. 1970 ortho	
104	Rat Sprague-Dawley Long-Evans	6 wk 5 d/wk 8 hr/d				780 (3/12 died)	Jenkins et al. 1970 ortho	

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

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (ppm)	LOAEL Less Serious (ppm)	LOAEL Serious (ppm)	Reference Chemical Form	Comments
Systemic								
105	Human	4 wk 5 d/wk 1-7.5 hr/d (I)	Resp	20	100 M (nose and throat irritation)		NIOSH 1981 para	
			Cardio	150 M				
			Hemato	150 M				
			Renal	150 M				
			Ocular	20 M	100 M (eye irritation)			
106	Rat NS	10 wk 5 d/wk 6 hr/d	Resp	810 M			Carpenter et al. 1975a mixed	
			Cardio	810 M				
			Gastro	810 M				
			Hemato	810 M				
			Musc/skel	810 M				
			Hepatic	810 M				
			Renal	810 M				
			Endocr	810 M				
			Bd Wt	810 M				
107	Rat NS	5, 9, 14, or 18 wk 5 d/wk 6 hr/d	Hepatic	300 M			Elovaara et al. 1980 mixed	

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

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (ppm)	LOAEL Less Serious (ppm)	LOAEL Serious (ppm)	Reference Chemical Form	Comments
108	Rat (Sprague-Dawley)	13 wk 6 d/wk 6 hr/d	Bd Wt	1800 M			Gagnaire et al. 2001 meta	
109	Rat (Sprague-Dawley)	13 wk 6 d/wk 6 hr/d	Bd Wt	1800 M			Gagnaire et al. 2001 ortho	
110	Rat (Sprague-Dawley)	13 wk 6 d/wk 6 hr/d	Bd Wt	1800 M			Gagnaire et al. 2001 para	
111	Rat (Wistar)	3 mo 5 d/wk 6 hr/d	Bd Wt	1000 M			Gralewicz et al. 1995 meta	
112	Rat (Wistar)	5 mo 5 d/wk 5 hr/d	Hepatic	92 M			Jajte et al. 2003 meta	
			Bd Wt	92 M				
113	Rat Sprague-Dawley Long-Evans	90-127 d 24 hr/d	Resp	78			Jenkins et al. 1970 ortho	
			Cardio	78				
			Hemato	78				
			Hepatic	78				
			Renal	78				

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

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (ppm)	LOAEL Less Serious (ppm)	LOAEL Serious (ppm)	Reference Chemical Form	Comments
114	Rat Sprague-Dawley Long-Evans	6 wk 5 d/wk 8 hr/d	Resp	780			Jenkins et al. 1970 ortho	
			Cardio	780				
			Hemato	780				
			Hepatic	780				
			Renal	780				
115	Rat Wistar	3 mo 5 d/wk 6 hr/d	Hemato	1000 M			Korsak et al. 1992 meta	
			Bd Wt	1000 M				
116	Rat (Wistar)	3 mo 5 d/wk 6 hr/d	Hemato	50 M	100 M (19% decreased erythrocytes; 35% increased leukocytes)		Korsak et al. 1994 meta	
			Bd Wt	100 M				
117	Rat CFY	4 wk 5 d/wk 6 hr/d	Cardio			230 M (increased wall thickness in coronary micro-vessels)	Morvai et al. 1987 mixed	
118	Rat Wistar	6 mo 5 d/wk 6 hr/d	Hepatic	100 M			Rydzynski et al. 1992 meta	

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

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (ppm)	LOAEL Less Serious (ppm)	LOAEL Serious (ppm)	Reference Chemical Form	Comments
119	Rat Wistar	3 mo 5 d/wk 6 hr/d	Hepatic	1000 M			Rydzynski et al. 1992 meta	
120	Rat CFY	6 mo 7 d/wk 8 hr/d	Hepatic	1096 M			Tatrai et al. 1981 ortho	
			Bd Wt		1096 M (12% decrease in body weight)			
121	Rat Sprague-Dawley	4 wk 5 d/wk 6 hr/d	Hepatic		600 M (11% increase in relative liver weight)		Toftgard et al. 1981 mixed	
122	Dog	13 wk 5 d/wk 6 hr/d	Resp	810 M			Carpenter et al. 1975a mixed	
			Cardio	810 M				
			Gastro	810 M				
			Hemato	810 M				
			Musc/skel	810 M				
			Hepatic	810 M				
			Renal	810 M				
			Endocr	810 M				
Neurological								
123	Human	4 wk 5 d/wk 1-7.5 hr/d		150 M			NIOSH 1981 para	

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

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (ppm)	LOAEL Less Serious (ppm)	LOAEL Serious (ppm)	Reference Chemical Form	Comments
124	Monkey Squirrel	90-127 d 24 hr/d		78 M			Jenkins et al. 1970 ortho	
125	Monkey Squirrel	6 wk 5 d/wk 8 hr/d		780 M			Jenkins et al. 1970 ortho	
126	Rat (Sprague-Dawley)	13 wk 6 d/wk 6 hr/d		1800 M			Gagnaire et al. 2001 meta	
127	Rat (Sprague-Dawley)	13 wk 6 d/wk 6 hr/d		1800 M			Gagnaire et al. 2001 ortho	
128	Rat (Sprague-Dawley)	13 wk 6 d/wk 6 hr/d		450 M	900 M (loss of cochlear hair cells without functional hearing loss)	1800 M (extensive cochlear hair cell loss; altered auditory evoked potentials; persistent 35-42 dB hearing loss)	Gagnaire et al. 2001 para	
129	Rat (Sprague-Dawley)	13 wk 6 d/wk 6 hr/d		500 M		1000 M (13-19 dB hearing losses in 2-16 kHz frequencies; in all rats, significant loss of outer hair cells of cochlea)	Gagnaire et al. 2006 mixed	20% o-xylene, 20% p-xylene, 40% m-xylene, 20% ethylbenzene.

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

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (ppm)	LOAEL Less Serious (ppm)	LOAEL Serious (ppm)	Reference Chemical Form	Comments
130	Rat (Sprague-Dawley)	13 wk 6 d/wk 6 hr/d		500 M		1000 M (in all rats, significant loss of hair cells in outer rows of organ of Corti)	Gagnaire et al. 2006 mixed	30% o-xylene, 10% p-xylene, 50% m-xylene, 10% ethylbenzene.
131	Rat (Wistar)	4 wk 5 d/wk 6 hr/d			100 M (impaired passive and active avoidance learning)		Gralewicz and Wiaderna 2001 meta	
132	Rat (Wistar)	3 mo 5 d/wk 6 hr/d			100 M (learning deficit in radial arm maze test)		Gralewicz et al. 1995 meta	
133	Rat (Sprague-Dawley)	4 wk 5 d/wk 6 hr/d		80 M			Hillefors-Berglund et al. 1995 mixed	
134	Rat Albino	30 d 24 hr/d			800 M (decreased acetylcholine in striatum, increased glutamine in midbrain, and norepinephrine in hypothalmus)		Honma et al. 1983 mixed	
135	Rat Wistar	3 mo 5 d/wk 6 hr/d			1000 M (decreased rotarod performance and spontaneous motor activity)		Korsak et al. 1992 meta	

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

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (ppm)	LOAEL Less Serious (ppm)	LOAEL Serious (ppm)	Reference Chemical Form	Comments
136	Rat Wistar	6 mo 5 d/wk 6 hr/d			100 M (decreased rotarod performance and spontaneous motor activity)		Korsak et al. 1992 meta	
137	Rat (Wistar)	3 mo 5 d/wk 6 hr/d			50[c] M (decreased latency of paw-lick response)		Korsak et al. 1994 meta	
138	Rat Sprague-Dawley	61 d 7 d/wk 8 hr/d			1009 M (reversible decrease in auditory brainstem response)		Nylen and Hagman 1994 mixed	
139	Rat Fischer- 344	6 wk 7 d/wk 14 hr/d				800 M (hearing loss)	Pryor et al. 1987 mixed	
140	Rat Wistar	18 wk 5 d/wk 6 hr/d			300 M (decreased membrane lipids in axon membranes)		Savolainen and Seppalainen 1979 mixed	
141	Rat Wistar	18 wk 5 d/wk 6 hr/d			300 M (transient decreases in preening behavior)		Savolainen et al. 1979a mixed	
142	Mouse NMRI- BOM	7 wk 5 d/wk 4 hr/d			1600 F (decreased alpha-adrenergic binding in brain)		Rank 1985 meta	

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

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (ppm)	LOAEL Less Serious (ppm)	LOAEL Serious (ppm)	Reference Chemical Form	Comments
143	Dog Beagle	90-127 d 24 hr/d		78 M			Jenkins et al. 1970 ortho	
144	Dog Beagle	6 wk 5 d/wk 8 hr/d				780 M (tremor)	Jenkins et al. 1970 ortho	
145	Gerbil Mongolian	3 mo 30 d/mo 24 hr/d			160 (regional increases in DNA and astro-glial proteins)		Rosengren et al. 1986 mixed	
Reproductive								
146	Rat Sprague-Dawley	61 d 7 d/wk 18 hr/d		1000			Nylen et al. 1989 mixed	
Developmental								
147	Rat CD	166 d 7 d/wk 6 hr/d		250	500 F (7% decrease in fetal weight)		Bio/dynamics 1983 mixed	

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

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (ppm)	LOAEL Less Serious (ppm)	LOAEL Serious (ppm)	Reference Chemical Form	Comments
CHRONIC EXPOSURE								
Systemic								
148	Human	average 7 yr 8 hr/d	Resp		14 M (nose and throat irritation)		Uchida et al. 1993 mixed	
			Gastro		14 M (increased prevalence of nausea and poor appetite)			
			Hemato	14 M				
			Hepatic	14 M				
			Renal	14 M				
			Ocular		14 M (eye irritation)			
149	Rat CFY	1 yr 7 d/wk 8 hr/d	Hepatic	1096 M			Tatrai et al. 1981 ortho	
			Bd Wt		1096 M (12% decrease in body weight)			

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

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (ppm)	LOAEL Less Serious (ppm)	LOAEL Serious (ppm)	Reference Chemical Form	Comments
Neurological								
150	Human	average 7 yr 8 hr/d			14[d] (increased prevalence of anxiety, forgetfulness, inability to concentrate and other subjective symptoms)		Uchida et al. 1993 mixed	

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

b Used to derive an acute-duration minimal risk level (MRL) for mixed xylenes based on a minimal LOAEL of 50 ppm for m-xylene in humans; concentration divided by an uncertainty factor of 30 (3 for use of a minimal LOAEL and 10 for human variability).

c Used to derive an intermediate-duration minimal risk level (MRL) for mixed xylenes based on a minimal LOAEL of 50 ppm for m-xylene in rats; this LOAEL was converted to a human equivalent concentration using a dosimetric adjustment (EPA 1994). The human equivalent LOAEL of 50 ppm was divided by an uncertainty factor of 90 (3 for use of a minimal LOAEL, 3 for extrapolation from animals to humans with dosimetric adjustment, and 10 for human variability).

d Used to derive a chronic-duration minimal risk level (MRL) for mixed xylenes based on a LOAEL of 14 ppm (geometric mean) for mixed xylenes in humans; concentration divided by an uncertainty factor of 100 (10 for use of a LOAEL and 10 for human variability) and a modifying factor of 3 to account for the lack of supporting studies evaluating the chronic neurotoxicity of xylene.

Bd Wt = body weight; Cardio = cardiovascular; d = day(s); dB = decibel; EC50 = effective concentration; Endocr = endocrine; F = Female; Gastro = gastrointestinal; Gd = gestational day,50%; hemato = hematological; hr = hour(s); KHz = kilohertz; LC50 = lethal concentration, 50%; LOAEL = lowest-observed-adverse-effect level; M = male; min = minute(s); mo = month(s); Musc/skel = musculoskeletal; NOAEL = no-observed-adverse-effect level; Resp = respiratory; x = time(s); wk = week(s); yr = year(s)

Figure 3-1 Levels of Significant Exposure to Xylene - Inhalation

Acute (≤14 days)

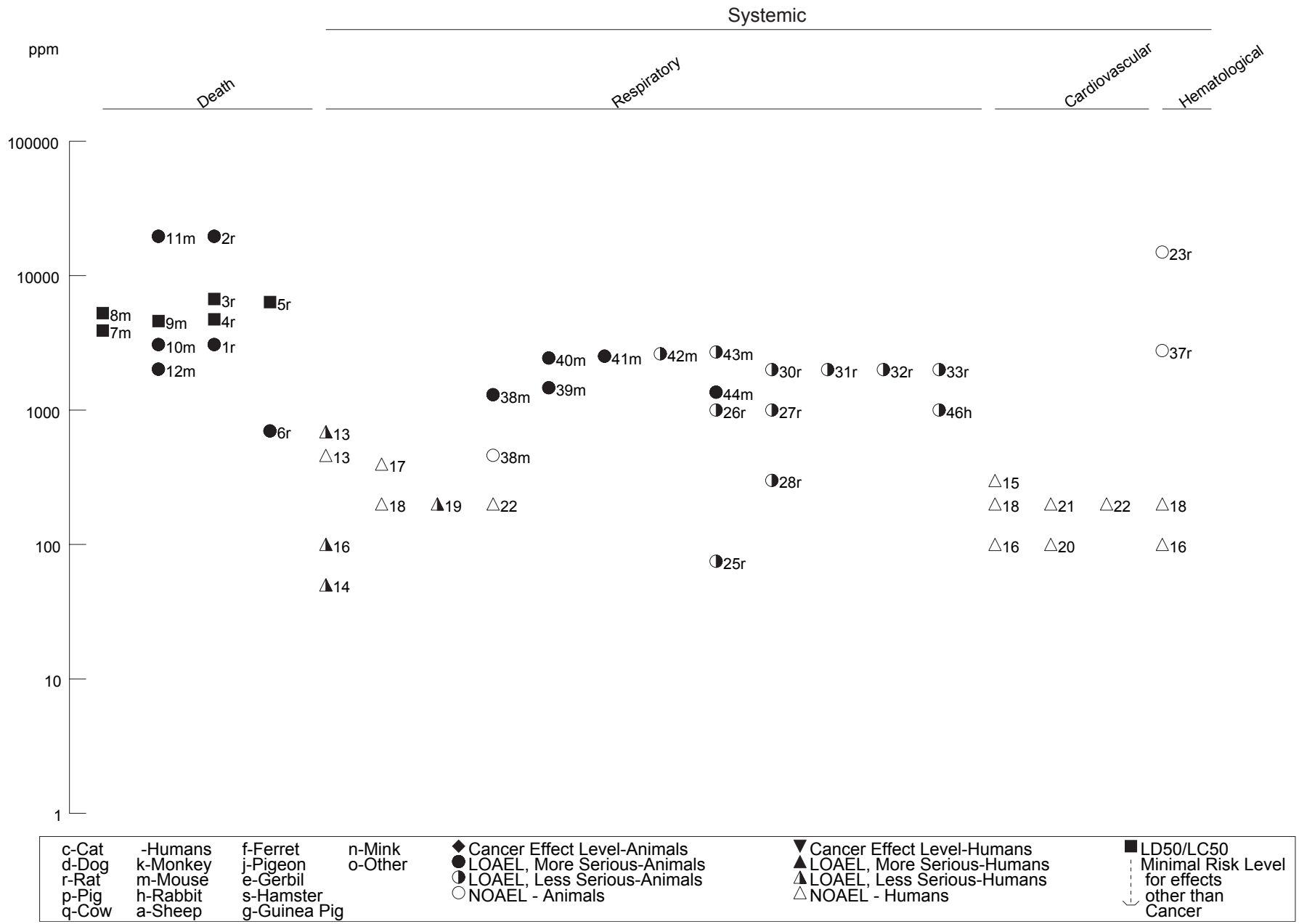


Figure 3-1 Levels of Significant Exposure to Xylene - Inhalation *(Continued)*

Acute (≤14 days)

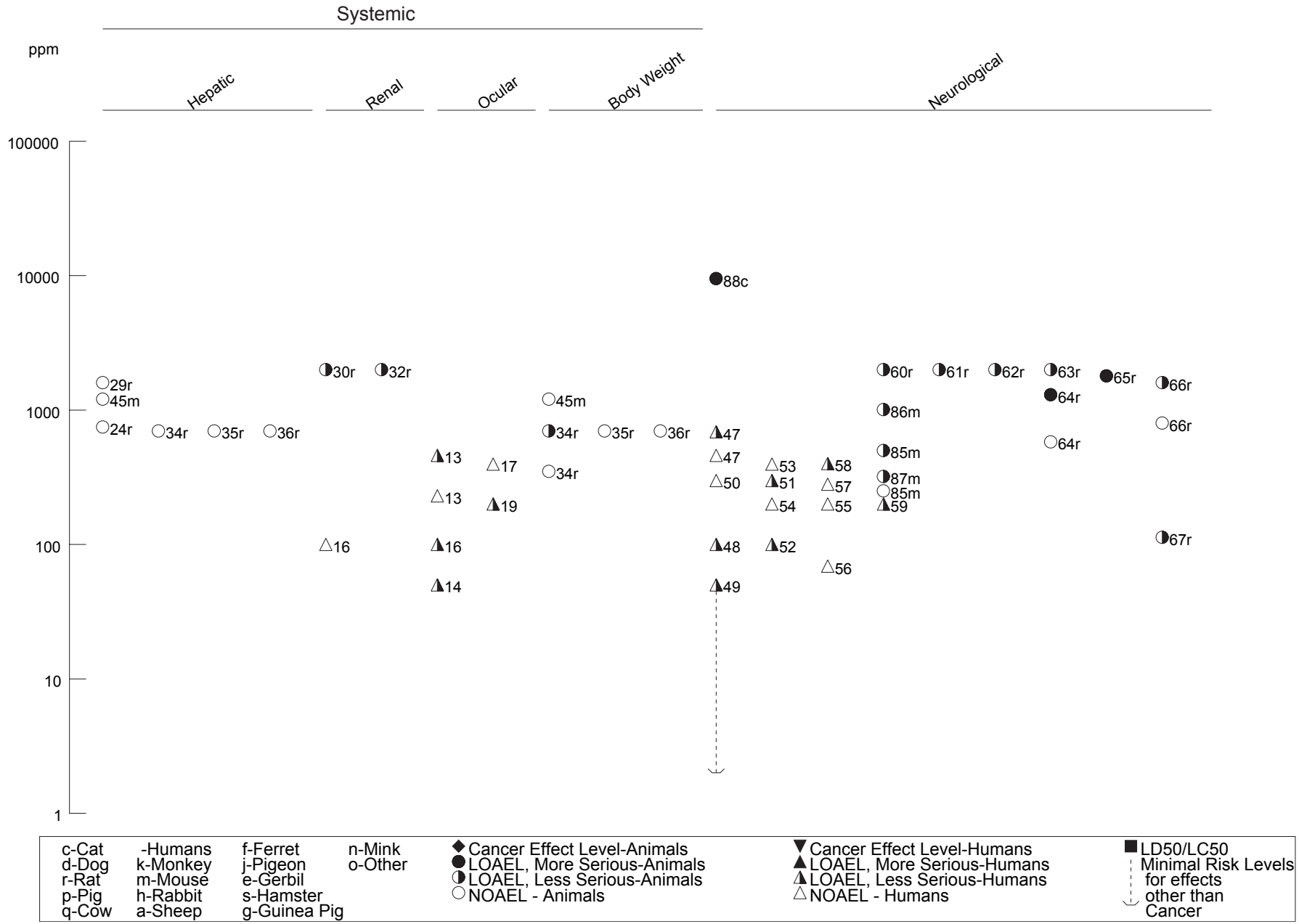


Figure 3-1 Levels of Significant Exposure to Xylene - Inhalation *(Continued)*

Acute (≤14 days)

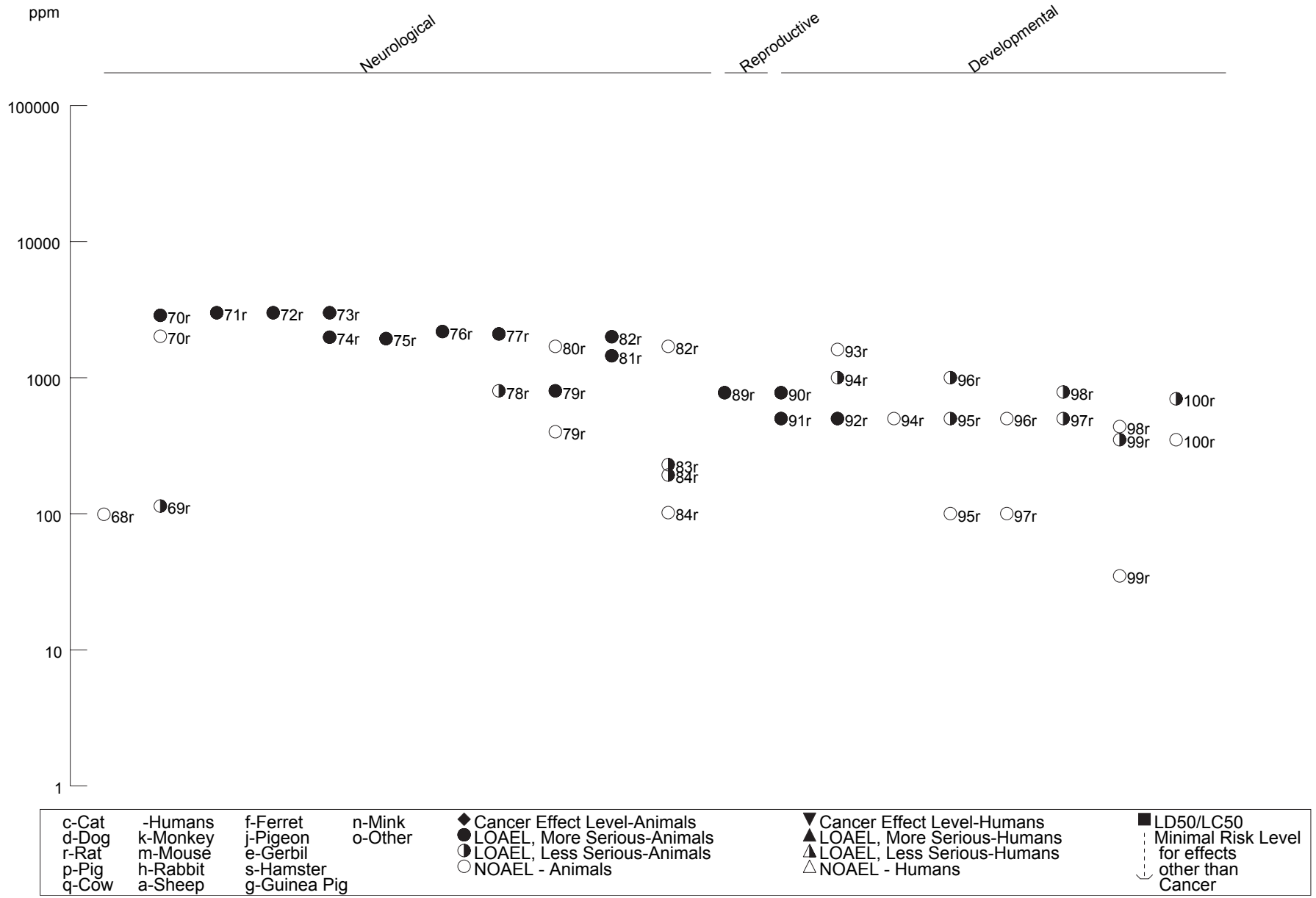


Figure 3-1 Levels of Significant Exposure to Xylene - Inhalation *(Continued)*

Acute (≤14 days)

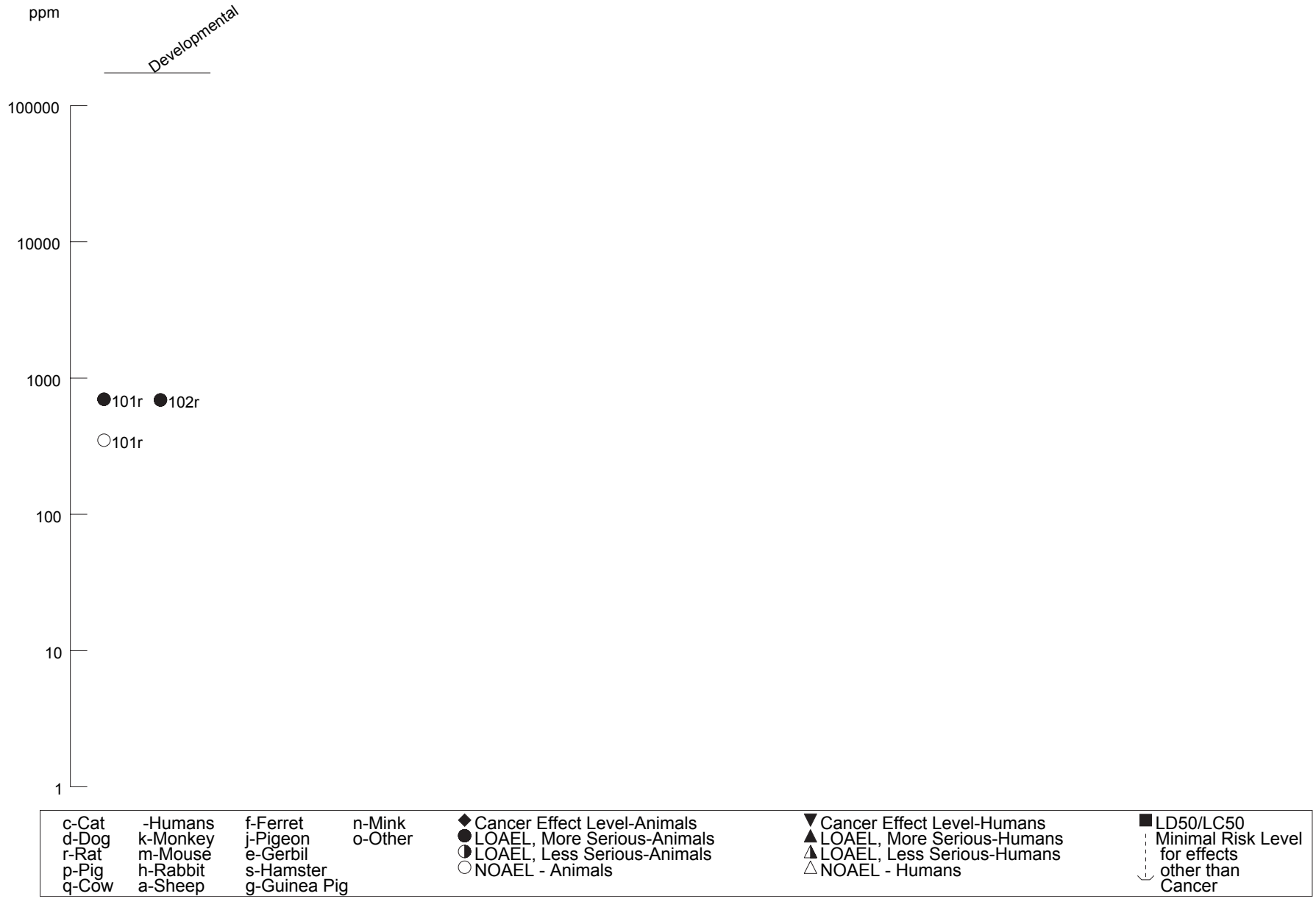


Figure 3-1 Levels of Significant Exposure to Xylene - Inhalation *(Continued)*

Intermediate (15-364 days)

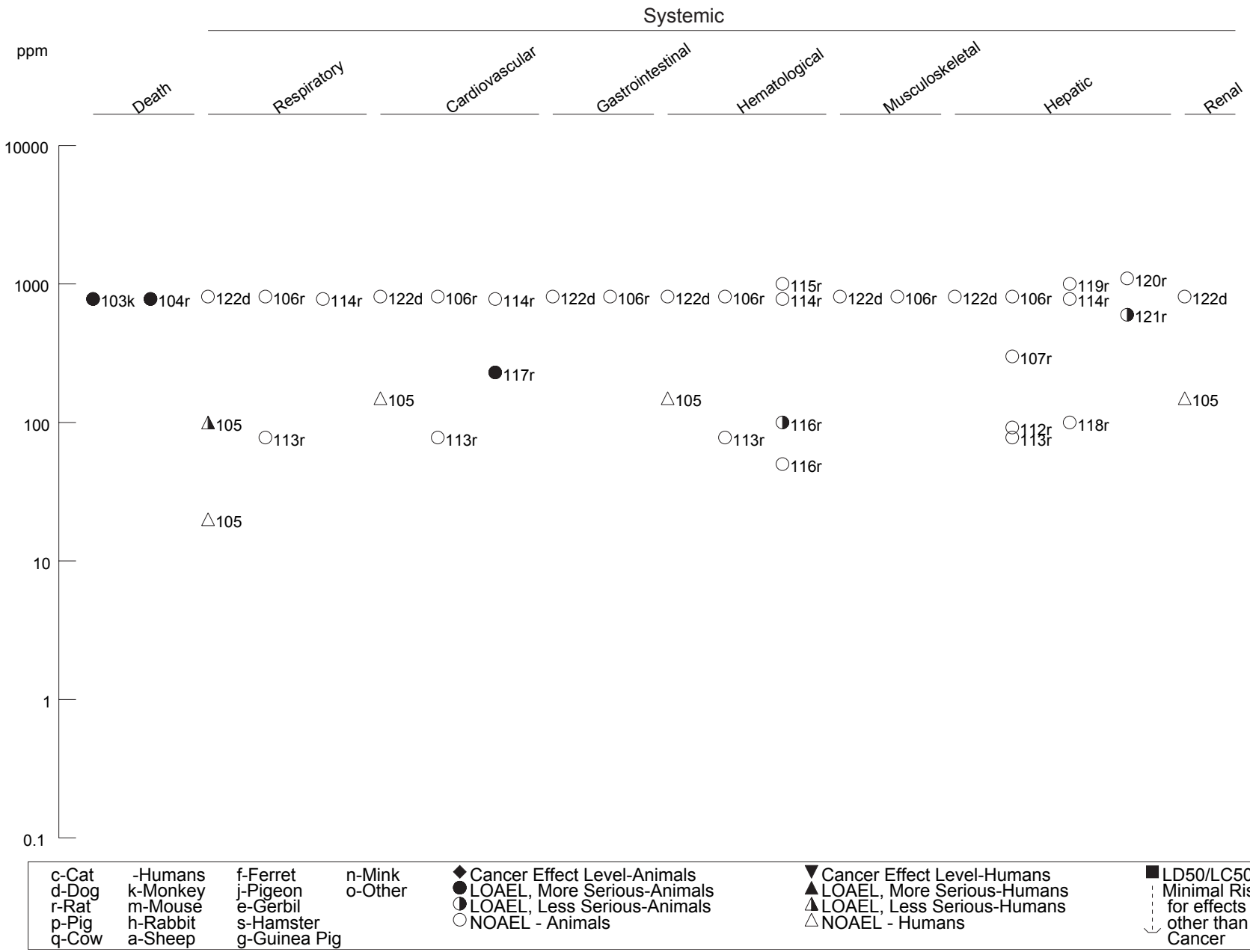


Figure 3-1 Levels of Significant Exposure to Xylene - Inhalation *(Continued)*

Intermediate (15-364 days)

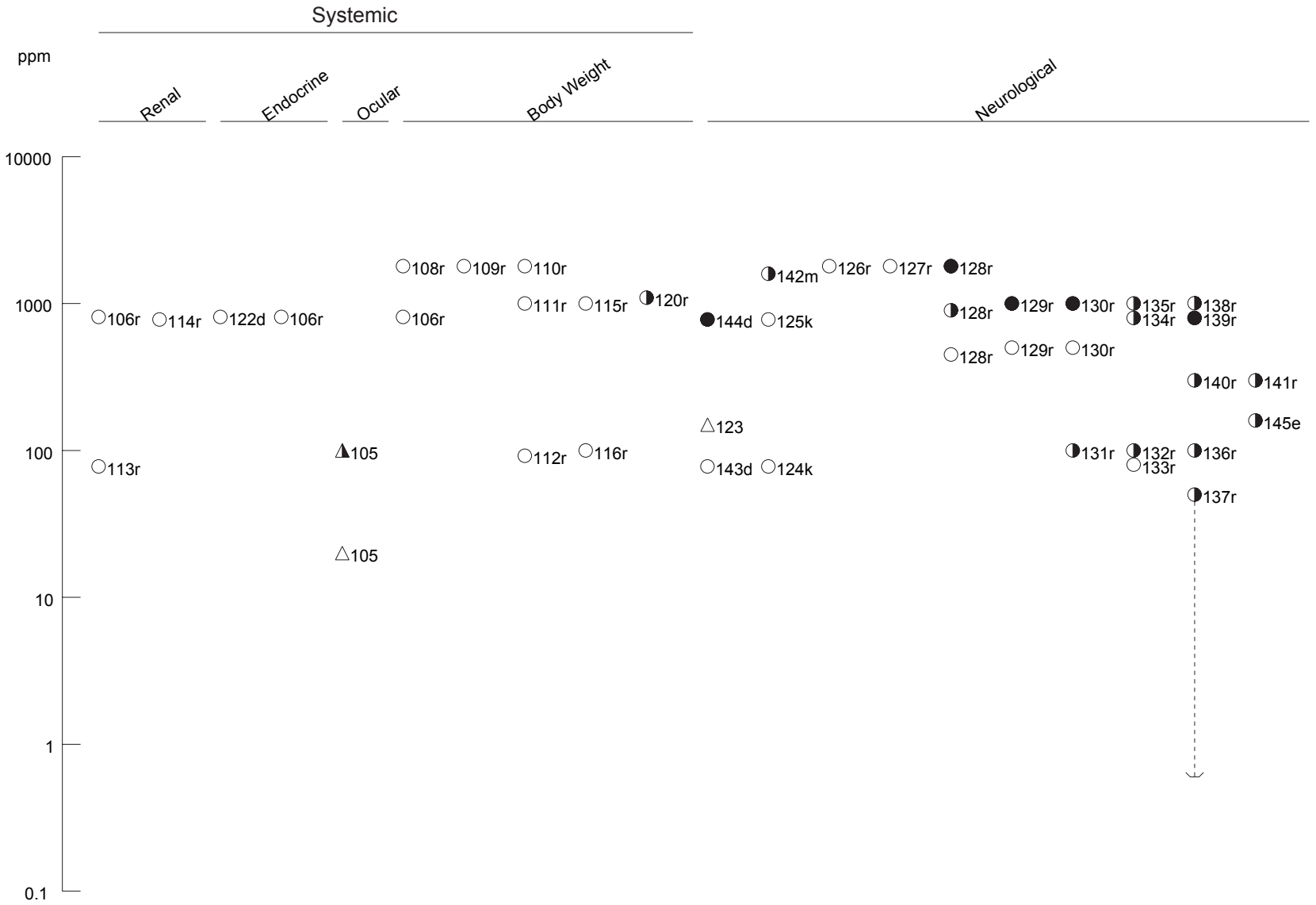


Figure 3-1 Levels of Significant Exposure to Xylene - Inhalation *(Continued)*

Intermediate (15-364 days)

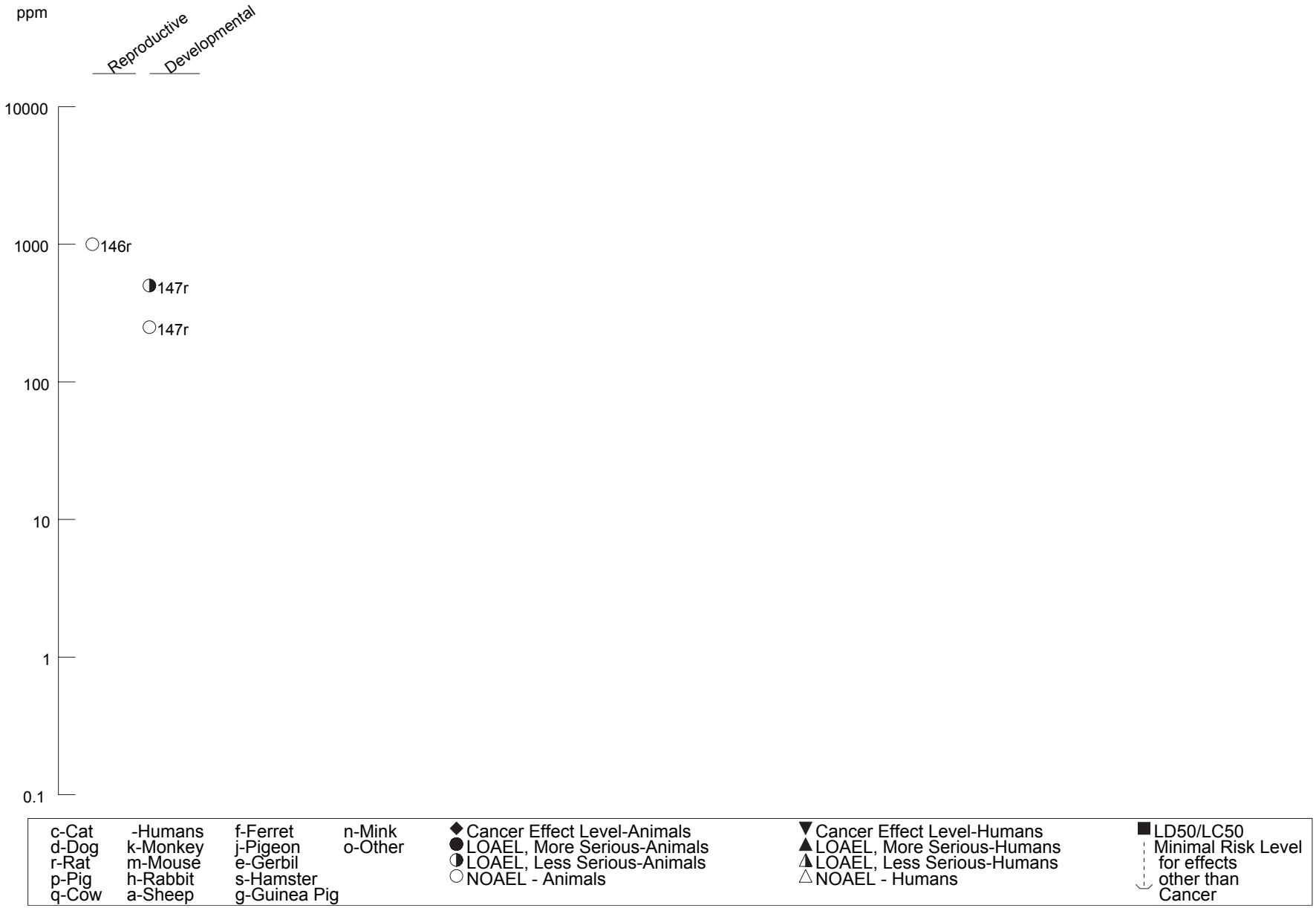
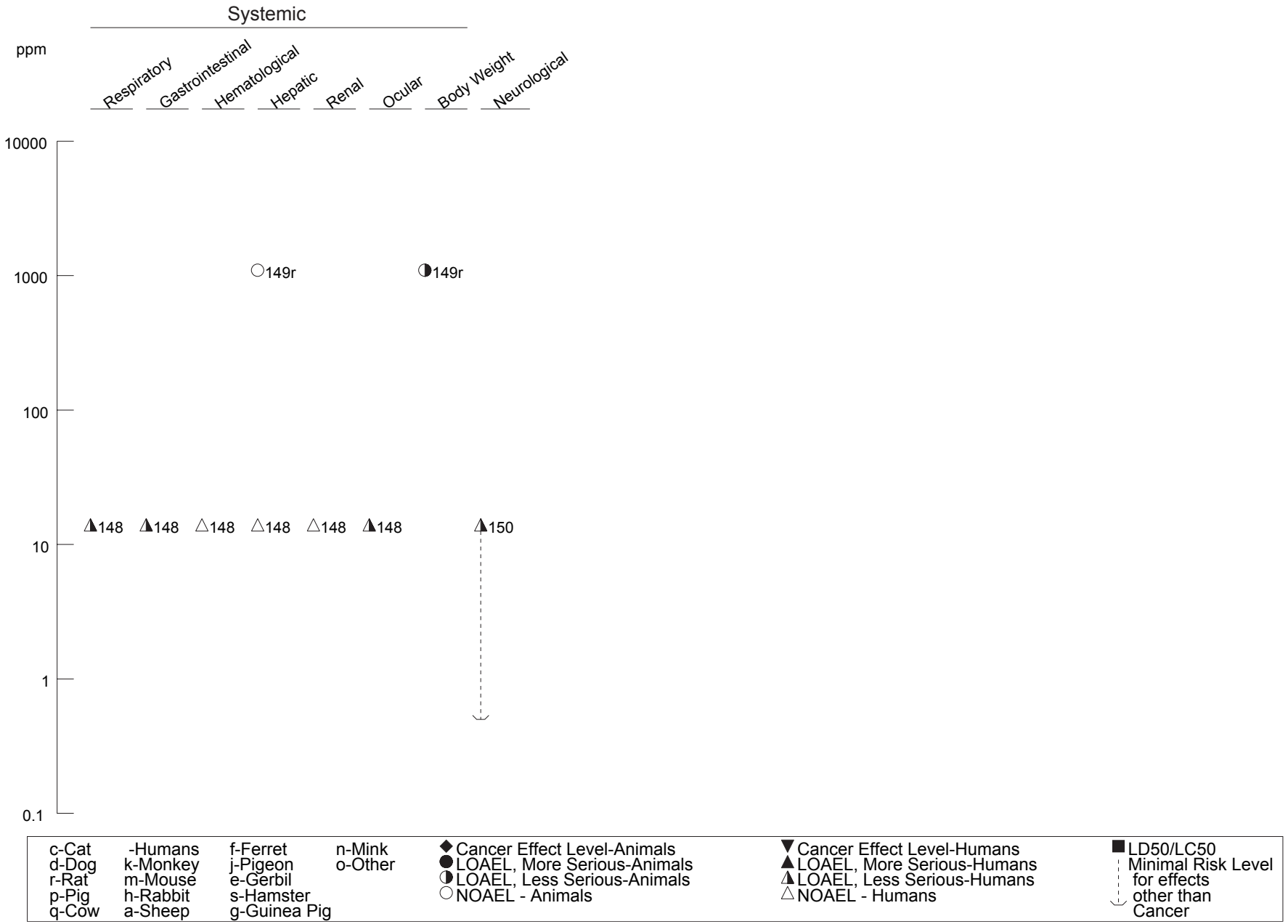


Figure 3-1 Levels of Significant Exposure to Xylene - Inhalation *(Continued)*

Chronic (≥365 days)



30 minutes (Hastings et al. 1986). Slight, but statistically significant increases in the average rating for subjective symptoms of respiratory effects were observed following exposure to *m*-xylene at 50 ppm (Ernstgard et al. 2002) for discomfort in the nose in both sexes after 60 and 118 minutes, in discomfort in the throat or airways in women after 60 minutes, and in breathing difficulty in men at 118 minutes and women at both timepoints. Small but statistically significant changes in objective tests of pulmonary function were reported in women, but not men, measured 3 hours after the end of the 2-hour exposure: decreased forced vital capacity (FVC), increased forced expiratory flow at 75% FVC (FEF_{75}), and increased ratio of forced expiratory volume in 1 minute (FEV_1) to forced vital capacity (FEV_1/FVC). This study is the basis for the acute-duration inhalation MRL for which respiratory and neurologic toxicity are the critical effects. Chest x-rays obtained from volunteers exposed to a time-weighted-average (TWA) concentration of 200 ppm *m*-xylene for 3.67 hours/day for 4 days showed no adverse effects on the lungs (Seppalainen et al. 1989). Also, no effects on pulmonary ventilation volume were observed in volunteers exposed to 150 ppm *p*-xylene for 5 days/week in a 4-week trial (NIOSH 1981).

At much higher concentrations, however, the lung may be adversely affected. An autopsy revealed that exposure to an estimated 10,000 ppm of xylene produced severe lung congestion with focal intra-alveolar hemorrhage and pulmonary edema in one worker who died following exposure to xylene fumes for several hours while painting (Morley et al. 1970). Another worker exposed in the same incident exhibited patchy diffuse opacities in radiograms and moist rales in both lungs; a third exposed worker showed no evidence of lung effects. Case reports indicate that acute-duration inhalation exposure to mixed xylene and *p*-xylene has been associated with irritation of the nose and throat (Carpenter et al. 1975a; Klaucke et al. 1982; Nelson et al. 1943; Nersesian et al. 1985; NIOSH 1981). A worker at a chemical company who was exposed to heated xylene from a pressurized hose experienced throat pain and dyspnea (Narvaez and Song 2003).

Chronic occupational exposure of workers to an unspecified concentration of vapors of mixed xylene has also been associated with labored breathing and impaired pulmonary function (Hipolito 1980; Roberts et al. 1988). A significant ($p<0.01$) increase in the prevalence of nose and throat irritation was reported by workers chronically exposed to mixed xylene vapors at a geometric mean TWA concentration of 14 ppm (Uchida et al. 1993). This study is the basis for the chronic-duration inhalation MRL for which respiratory and neurological toxicity are the critical effects.

Adverse respiratory effects noted in rats, mice, and guinea pigs following acute and intermediate inhalation exposure to xylene are similar to those observed in humans. They include decreased

respiration, labored breathing, irritation of the respiratory tract, pulmonary edema, pulmonary hemorrhage, and pulmonary inflammation (Carpenter et al. 1975a; De Ceaurriz et al. 1981; Furnas and Hine 1958; Korsak et al. 1990). Exposure to concentrations of 2,440 ppm mixed xylene for 6 minutes (Korsak et al. 1988) to 1,467 ppm *o*-xylene for 5 minutes (De Ceaurriz et al. 1981), or to 1,361 ppm *m*-xylene for 6 minutes (Korsak et al. 1993) produced a 50% decrease in respiratory rate in mice. Comparison of the individual xylene isomers showed that the irritant effects of *m*- and *o*-xylene as quantified by measurements of respiratory rate in mice are more pronounced than those of *p*-xylene, with *o*-xylene having the most prolonged effect (Korsak et al. 1990). In rats that died as a result of exposure to 9,900 ppm mixed xylene for 4 hours, atelectasis, hemorrhage, and edema of the lungs were observed (Carpenter et al. 1975a). Biochemical changes detected in the lungs after acute-duration intermittent exposure include transiently decreased lung surfactant levels at 300 ppm *p*-xylene (Silverman and Schatz 1991) and decreased pulmonary microsomal enzyme activities at 2,000 ppm mixed xylene, 75–2,000 ppm *m*-xylene, 2,000 ppm *o*-xylene, or 1,000 or 3,400 ppm *p*-xylene (Day et al. 1992; Elovaara et al. 1980, 1987; Patel et al. 1978; Silverman and Schatz 1991; Toftgard and Nilsen 1982). The LOAEL of 75 ppm for *m*-xylene was based on decreased P-450 and 7-ethoxycoumarin *O*-deethylase activities noted in the lungs of rats exposed for 24 hours (Elovaara et al. 1987). The decrease in pulmonary microsomal activity by selective inactivation of enzymes can result from damage to lung tissue caused by the toxic metabolite of xylene, a methylbenzaldehyde (Carlone and Fouts 1974; Patel et al. 1978; Smith et al. 1982); the selective inactivation of enzymes may also result in anoxia.

No effect on absolute or relative lung weights was observed in male rats intermittently exposed to *m*-xylene at concentrations as high as 100 ppm for 13 weeks (Korsak et al. 1994). No histopathological changes in the lungs were evident in rats, dogs, guinea pigs, or monkeys following intermediate exposure for 90–127 days to concentrations of 78 ppm *o*-xylene on a continuous basis (Jenkins et al. 1970) or 13 weeks to 810 ppm mixed or 6 weeks to 780 ppm *o*-xylene, 5 weeks to 300 ppm *m*-xylene, or for 5 days to 300 ppm *p*-xylene on an intermittent basis (Carpenter et al. 1975a; Elovaara et al. 1987; Jenkins et al. 1970; Silverman and Schatz 1991).

No animal studies were located that evaluated the respiratory effects of mixed xylene or single xylene isomers following chronic inhalation exposure.

An acute-duration inhalation MRL of 2 ppm was calculated for mixed xylenes based on a LOAEL for neurological and respiratory effects in human subjects exposed to 50 ppm *m*-xylene for 2 hours (Ernstgard et al. 2002; see footnote in Table 3-1). A chronic-duration inhalation MRL of 0.05 ppm was

calculated for mixed xylenes based on a LOAEL of 14 ppm for subjective neurological and respiratory symptoms in workers exposed to mixed xylene 8 hours/day, 5 days/week for an average of 7 years (Uchida et al. 1993; see footnote in Table 3-1).

Cardiovascular Effects. Limited human data are available regarding the cardiovascular effects of xylene following inhalation exposure. Although tachycardia was reported by one of nine persons exposed to unidentified levels of xylene as a result of its use in a sealant in a heating duct, no effects on heart rate, blood pressure, or cardiac function were noted in humans exposed to ≤299 ppm mixed xylene for an acute duration (70 minutes to 7 hours) (Gamberale et al. 1978), 200 ppm *m*-xylene (Ogata et al. 1970; Seppalainen et al. 1989), or 150 ppm *p*-xylene (NIOSH 1981; Ogata et al. 1970). Furthermore, two survivors exposed to an estimated 10,000 ppm xylene in an industrial accident had normal pulse, blood pressure, and heart sounds upon hospitalization. Chronic occupational exposure to xylene along with other chemical agents has resulted in complaints of heart palpitations, chest pain, and an abnormal electrocardiogram (ECG) (Hipolito 1980; Kilburn et al. 1985). However, the contribution of other chemical exposures to these effects cannot be eliminated.

Data regarding cardiovascular effects in animals are limited. Morphological changes in coronary microvessels (increased wall thickness) were noted in rats exposed to 230 ppm xylene (unspecified composition) for 4 weeks (Morvai et al. 1987). Other effects seen in rats inhaling unspecified (lethal) concentrations of xylene of unknown composition included ventricular repolarization disturbances and occasional arrhythmias; the toxicity of unknown components was not reported (Morvai et al. 1976). However, no adverse effects on the heart were observed upon histopathological examination of rats and dogs exposed intermittently for 10–13 weeks to mixed xylene at concentrations as high as 810 ppm (Carpenter et al. 1975a) or rats, guinea pigs, dogs, or monkeys exposed to *o*-xylene at 78 ppm on a continuous basis for 90–127 days or 780 ppm on an intermittent basis for 6 weeks (Jenkins et al. 1970). No effect on absolute or relative heart weights was observed in male rats intermittently exposed to *m*-xylene at concentrations as high as 100 ppm for 13 weeks (Korsak et al. 1994). No information was located regarding cardiovascular effects in animals after chronic exposure to mixed xylene or its individual isomers.

Gastrointestinal Effects. Symptoms of nausea, vomiting, and gastric discomfort have been noted in workers exposed to xylene vapors (concentration unspecified) (Goldie 1960; Hipolito 1980; Klaucke et al. 1982; Nersesian et al. 1985; Uchida et al. 1993). These symptoms subsided after cessation of the xylene exposure. Anorexia and vomiting were also observed in a patient admitted to the hospital after sniffing

paint containing xylene and other unknown substances over a 2-week period in an effort to become intoxicated (Martinez et al. 1989) and nausea was more frequently reported in males acutely exposed to m-xylene for 2 hours, as compared to controls (Ernstgard et al. 2002).

Limited data were located regarding gastrointestinal effects in animals. No lesions were observed in the gastrointestinal tract of rats and dogs exposed to concentrations as high as 810 ppm mixed xylene for 13 weeks (Carpenter et al. 1975a). No studies were located regarding gastrointestinal effects in animals after acute or chronic inhalation exposure to mixed xylene or the isomers of xylene.

Hematological Effects. Human data are limited regarding the effects of xylene on the blood. Female volunteers had normal blood counts after exposure to 100 ppm *p*-xylene for 1–7.5 hours/day for 5 days (NIOSH 1981). Hemoglobin content of the blood was unaffected in two workers exposed to an estimated 10,000 ppm of mixed xylene in an industrial accident (Morley et al. 1970). Decreased white blood cell counts were observed in two women with chronic occupational exposure to xylene (Hipolito 1980; Moszczynski and Lisiewicz 1983, 1984a), but exposure to other chemicals cannot be ruled out as an alternative explanation for the effects observed.

Previously, chronic occupational exposure to xylene by inhalation was thought to be associated with a variety of hematological effects (NIOSH 1975). However, exposure in all cases was to solvent mixtures known or suspected to contain benzene as well. Because benzene is an agent known to cause leukemia and other blood dyscrasias in humans (Agency for Toxic Substances and Disease Registry 2005), these effects cannot be solely attributed to xylene.

An occupational study in which no benzene exposure was involved (Uchida et al. 1993) found no hematological effects (red blood cell, white blood cell and platelet counts, and hemoglobin concentrations were unchanged). Workers (175) were exposed to a geometric mean TWA of 14 ppm xylene for an average of 7 years, and mixed xylene exposure accounted for 70% or more of the total exposure (Uchida et al. 1993). This study suggests that occupational exposure to relatively low concentrations of xylenes does not cause hematological effects.

No effect on erythrocyte fragility was observed in rats exposed to 15,000 ppm mixed xylene for 45 minutes (Carpenter et al. 1975a). No adverse hematological effects have been observed in rats exposed to 2,764 ppm mixed xylene for 5 hours/day for 9 days (Wronska-Nofer et al. 1991). In rats intermittently exposed to 100 ppm *m*-xylene for 90 days, erythrocyte counts were reduced by 18.5% and

leukocyte counts were increased by 35% (Korsak et al. 1994). Increases in leukocyte count were reported in rats and dogs exposed intermittently to 780 ppm *o*-xylene for 6 weeks (Jenkins et al. 1970), but it is unknown whether these increases were statistically significant. However, no effects on hematological parameters were observed in rats or dogs following intermediate-duration intermittent exposure to concentrations as high as 810 ppm of mixed xylene (Carpenter et al. 1975a) or in guinea pigs exposed to 78 ppm *o*-xylene continuously or 780 ppm *o*-xylene intermittently (Jenkins et al. 1970) for an intermediate duration.

Musculoskeletal Effects. A 1993 occupational study indicates that workers exposed to xylenes (geometric mean TWA 14 ppm) reported reduced grasping power and reduced muscle power in the extremities more frequently than the unexposed controls (Uchida et al. 1993). This effect was a neurological effect rather than a direct effect on the muscles. No additional data were available regarding musculoskeletal effects in humans following inhalation exposure to mixed xylene or its individual isomers. Animal data regarding musculoskeletal effects following xylene inhalation are limited but provide no indication that xylene produces musculoskeletal effects. No lesions were observed in the skeletal muscle of rats and dogs exposed for an intermediate exposure to concentrations as high as 810 ppm mixed xylene (Carpenter et al. 1975a).

Hepatic Effects. Human data regarding hepatic effects following inhalation of xylene are limited to several case and occupational studies (Klaucke et al. 1982; Morley et al. 1970; Uchida et al. 1993); other occupational studies involve exposure to other compounds such as toluene (Dolara et al. 1982; Kurppa and Husman 1982). Two of these studies suggest that acute-duration exposure to high levels of xylene may result in hepatic toxicity. Two painters who survived exposure to an estimated 10,000 ppm of xylene and several workers who were exposed to an estimated 700 ppm of xylene had transiently elevated serum transaminase levels (Klaucke et al. 1982; Morley et al. 1970). The one painter who died had hepato-cellular vacuolation following exposure to xylene for 18.5 hours. An occupational study in which workers were exposed an average of 7 years to >70% mixed xylenes (geometric mean TWA 14 ppm) found no changes in serum biochemistry values that reflect liver function (total bilirubin, aspartate aminotransferase, alanine aminotransferase, gamma glutamyl transpeptidase, alkaline phosphatase, and leucine aminopeptidase) (Uchida et al. 1993). This study suggests that low-level occupational exposure to xylenes does not result in hepatic effects.

Animal studies using rats indicate that mixed xylene, *m*-xylene, *o*-xylene, or *p*-xylene generally induce a wide variety of hepatic enzymes, as well as increased hepatic cytochrome P-450 content in rats (Elovaara

1982; Elovaara et al. 1980; Patel et al. 1979; Savolainen et al. 1978; Selgrade et al. 1993; Toftgard and Nilsen 1981, 1982; Toftgard et al. 1981; Ungvary et al. 1980a). Following acute exposures to mixed xylene (Savolainen et al. 1978; Ungvary 1990; Wisniewska-Knypl et al. 1989), *m*-xylene (Elovaara 1982; Ungvary et al. 1980b), *o*-xylene (Tatrai and Ungvary 1980; Ungvary et al. 1980a), or *p*-xylene (Patel et al. 1979; Simmons et al. 1991; Ungvary et al. 1980b), effects have been observed including increased relative liver weight (Simmons et al. 1991; Tatrai and Ungvary 1980; Ungvary et al. 1980a, 1980b), cytochrome P-450 content (Simmons et al. 1991; Ungvary 1990; Ungvary et al. 1980a; Wisniewska-Knypl et al. 1989), microsomal protein (Elovaara 1982), microsomal enzyme activity (Elovaara 1982; Savolainen et al. 1978; Ungvary 1990; Ungvary et al. 1980a; Wisniewska-Knypl et al. 1989), proliferation of the endoplasmic reticulum (Ungvary 1990; Wisniewska-Knypl et al. 1989), and decreased hexobarbital sleep time (Ungvary 1990; Ungvary et al. 1980a). Similar changes were observed in rabbits and mice (Ungvary 1990). Although histopathological examination of livers in most studies showed no adverse effects (Elovaara 1982; Simmons et al. 1991; Ungvary et al. 1980b), minor histopathological changes suggesting mild hepatic toxicity included decreased glycogen content, dilation of the cisterns of the rough endoplasmic reticulum, separation of ribosomes from the membranes, variously shaped mitochondria, and increased autophagous bodies (Tatrai and Ungvary 1980; Ungvary 1990). Also, increased serum transaminases were observed following a 4-hour exposure of rats to 1,000 ppm *p*-xylene (Patel et al. 1979).

Many similar hepatic effects appear after intermediate-duration exposure to mixed xylene or *o*-xylene. They include increased absolute and/or relative hepatic weight in rats (Kyrklund et al. 1987; Tatrai and Ungvary 1980; Tatrai et al. 1981; Toftgard et al. 1981; Ungvary 1990; Ungvary et al. 1980a), increased cytochrome P-450 (Tatrai et al. 1981; Ungvary 1990; Ungvary et al. 1980a); increased microsomal enzyme activity (Elovaara et al. 1980, 1987; Tatrai et al. 1981; Toftgard et al. 1981; Ungvary 1990; Ungvary et al. 1980a), proliferation of the smooth and rough endoplasmic reticulum (Rydzynski et al. 1992; Tatrai and Ungvary 1980; Tatrai et al. 1981; Ungvary 1990) and decreased hexobarbital sleeping time because of enhanced metabolism of the drug (Tatrai et al. 1981; Ungvary 1990; Ungvary et al. 1980a). Similar effects were observed in rabbits and mice (Ungvary 1990). As in the acute studies, several intermediate-duration studies in rats, guinea pigs, monkeys, or dogs, reported no effect on serum transaminases (Carpenter et al. 1975a; Tatrai et al. 1981) or hepatic morphology (Carpenter et al. 1975a; Jenkins et al. 1970). Ultrastructural examination of livers showed only minor changes: decreased hepatic glycogen in rats (Tatrai and Ungvary 1980; Ungvary 1990; Ungvary et al. 1980b), ultrastructural changes in hepatic rough endoplasmic reticulum and mitochondria in rats (Tatrai and Ungvary 1980; Ungvary 1990), increased autophagous bodies (Tatrai et al. 1981; Ungvary 1990), and changes in the distribution

of hepatocellular nuclei in rats (Tatrai and Ungvary 1980). Some authors have characterized the hepatic changes as adaptive rather than adverse (Tatrai and Ungvary 1980; Ungvary 1990). No effects on hepatic microsomal proteins, cytochrome P-450, lipid peroxidation (as indicated by levels of malondialdehyde), triglycerides, serum enzymes (AST, ALT, SDH), or absolute or relative liver weights were observed in rats exposed to *m*-xylene at concentrations as high as 100 ppm for 13 weeks (Korsak et al. 1994). No changes in the levels of lipid peroxidation (malondialdehyde levels), glutathione, or glutathione-S-transferase activity were observed in rats intermittently exposed to 92 ppm *m*-xylene for 5 months (Jajte et al. 2003).

Increased liver weight and microsomal enzyme activity were reported in a study in which rats were exposed to 1,096 ppm *o*-xylene for 1 year (Tatrai et al. 1981). Electron microscopic examination of liver revealed a proliferation of the endoplasmic reticulum and only very minor effects on mitochondria as exemplified by increased numbers of peroxisomes.

Renal Effects. Although urinalyses (using a dip-stick technique) of volunteers exposed to *p*-xylene at 100 ppm for 5 days or up to 150 ppm in a multi-week exposure paradigm showed no adverse effects on the kidneys (NIOSH 1981), limited data from case reports and occupational studies suggest that inhalation exposure to solvent mixtures containing xylene may be associated with adverse renal effects in humans (Martinez et al. 1989; Morley et al. 1970). These effects included increased blood urea (Morley et al. 1970), distal renal tubular acidemia (Martinez et al. 1989), and decreased urinary clearance of endogenous creatinine (Morley et al. 1970). Other studies that reported increased urinary levels of β-glucuronidase (Franchini et al. 1983), or increased urinary excretion of albumin, erythrocytes, and leukocytes (Askergren 1981, 1982) are confounded by concurrent exposure to substantial amounts of toluene, a known renal toxicant.

In an occupational study in which the exposure was predominantly to mixed xylenes (geometric mean TWA 14 ppm) for an average of 7 years (Uchida et al. 1993), no effects on measures of kidney function (serum creatinine or urinalysis for urobilinogen, sugar, protein, and occult bleeding) were noted. This study suggests that low-level occupational exposure to xylenes does not result in kidney effects.

The renal effects of mixed xylene and *o*-xylene following inhalation exposure have been evaluated in acute and intermediate studies with rats, guinea pigs, dogs, and monkeys (Carpenter et al. 1975a; Elovaara 1982; Jenkins et al. 1970; Toftgard and Nilsen 1982). Effects noted in these studies at xylene concentrations of 50–2,000 ppm have included increased renal enzyme activity, increased renal

cytochrome P-450 content, and increased kidney-to-body weight ratios (*o*-xylene-exposed rats) (Elovaara 1982; Toftgard and Nilsen 1982). However, histopathologic examination of rats, guinea pigs, dogs, and monkeys did not reveal any renal lesions after inhalation of 810 ppm mixed xylene or 78 ppm *o*-xylene for an intermediate period of 13 weeks and 90–127 days, respectively (Carpenter et al. 1975a; Jenkins et al. 1970). No effect on absolute or relative kidney weights was observed in male rats intermittently exposed to *m*-xylene at concentrations as high as 100 ppm for 13 weeks (Korsak et al. 1994).

No studies were located regarding renal effects following chronic inhalation exposure to mixed xylene or its isomers.

Endocrine Effects. No human data were available regarding endocrine effects following inhalation exposure to mixed xylene or xylene isomers. Inhalation exposure to 810 ppm mixed xylene for 13 weeks produced no adverse adrenal, thyroid, or parathyroid effects in the dog (Carpenter et al. 1975a). No effect on absolute or relative adrenal weights was observed in male rats intermittently exposed to *m*-xylene at concentrations as high as 100 ppm for 13 weeks (Korsak et al. 1994).

Ocular Effects. Human data indicate that acute inhalation exposures to 460 ppm mixed xylene and 100 ppm *p*-xylene vapors produce mild and transient eye irritation (Carpenter et al. 1975a; Hastings et al. 1986; Klaucke et al. 1982; Nelson et al. 1943; Nersesian et al. 1985; NIOSH 1981). This effect is probably the result of direct contact of the xylene vapor with the eye and as such is described under Ocular Effects in Section 3.2.3.2.

No animal data were available regarding ocular effects following inhalation exposure to mixed xylenes or xylene isomers.

Body Weight Effects. No studies were located regarding body weight effects in humans following inhalation exposure to mixed xylenes or xylene isomers.

A number of intermediate-duration intermittent inhalation studies of xylene have examined body weight effects in animals (Carpenter et al. 1975a; Gagnaire et al. 2001, 2006; Gralewicz and Wiaderna 2001; Jajte et al. 2003; Korsak et al. 1992, 1994; Rosengren et al. 1986; Tatrai et al. 1981). Except for the study by Tatrai et al. (1981) in which a 12% decrease in body weight was observed in rats exposed to 1,096 ppm *o*-xylene for 6 months, no significant adverse effects on body weight were noted.

Metabolic Effects. Metabolic acidosis was reported in a man who sniffed paint containing xylenes, but other solvents in the paint may have contributed to the effect (Martinez et al. 1989). No data were located concerning metabolic effects in animals following inhalation exposure to xylenes.

3.2.1.3 Immunological and Lymphoreticular Effects

Limited data were available regarding immunological and lymphoreticular effects of xylene in humans. Decreased lymphocytes (Moszczynski and Lisiewicz 1983, 1984a) and decreased serum complement (Smolik et al. 1973) have been observed in workers exposed to xylene. However, no determination can be made regarding the association between inhalation of xylene and immunological effects from the available human studies, because workers were concurrently exposed to other chemical agents.

Acute exposure (4 days, 4 hours/day) of mice to 1,208 ppm *p*-xylene had no effect on natural killer cell activity, although mortality from murine cytomegalovirus was increased (Selgrade et al. 1993). The investigators (Selgrade et al. 1993) attributed the enhanced virus susceptibility to increased liver toxicity rather than to an effect on the immune system. Intermittent exposure of rats and dogs to mixed xylenes for 10 or 13 weeks at concentrations as high as 810 ppm resulted in no effect on spleen weight (Carpenter et al. 1975a).

3.2.1.4 Neurological Effects

The neurological effects of xylene in humans following inhalation exposure have been evaluated in a number of experimental studies, case reports, and occupational studies. Results of experimental studies with humans indicate that acute inhalation exposure to mixed xylene or *m*-xylene causes impaired short-term memory, impaired reaction time, performance decrements in numerical ability, and alterations in equilibrium and body balance (Carpenter et al. 1975a; Dudek et al. 1990; Gamberale et al. 1978; Riihimaki and Savolainen 1980; Savolainen and Linnavuo 1979; Savolainen and Riihimaki 1981a; Savolainen et al. 1979b, 1984, 1985a).

Dizziness was reported by the majority of subjects exposed to 690 ppm mixed xylene for 15 minutes, but in only one of six persons exposed at 460 ppm (Carpenter et al. 1975a). In objective measures of neurological function, exposure to 100 ppm mixed xylene for 4 hours resulted in prolonged reaction time (Dudek et al. 1990) and exposure to 299 ppm mixed xylene for 70 minutes during exercise resulted in impaired short-term memory and reaction time (Gamberale et al. 1978). No impairment in performance tests was observed in sedentary subjects exposed at 299 ppm for 70 minutes (15 men) (Gamberale et al.

1978) or at 396 ppm for 30 minutes (10 men) (Hastings et al. 1986). The difference between the effects in the absence and presence of exercise may be due to increased xylene respiratory uptake during exercise.

Slight, but statistically significant, increases in the average rating for subjective symptoms of neurological effects were observed following exposure to 50 ppm *m*-xylene vapor compared to controls (Ernstgard et al. 2002). After 60 and 118 minutes of exposure, severity ratings for feelings of intoxication were elevated in men and women, and ratings for headache were elevated in men. The ratings for dizziness were increased in exposed men after 118 minutes of exposure. (This study was chosen as the basis for the acute-duration inhalation MRL, for these subjective neurological effects and changes in objective and subjective measures of respiratory function.) Electroencephalograms obtained from nine men exposed to *m*-xylene at 200 ppm (TWA) for 4 hours showed only minor changes (Seppalainen et al. 1991). These changes were characterized as a slight increase in alpha-wave frequency and percentage early in the exposure period and a decrease in exercise-induced increases in theta and delta waves indicating central nervous system effects. Studies using the *m*-isomer of xylene have also indicated that some tolerance may occur during acute exposures. While exposure to stable concentrations of *m*-xylene for 7 hours or 4 hours, twice a week in the range of up to approximately 280 ppm had no effect on body sway, coordination, or reaction time (Ogata et al. 1970; Savolainen 1980; Savolainen et al. 1980b), exposure for 6 hours or 6–9 days to levels fluctuating between 64 and 400 ppm produced impairment in human body balance and/or reaction time (Savolainen and Linnavuo 1979; Savolainen and Riihimaki 1981a; Savolainen et al. 1979b, 1980a, 1984, 1985a). A 3-hour exposure of nine male volunteers to *m*-xylene at 200 ppm during exercise resulted in a slight but significant ($p<0.05$) change in the N135 component of a pattern visual evoked potential (Seppalainen et al. 1989). Laine et al. (1993) saw no clear effects on visual reaction times or auditive choice reaction times in nine male volunteers exposed to levels of *m*-xylene fluctuating between 135 and 400 ppm (TWA 200 ppm) with or without exercise. Levels of *m*-xylene fluctuating between 135 and 400 ppm produced a slight decrease in the latency of visual evoked potentials (Seppalainen et al. 1989), but no clear effects on visual reaction times or auditory choice reaction times (Laine et al. 1993).

Objective measures of neurological function (electroencephalography, tests of motor activity and cognitive performance) in humans are not affected by acute or intermediate, intermittent or continuous inhalation exposure to *p*-xylene for 4 hours or up to 7 hours for 5 days at concentrations ranging from 69 to 150 ppm (NIOSH 1981; Olson et al. 1985). Differences in such factors as the xylene isomer, the neurological parameter, exposure conditions and concentrations, rapid development of tolerance, and total

xylene uptake may account for the variability in results. However, some sex difference in subjective reports of central nervous system effects was observed (NIOSH 1981). Three women exposed to *p*-xylene at 100 ppm for 1–7.5 hours/day, for 5 days, showed no effects on electroencephalograms, evoked potentials, or cognitive performance, but frequently reported headache and dizziness as a result of exposure (NIOSH 1981). In contrast, four men exposed at concentrations of up to 150 ppm *p*-xylene under the same exposure conditions reported no increase in headaches or dizziness.

Available case reports and occupational studies together provide suggestive evidence that acute and chronic inhalation exposure to xylene or solvent mixtures containing xylene may be associated with neurological effects; however, most studies are difficult to evaluate because the exposure conditions either have not been well characterized or the subjects may have been exposed to other chemicals in addition to xylene. The neurological symptoms observed in these studies include headache, nausea, dizziness, difficulty concentrating, impaired memory, slurred speech, ataxia, fatigue, agitation, confusion, tremors, labored breathing, and sensitivity to noise (Arthur and Curnock 1982; Goldie 1960; Gupta et al. 1990; Hipolito 1980; Klaucke et al. 1982; Martinez et al. 1989; Morley et al. 1970; Nersesian et al. 1985; Roberts et al. 1988). In several case reports, isolated instances of unconsciousness, amnesia, brain hemorrhage, and epileptic seizure have been associated with acute inhalation exposure to solvent mixtures containing xylene (Arthur and Curnock 1982; Goldie 1960; Martinez et al. 1989; Morley et al. 1970). Long-term occupational exposure (≥10 years) to mixed solvents among spray painters was associated with an increase in depression and "loss of interest," but no significant effects on psychological performance tests or CAT-scan measures of brain atrophy (Triebig et al. 1992a, 1992b). Workers exposed to mixed solvents for <10–>30 years exhibited significantly reduced conduction velocities in the radial and tibial nerves, as well as duration-related increases in symptoms of numbness, cramps, and weakness (Jovanovic et al. 2004). Because other chemicals were present with xylenes in many of these studies, the effects observed cannot be conclusively attributed to xylene exposure.

Another occupational study in which xylene exposure was most well defined and represented 70% of the solvent exposure (Uchida et al. 1993) reported an increase in subjective symptoms including an increased prevalence of anxiety, forgetfulness, inability to concentrate, and dizziness among workers exposed to an average TWA concentration of 21 ppm (14 ppm geometric mean) of mixed xylenes for an average of 7 years. No objective measures of neurological impairment were tested in this study. Subjective symptoms of neurological and respiratory toxicity in this study were selected as co-critical effects for the chronic-duration inhalation MRL.

Results of experimental studies with animals also provide evidence that mixed xylene and its isomers are neurotoxic following inhalation exposure. Signs of neurotoxicity observed in rats, mice, dogs, cats, and gerbils following acute and intermediate inhalation exposure to the various xylene isomers include narcosis, prostration, incoordination, tremors, muscular spasms, labored breathing, behavioral changes, hyperreactivity to stimuli, altered visual evoked potentials, elevated auditory thresholds, hearing loss, and decreased acetylcholine in midbrain and norepinephrine in hypothalamus (suggestive of effect on motor control, sleep, and memory maintenance) (Andersson et al. 1981; Bushnell 1989; Carpenter et al. 1975a; De Ceaurriz et al. 1983; Furnas and Hine 1958; Ghosh et al. 1987; Honma et al. 1983; Korsak et al. 1988, 1990; Kyrklund et al. 1987; Molnar et al. 1986; Pryor et al. 1987; Rank 1985; Rosengren et al. 1986; Savolainen and Seppalainen 1979; Savolainen et al. 1978, 1979b; Wimolwattanapun et al. 1987).

Exposure levels associated with neurological effects in animals are well defined. A comparative study determined that the minimal alveolar concentrations needed to induce anesthesia in rats were similar for all three isomers (0.00118, 0.00139, and 0.00151 atm, respectively, for *o*-, *m*-, and *p*-xylene), but only *p*-xylene also induced excitation (strong tremors) (Fang et al. 1996). Acute exposure to unspecified levels of mixed xylene resulted in respiratory paralysis (Morvai et al. 1976), 1,600 ppm *p*-xylene produced hyperactivity (Bushnell 1989), and 1,300 ppm mixed xylene produced incoordination in rats, which did not persist after exposure ended; no overt signs of toxicity were noted at 580 ppm (Carpenter et al. 1975a). All three xylene isomers produced narcosis in rats after 1–4 hours of exposure to concentrations of approximately 2,000 ppm (Molnar et al. 1986). No behavioral signs of xylene intoxication were observed in dogs or monkeys exposed continuously to 78 ppm *o*-xylene for up to 127 days, but dogs exposed to 780 ppm *o*-xylene intermittently for 6 weeks exhibited tremors during exposure (Jenkins et al. 1970).

The neurotoxicity of xylenes has been evaluated in neurobehavioral tests on animals exposed by inhalation. Mice exposed for 30 minutes by inhalation to any of the isomers exhibited impaired operant performance at the same minimal effective concentration, 1,400 ppm, but the median effective concentrations varied to a limited degree—5,179 ppm for *o*-xylene, 5,611 ppm for *p*-xylene, and 6,176 ppm for *m*-xylene (Moser et al. 1985). The order of potency was different for impairment of motor coordination in mice undergoing the inverted screen test, with a minimal effective concentration of 2,000 ppm for *p*-xylene and 3,000 ppm for the two other isomers (Moser et al. 1985); the median effective concentrations were 2,676, 3,640, and 3,790 ppm, respectively, for *p*-, *o*-, and *m*-xylene. Acute exposures to concentrations inducing behavioral changes in rats and mice ranged from 114 ppm for effects of mixed xylene on operant conditioning or self-stimulation behavior (Ghosh et al. 1987;

Wimolwattanapun et al. 1987), 500 ppm for reduced response rate in schedule-controlled operant behavior in mice exposed to *m*-xylene (Bowen et al. 1998), to 1,010 ppm for *o*-xylene-induced immobility in a "behavioral despair swimming test" (De Ceaurriz et al. 1983). Exposure of male rats to 230 ppm *o*-xylene for 4 hours shortened the duration of response (extension of hindlimbs) to an applied electrical shock by 18.8% (Vodickova et al. 1995); doubling the exposure concentration correspondingly doubled the magnitude of the response. In the same report, exposure of female mice to 320 ppm *o*-xylene for 2 hours shortened the duration of response (velocity of tonic extension, i.e., the reciprocal of the latency) was reduced by 11%; unlike rats, exposure at twice the concentration, increased the magnitude of the response by a factor of 3.7. Impaired rotarod performance was observed in rats acutely exposed to mixed xylene and the individual xylene isomers at concentrations of ≥3,000 ppm (Korsak et al. 1990). In intermediate-duration inhalation studies with rats, exposure to 100 ppm *m*-xylene intermittently for 3 or 6 months or to 1,000 ppm for 3 months showed decreased rotarod performance and decreased spontaneous activity (Korsak et al. 1992, 1994). The effect was greater following the 3-month exposure at 1,000 ppm than the 6-month exposure at 100 ppm suggesting that for effects on motor activity, concentration is more important than duration of exposure. The persistence of neurological effects of xylene was examined in some studies that evaluated tested animals some weeks after the last exposure. Rats intermittently exposed to 100 ppm *m*-xylene for 4 weeks exhibited impaired passive avoidance learning (tested 5 weeks after exposure) and impaired acquisition, but not retention, of the two-way active avoidance response (tested 9 weeks after exposure) (Gralewicz and Wiaderna 2001). Exposure had no lasting effect in tests for short-term memory, responsiveness to a thermal stimulus (paw-lick latency), or spontaneous activity (open-field test) or the retention of a learned active avoidance response.

Sensory deficits resulting from xylene exposure have been observed under controlled testing conditions. Acute exposure to 1,600 ppm, but not 800 ppm *p*-xylene depressed the amplitude of visual evoked potentials (Dyer et al. 1988). Hearing deficits have been reported in animals exposed to xylene for acute or intermediate durations. Hearing loss occurred in rats exposed to 1,450 ppm mixed xylene for 8 hours, whereas exposure to 1,700 ppm for 4 hours produced no effects on hearing (Pryor et al. 1987) indicating that the duration of exposure is important for the observation of ototoxic effects in conditioned avoidance tests. In rats acutely exposed to 1,800 ppm mixed xylenes, 18–30 dB hearing losses were observed at mid-range frequencies (Crofton et al. 1994). The amplitude of brainstem auditory evoked potentials was reduced by 50% in rats acutely exposed at 2,000 ppm, but not at 1,700 ppm (Rebert et al. 1995). Hearing loss was also evident after exposure for 6 weeks to 800 ppm mixed xylene (Pryor et al. 1987). In rats exposed intermittently to 900 ppm *p*-xylene for 13 weeks, there was some hair cell loss in the organ of Corti, but no effect on auditory neurophysiology (Gagnaire et al. 2001). At 1,800 ppm, extensive

ototoxicity (altered brainstem auditory evoked potentials, measurable hearing loss [deficits of 35–42 dB] and significant hair cell loss in the organ of Corti) was observed as early as the fourth week and did not improve during an 8-week recovery period; neither *o*-xylene nor *m*-xylene were ototoxic in this study. All rats exposed for 13 weeks to 1,000 or 2,000 ppm mixed xylenes containing ethylbenzene exhibited significant irreversible ototoxicity including hearing deficits and loss of hair cells (Gagnaire et al. 2006). Exposures to these mixtures at 250 or 500 ppm resulted in a small loss of hair cells in one of eight rats per group, but no effect on auditory thresholds. According to the authors, ethylbenzene contributes significantly to the ototoxicity of technical xylene.

Changes in neuronal cells and brain biochemistry have been noted in animals following exposure to xylenes. Acute exposure to *p*-xylene caused decreased axonal transport at concentrations as low as 800 ppm (Padilla and Lyerly 1989); however, no such decrease was apparent 3 days after exposures had ceased. At 1,600 ppm, however, the decrease in axonal transport persisted for 13 days after exposure. Acute inhalation of 2,000 ppm mixed xylene produced increased dopamine and/or noradrenaline levels in the hypothalamus of rats; no behavioral changes were assessed (Andersson et al. 1981). Levels of these catecholamines in the hypothalamus of rats were also increased following inhalation of 2,000 ppm *m*-, *o*-, or *p*-xylene (Andersson et al. 1981). Brain concentrations of deoxyribonucleic acid (DNA) and/or astroglial proteins increased in rats (at 300–320 ppm) and gerbils (at 160 ppm) after intermediate continuous exposure of 3–4.5 months to xylene (Rosengren et al. 1986; Savolainen and Seppalainen 1979). In addition, increased levels of brain enzymes, changes in axon membranes, and behavioral changes occurred in rats after exposure to 300 ppm of mixed xylene for 18 weeks (Savolainen and Seppalainen 1979; Savolainen et al. 1979a). Alterations in neurotransmitter levels were observed in some brain areas at 800 ppm mixed xylene for 30 days (Honma et al. 1983). However, no significant long-term alterations in fatty acid levels were noted in the brains of rats after intermediate-duration exposure of 30 or 90 days to 320 ppm mixed xylene (Kyrklund et al. 1987). At 1,600 ppm *m*-xylene for 7 weeks, decreased α-adrenergic binding compared to the controls was observed in the hypothalamus of exposed mice (Rank 1985). No persistent changes in brain weight or weights of the caudate-putamen or subcortical limbic areas, or in agonist binding to the dopamine D_2 receptor in those regions occurred in rats exposed to 80 ppm *p*-xylene for 4 weeks and examined 5 weeks after the last exposure (Hillefors-Berglund et al. 1995).

No animal studies were located regarding neurological effects following chronic inhalation exposure to mixed xylene or its isomers.

The highest NOAEL values and all LOAEL values for each reliable study for neurological effects in each species and duration category are recorded in Table 3-1 and plotted in Figure 3-1. An acute-duration inhalation MRL of 2 ppm was calculated for mixed xylenes based on a LOAEL for subjective neurological effects and measured and subjective respiratory effects in human subjects exposed to 50 ppm *m*-xylene for 2 hours (Ernstgard et al. 2002; see footnote in Table 3-1). An intermediate-duration inhalation MRL of 0.6 ppm was calculated for mixed xylenes based on a minimal LOAEL of 50 ppm for decreased mean latency of the paw-lick response in rats exposed to *m*-xylene for 6 hours/day, 5 days/week for 3 months (Korsak et al. 1994; see footnote in Table 3-1). A chronic-duration inhalation MRL of 0.05 ppm was calculated for mixed xylenes based on a LOAEL of 14 ppm for subjective neurological and respiratory symptoms in workers exposed to mixed xylene 8 hours/day, 5 days/week for an average of 7 years (Uchida et al. 1993; see footnote in Table 3-1).

3.2.1.5 Reproductive Effects

A few occupational studies evaluated reproductive effects in workers exposed to xylenes. A case-control study of spontaneous abortions among Finnish workers for whom there was biomonitoring data for exposure to organic solvents found no statistically significant increase in the odds ratio associated with exposure to xylene (Lindbohm et al. 1990). Spontaneous abortions in early pregnancy were significantly increased among 37 women exposed to xylene and formalin in pathology or histology laboratories (Taskinen et al. 1994). Although the increased odds ratio (OR=3.1; 95% confidence interval=1.3–7.5) for spontaneous abortion was statistically significant for women exposed to xylene at least 3–4 days/week, the analysis cannot be considered definitive because of simultaneous exposure to other solvents and chemicals. A cross-sectional study among 1,408 petrochemical workers in China reported an increased prevalence of oligomenorrhea among workers exposed to organic solvents, but the contribution of xylene cannot be definitively determined as all workers who were exposed to xylene were also exposed to other solvents such as benzene, toluene, and styrene (Cho et al. 2001).

Continuous exposure of CFY rats for 8 days on days 7–14 during pregnancy to 775 ppm mixed xylene produced an increased number of resorptions without any maternal toxicity; reduced fertility was also observed (Balogh et al. 1982). However, no adverse reproductive effects were noted following inhalation exposure of male and female CD rats to mixed xylene at concentrations as high as 500 ppm during premating, mating, pregnancy, and lactation (Bio/dynamics 1983). No effect on absolute or relative testicular weights was observed in rats intermittently exposed to *m*-xylene at concentrations as high as 100 ppm for 13 weeks (Korsak et al. 1994). Inhalation exposure of male Sprague-Dawley rats to

1,000 ppm mixed xylene for 61 days produced no alterations in testes, accessory glands, or circulating male hormone levels (Nylen et al. 1989). Strain differences may account for the differential response to mixed xylene in these studies. The highest NOAEL and LOAEL values for each reliable study for reproductive effects in rat for each duration category are recorded in Table 3-1 and plotted in Figure 3-1.

3.2.1.6 Developmental Effects

Although the human data regarding the developmental effects of xylene suggest a possible relationship between solvent (unspecified) exposure and developmental toxicity, these data are limited for assessing the relationship between inhalation of xylene and developmental effects because the available studies involved concurrent exposure to other solvents in addition to xylene in the workplace (Holmberg and Nurminen 1980; Kucera 1968; Taskinen et al. 1989; Windham et al. 1991), and because of the small number of subjects ranging from 9 to 61 (Taskinen et al. 1989; Windham et al. 1991).

Both mixed xylene and the individual isomers produce fetotoxic effects in laboratory animals. Effects of mixed xylene observed in rats, mice, and rabbits included increased incidences of skeletal variations in fetuses, delayed ossification, fetal resorptions, hemorrhages in fetal organs, and decreased fetal body weight (Balogh et al. 1982; Bio/dynamics 1983; Hass and Jakobsen 1993; Hudak and Ungvary 1978; Litton Bionetics 1978a; Mirkova et al. 1983; Ungvary 1985; Ungvary and Tatrai 1985). Balogh et al. (1982) reported delayed ossification in all exposure levels (53–775 ppm), but as the incidences were inversely related to dose, this end point is not included in LSE Table 3-1. The levels at which these effects were observed depended upon the composition and concentration of mixed xylene, the time of exposure, and the choice of strain and test species used. In addition, animals in a number of studies were exposed 24 hours/day (Balogh et al. 1982; Hudak and Ungvary 1978; Ungvary 1985; Ungvary and Tatrai 1985), whereas animals in other studies (Bio/dynamics 1983; Hass and Jakobsen 1993; Litton Bionetics 1978a; Mirkova et al. 1983) were exposed 6 hours/day. The study conducted by Litton Bionetics (1978a) used a formulation of mixed xylene with a comparatively high percentage (36%) of ethylbenzene. Developmental effects were reported following maternal exposure to concentrations as low as 12 ppm mixed xylene in rats (Mirkova et al. 1983), but the health of the test animals may have been compromised due to poor animal husbandry. This is suggested by the relatively low conception rates and the high incidence of fetal hemorrhages seen in the controls. Maternal toxicity was observed at 775 ppm in the study by Balogh et al. (1982) and at 138 ppm in the study by Ungvary (1985); however, no maternal toxicity occurred at exposure levels of 100–400 ppm in the studies by Bio/dynamics (1983), Hass and Jacobsen (1993), Hudak and Ungvary (1978), and Litton Bionetics (1978a). Insufficient evidence was

presented to determine whether maternal toxicity occurred in the studies by Mirkova et al. (1983) and Ungvary and Tatrai (1985). Many of the studies (Balogh et al. 1982; Bio/dynamics 1983; Hudak and Ungvary 1978; Mirkova et al. 1983; Ungvary 1985; Ungvary and Tatrai 1985) had limitations that made them difficult to assess (e.g., unknown composition of xylene and insufficient number of doses to form a dose-response relationship; lack of detail with regard to both methods and data obtained). Furthermore, several of these studies (Balogh et al. 1982; Ungvary and Tatrai 1985; Ungvary et al. 1980b) reported the incidence of fetal effects, such as delayed skeletal ossification and extra ribs, on the basis of total fetuses per exposure group, but not on a per-litter basis. Without litter-specific results, it is not possible to adjust for possible covariance with litter size, as reported by Litton Bionetics (1978a). Given the uncertainty of the significance of these effects, increases in the incidences of delayed ossification or skeletal variants were not used to establish NOAELs or LOAELs unless litter-specific data were provided. Based on the reliable data in these reports (fetal weight, fetal death, implantation losses), developmental toxicity in rats occurred at concentrations of ≥350 ppm; these effects are noted in Table 3-1.

A number of studies reported neurobehavioral deficits in rats resulting from exposure to xylene during gestation. ATSDR considers these effects to be serious. Hass and Jakobsen (1993) reported decreased rotarod performance in 1- and 2-day-old rat pups exposed to 200 ppm mixed xylenes 6 hours/day on gestation days 4–20. Female offspring of rats exposed to 500 ppm mixed xylenes for 6 hours/day on gestation days 7–20 showed significant reductions in absolute brain weight after weaning and neurobehavioral deficits (delayed air righting reflex, nonsignificant trends for reduced motor coordination in the Rotarod test, and increased latencies in the Morris water maze test) (Hass et al. 1995). Hass et al. (1995) suggested that the delayed air righting reflex and impaired spatial navigation in the water maze (locating a re-situated platform) may have been related to ototoxicity, since both tasks involve the vestibular system and cochlear hair cells. The latency in locating a moved platform was significantly increased in the first learning trial by exposed offspring tested at week 28, but was not significantly different from controls at 55 weeks (Hass et al. 1997).

Inhalation of *o*- or *p*-xylene at concentrations similar to those at which mixed xylene caused fetal toxicity, produced decreased fetal weight, skeletal retardation, and post-implantation loss in rats, mice, and rabbits following maternal acute exposure during gestation days 7–14/15 (Ungvary and Tatrai 1985; Ungvary et al. 1980b, 1981); no maternal toxicity was observed. A NOAEL value of 1,612 ppm *p*-xylene for developmental effects was determined from one study with rats (Rosen et al. 1986). The large variation in concentrations of xylene producing developmental effects and those producing no developmental effects may be influenced by a number of factors (e.g., strain and species of animal, purity of xylene,

method of exposure, exposure pattern and duration, etc.). For example, Rosen et al. (1986) exposed animals for 6 hours/day, whereas animals were exposed 24 hours/day in studies by Ungvary and Tatrai (1985) and Ungvary et al. (1980b, 1981). No information on maternal toxicity was available for the studies by Ungvary and Tatrai (1985) or Ungvary et al. (1981); however, in the studies by Rosen et al. (1986) and Ungvary et al. (1980b), signs of maternal toxicity in rats following inhalation of the isomers included decreased weight gain, decreased food consumption, and increased liver-to-body weight ratios. *m*-Xylene was the only isomer that resulted in lasting maternal growth inhibition or maternal mortality (Ungvary et al. 1980b). Thus, it is difficult to determine from these studies whether mixed xylenes are selectively toxic to the fetus or the observed developmental toxicity was secondary to maternal toxicity. As mentioned above, retardation in skeletal growth or variations, in the absence of litter-specific data, were not used to determine NOAELs or LOAELs for exposure to specific isomers of xylene (Ungvary et al. 1980b).

Results of a well-documented comparative standard developmental toxicity assay in rats exposed for 6 hours/day on gestational days 6–20, suggest that developmental toxicities of *m*- and *p*-xylene are secondary to maternal toxicity (Saillenfait et al. 2003). Statistically significant maternal effects (reduced 'corrected weight gain'—body weight gain exclusive of gravid uterus weight) and fetal toxicity (reduced fetal weight) occurred at exposures of ≥1,000 ppm, but not 500 ppm for these two isomers. Both *m*- and *p*-xylene at 2,000 ppm induced significant increases in delayed ossification, a skeletal variation related to retarded growth, although the affected bones were different for the two isomers. Dams exposed at ≥1,000 ppm to *o*-xylene or mixed xylenes showed similar effects, although the reduced corrected weight gain for mixed xylenes was only significant for gestational days 6–13. However, both *o*-xylene and mixed xylene caused significantly reduced fetal body weights at 500 ppm, the maternal NOAEL. Exposure to mixed xylenes did not elicit any developmental structural anomalies at concentrations as high as 2,000 ppm, whereas *o*-xylene elicited increases in the percent of fetuses per litter with skeletal variations (incomplete ossification) at 2,000 ppm and in the percent of fetuses with any variation at 1,000 ppm. This assay did not include testing for postnatal neurobehavioral effects.

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

3.2.1.7 Cancer

Human data regarding cancer are limited to three occupational studies and one population-based study. Two studies examined the cancer and leukemia risks among solvent-exposed workers and suggested a possible relationship between coal-based xylene exposure and leukemia (Arp et al. 1983; Wilcosky et al. 1984). Both contain limitations (e.g., small number of subjects ranging from 9 to 85 male workers, no exposure concentrations, unknown composition of xylene) that preclude a definitive conclusion regarding inhalation of xylene and cancer. A retrospective cohort mortality study evaluated 14,457 workers at an aircraft maintenance facility, 108 of whom had been exposed to xylene (Spirtas et al. 1991; Stewart et al. 1991). In this study, evaluations of deaths from multiple myeloma or non-Hodgkin's lymphoma revealed no cases among xylene-exposed workers, representing 1,837 and 444 person-years of exposure, for men and women, respectively (Spirtas et al. 1991; Stewart et al. 1991). A population-based case-control study that estimated job-related exposure to xylene reported limited evidence of increased risks for cancers of the colon and rectum following high exposure to xylene (Gerin et al. 1998). The authors indicated that the marginally significant excesses in rectal cancer in xylene-exposed workers may have been due to the confounding effect of exposure to styrene or another material associated with styrene exposure. Limitations of this study included a lack of information on exposure concentrations and the composition of xylene, a small number of cases, and the fact that most of the xylene-exposed workers were also exposed to toluene and benzene.

No studies were located regarding cancer in animals exposed via inhalation to mixed xylene or xylene isomers.

3.2.2 Oral Exposure

3.2.2.1 Death

Death in humans following accidental or intentional ingestion of xylene was reported by Abu Al Ragheb et al. (1986). Levels of xylene found in blood and gastric and duodenal contents were 110, 8,800, and 33,000 mg/L, respectively, indicating ingestion of a large, but undetermined, quantity of xylene. Death was attributed to respiratory failure secondary to depression of the respiratory center in the brain.

Mortality was observed in laboratory animals following the ingestion of mixed xylene or isomers of xylene. Reported acute oral LD_{50} values in rats for mixed xylene range from 3,523 mg/kg when administered in corn oil (NTP 1986) to 8,600 mg/kg when administered undiluted (Hine and Zuidema

1970). It appears that the absorption of xylene was enhanced by corn oil due to its greater lipophilicity. The acute oral LD_{50} values for mixed xylene administered in corn oil to male and female mice were 5,627 and 5,251 mg/kg, respectively (NTP 1986). The acute oral LD_{50} for *m*-xylene (neat) in rats was 6,661 mg/kg (Smyth et al. 1962). According to a study by Gerarde (1959), *m*-xylene may be slightly less toxic than the other two isomers, since a single oral dose of 4,320 mg/kg *of m*-xylene resulted in death in 3/10 rats, whereas 4,400 mg/kg of *o*-xylene or 4,305 mg/kg of *p*-xylene produced death in 7/10 and 6/10 rats, respectively. In acute-duration repeated-dose gavage studies, mortality was observed in 8 of 10 rats given 2,000 mg/kg mixed xylene and 10 of 10 mice given 4,000 mg/kg mixed xylene in corn oil for 14 days (NTP 1986) and in 2 females from a group of 10 male and 10 female rats that received 2,000 mg/kg/day *p*-xylene for 10 days (Condie et al. 1988).

Survival was significantly lowered in male rats exposed to mixed xylene 5 days/week at chronic oral doses of 500 mg/kg but not at 250 mg/kg (NTP 1986); females were not affected. Although mortality appeared to be dose related in the treated rats, many of the early deaths were related to an error in gavage methodology; non-accidental deaths occurred with an incidence of 22% in controls, 32% at 250 mg/kg/day, and 38% at 500 mg/kg/day.

All LD_{50} values and LOAEL values from each reliable study for death in each species and duration category are recorded in Table 3-2 and plotted in Figure 3-2.

3.2.2.2 Systemic Effects

The systemic effects observed after oral exposure to xylene are discussed below. The highest NOAEL value and all LOAEL values from each reliable study for systemic effects in each species and duration category are recorded in Table 3-2 and plotted in Figure 3-2.

Respiratory Effects. Limited information was located regarding respiratory effects in humans following oral exposure to mixed xylene or xylene isomers. Postmortem examination of a man who committed suicide by ingesting xylene showed pulmonary congestion and edema (Abu Al Ragheb et al. 1986). Death resulted from centrally mediated respiratory depression.

A single oral dose of 4,000 mg/kg in mice or daily oral dosing of rats and mice by gavage with mixed xylene for 14 days at 2,000 mg/kg/day resulted in shallow and labored breathing immediately after dosing, but no compound-related effects were observed in the lungs at necropsy (NTP 1986). Mice given

Table 3-2 Levels of Significant Exposure to Xylene - Oral

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference Chemical Form	Comments
ACUTE EXPOSURE								
Death								
1	Rat Sprague-Dawley	10 d 1 x/d (GO)				2000 F (2/20 died)	Condie et al. 1988 para	
2	Rat NS	once (GO)				4305 (6/10 rats died)	Gerarde 1959 para	
3	Rat NS	once (GO)				4400 (7/10 died)	Gerarde 1959 ortho	
4	Rat NS	once (GO)				4320 (3/10 died)	Gerarde 1959 meta	
5	Rat Long- Evans	once (G)				8640 M (LD50)	Hine and Zuidema 1970 mixed	
6	Rat Albino-Wistar CFT	once (GO)				5950 F (4/6 died)	Muralidhara and Krishnakumari 1980 mixed	
7	Rat F344/N	14 d 1 x/d (GO)				2000 (8/10 died)	NTP 1986 mixed	
8	Rat F344/N	once (GO)				3523 M (LD50)	NTP 1986 mixed	
9	Rat Carworth-Wistar	once (G)				6661 M (LD50)	Smyth et al. 1962 meta	

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

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference Chemical Form	Comments
10	Mouse B6C3F1	once (GO)				5627 M (LD50) 5251[b] F (LD50)	NTP 1986 mixed	
11	Mouse B6C3F1	14 d 1 x/d (GO)				4000 (10/10 died)	NTP 1986 mixed	
Systemic								
12	Rat Sprague-Dawley	10 d 1 x/d (GO)	Hemato	2000			Condie et al. 1988 meta	
			Renal	2000				
			Bd Wt	2000				
13	Rat Sprague-Dawley	10 1 x/d (GO)	Hemato	2000			Condie et al. 1988 para	
			Renal	2000				
			Bd Wt	1000	2000 M (13% decrease in body weight)			
14	Rat Sprague-Dawley	10 d 1 x/d (GO)	Hemato	2000			Condie et al. 1988 ortho	
			Renal	2000				
			Bd Wt	1000	2000 M (14% decrease in body weight)			

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

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference Chemical Form	Comments
15	Rat Long- Evans	once (GO)	Other	1000 M	2000 M (mild hypothermia)		Dyer et al. 1988 para	
16	Rat F344/N	14 d 1 x/d (GO)	Resp	1000		2000 (shallow and labored breathing)	NTP 1986 mixed	
			Bd Wt	500	1000 M (18% decrease in body weight gain in males)			
17	Rat Sprague-Dawley	once (GO)	Resp		1000 F (decreased pulmonary microsomal activity)		Patel et al. 1978 para	
18	Mouse B6C3F1	14 d 1 x/d (GO)	Resp	1000		2000 (shallow breathing)	NTP 1986 mixed	
			Bd Wt	2000				
Immuno/ Lymphoret								
19	Rat Sprague-Dawley	10 d 1 x/d (GO)		1000	2000 (11% decrease in relative thymus weight)		Condie et al. 1988 para	
Neurological								
20	Rat Long- Evans	once (GO)		125[c] M	250 M (suppressed visual evoked potentials)		Dyer et al. 1988 para	

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

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference Chemical Form	Comments
21	Rat (Sprague-Dawley)	2 wk 5 d/wk 1 x/d (GO)		900 M			Gagnaire and Langlais 2005 meta	
22	Rat (Sprague-Dawley)	2 wk 5 d/wk 1 x/d (GO)			900 M (partial loss of outer hair cells in cochlea responsive to medium frequencies: 10-25 kHz)		Gagnaire and Langlais 2005 para	
23	Rat (Sprague-Dawley)	2 wk 5 d/wk 1 x/d (GO)		900 M			Gagnaire and Langlais 2005 ortho	
24	Rat Albino- Wistar	once (GO)				5950 F (coma)	Muralidhara and Krishnakumari 1980 mixed	
25	Rat F344/N	once (GO)				4000 (decreased hindleg movement, incoordination, prostration)	NTP 1986 mixed	
26	Mouse B6C3F1	1 x/d 5 d/wk 13 wk (GO)				2000 (weakness, lethargy, unsteadiness, tremors, and partial paralysis)	NTP 1986 mixed	

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

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference Chemical Form	Comments
Developmental								
27	Mouse CD-1	10 d Gd 6-15 3 x/d (GO)		1030 F		2060 F (cleft palate)	Marks et al. 1982 mixed	
28	Mouse ICR/SIM	5 d 1 x/d Gd 8-12 (GO)		2000 F			Seidenberg et al. 1986 meta	
INTERMEDIATE EXPOSURE								
Systemic								
29	Rat Sprague-Dawley	90 d 1 x/d (GO)	Hemato	750 F	1500 F (mild polycythemia and leukocytosis; increased spleen weight)		Condie et al. 1988 mixed	
			Hepatic	150	750 (14% increase in serum alanine aminotransferase in females, 14-17% increase in relative liver weight in both sexes)			
			Renal	150 F	750 F (minimal chronic nephropathy in 6/10 females)			
			Bd Wt	1500				

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

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference Chemical Form	Comments
30	Rat NS	3.5 wk 5 d/wk 1 x/d (GO)	Resp		800 M (decreased cytochrome P-450)		Elovaara et al. 1989 meta	
			Hepatic		800 M (increased plasma SGPT and relative liver weight)			
31	Rat F344/N	13 wk 5 d/wk 1 x/d (GO)	Resp	1000			NTP 1986 mixed	
			Cardio	1000				
			Gastro	1000				
			Hemato	1000				
			Musc/skel	1000				
			Hepatic	1000				
			Renal	1000				
			Ocular	1000				
			Bd Wt	500 M	1000 M (15% decrease in body weight gain)			

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

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference Chemical Form	Comments
32	Rat Sprague-Dawley	13 wk 7 d/wk 1 x/d (GO)	Resp	800			Wolfe 1988a meta	
			Cardio	800				
			Gastro	800				
			Hemato	800				
			Musc/skel	800				
			Hepatic	800 F	800 M (37% increased serum alanine aminotransferase)			
			Renal	800				
			Dermal	800				
			Ocular	800				
			Bd Wt	200 M	800 M (body weight gain decreased by 15%)			

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

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference Chemical Form	Comments
33	Rat Sprague-Dawley	13 wk 7 d/wk 1 x/d (GO)	Resp	800			Wolfe 1988b para	
			Cardio	800				
			Gastro	800				
			Hemato	800				
			Musc/skel	800				
			Hepatic	800				
			Renal	800				
			Dermal	800				
			Ocular	800				
			Bd Wt	800				
34	Mouse B6C3F1	13 wk 5 d/wk 1 x/d (GO)	Cardio	2000			NTP 1986 mixed	
			Gastro	2000				
			Hemato	2000				
			Musc/skel	2000				
			Hepatic	2000				
			Renal	2000				
			Ocular	2000				
			Bd Wt	2000				

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

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference Chemical Form	Comments
Neurological								
35	Rat Sprague-Dawley	90 d 1 x/d (GO)		750 M	1500 M (increased aggressiveness)		Condie et al. 1988 mixed	
36	Rat F344/N	13 wk 5 d/wk 1 x/d (GO)		1000			NTP 1986 mixed	
37	Rat Sprague-Dawley	13 wk 7 d/wk 1 x/d (GO)		800			Wolfe 1988a meta	
38	Rat Sprague-Dawley	13 wk 7 d/wk 1 x/d (GO)		800			Wolfe 1988b para	
39	Mouse B6C3F1	103 wk 5 d/wk 1x/d (GO)		500[d]	1000 (hyperactivity weeks 4-52)		NTP 1986 mixed	
Reproductive								
40	Rat F344/N	13 wk 5 d/wk 1 x/d (GO)		1000			NTP 1986 mixed	
41	Rat Sprague-Dawley	13 wk 7 d/wk 1 x/d (GO)		800			Wolfe 1988a meta	

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

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference Chemical Form	Comments
42	Rat Sprague-Dawley	13 wk 7 d/wk 1 x/d (GO)		800			Wolfe 1988b para	
43	Mouse B6C3F1	13 wk 5 d/wk 1 x/d (GO)		2000			NTP 1986 mixed	
CHRONIC EXPOSURE								
Death								
44	Rat F344/N	103 wk 5 d/wk 1 x/d (GO)				500 M (survival decreased by 16%)	NTP 1986 mixed	

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

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference Chemical Form	Comments
Systemic								
45	Rat F344/N	103 wk 5 d/wk 1 x/d (GO)	Resp	500			NTP 1986 mixed	
			Cardio	500				
			Gastro	500				
			Hemato	500				
			Musc/skel	500				
			Hepatic	500				
			Renal	500				
			Dermal	500				
			Ocular	500				
			Bd Wt	250	500 M (body weight decreased 5-8% after week 59)			

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

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference Chemical Form	Comments
46	Mouse B6C3F1	103 wk 5 d/wk 1 x/d (GO)	Resp	1000			NTP 1986 mixed	
			Cardio	1000				
			Gastro	1000				
			Hemato	1000				
			Musc/skel	1000				
			Hepatic	1000				
			Renal	1000				
			Dermal	1000				
			Ocular	1000				
			Bd Wt	1000				
Neurological								
47	Rat F344/N	103 wk 5 d/wk 1 x/d (GO)		500[e]			NTP 1986 mixed	
Reproductive								
48	Rat F344/N	103 wk 5 d/wk 1 x/d (GO)		500			NTP 1986 mixed	

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

Key to Figure[a]	Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL (mg/kg/day)	LOAEL Less Serious (mg/kg/day)	LOAEL Serious (mg/kg/day)	Reference Chemical Form	Comments
49	Mouse B6C3F1	103 wk 5 d/wk 1 x/d (GO)		1000			NTP 1986 mixed	

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

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

c Used to derive an acute-duration minimal risk level (MRL) for mixed xylenes based on a NOAEL of 125 mg/kg/day p-xylene. The NOAEL was divided by an uncertainty factor of 100 (10 for extrapolation from animals to humans and 10 for human variability).

d Used to derive an intermediate-duration minimal risk level (MRL) for mixed xylenes based on a NOAEL of 500 mg/kg and a LOAEL of 1000 mg/kg for mice exposed to mixed xylenes 5 days/week. The NOAEL was adjusted for intermittent exposure (5 days/7 days) and divided by an uncertainty factor of 100 (10 for extrapolation from animals to humans and 10 for human variability) and a modifying factor of 10 to account for the lack of testing for sensitive neurological endpoints and lack of developmental and multi-generational data.

e Used to derive a chronic-duration minimal risk level (MRL) for mixed xylenes based on a NOAEL of 250 mg/kg in rats exposed to mixed xylenes 5 days/week. At 500 mg/kg, a decrease in survival was observed, thus the next lowest dose was selected as the basis of the MRL. The NOAEL of 250 mg/kg/ was adjusted for intermittent exposure (5 days/7 days) and divided by an uncertainty factor of 100 (10 for extrapolation from animals to humans and 10 for human variability) and a modifying factor of 10 to account for the lack of testing for sensitive neurological endpoints and lack of developmental and multi-generational data.

Bd Wt = body weight; Cardio = cardiovascular; d = day(s); F = Female; Gastro = gastrointestinal; Gd = gestational day; hemato = hematological; LD50 = lethal dose, 50% kill; LOAEL = lowest-observed-adverse-effect level; M = male; Musc/skel = musculoskeletal; NOAEL = no-observed-adverse-effect level; Resp = respiratory; SGPT = serum glutamic pyruvic transaminase; x = time(s); wk = week(s)

Figure 3-2 Levels of Significant Exposure to Xylene - Oral

Acute (≤14 days)

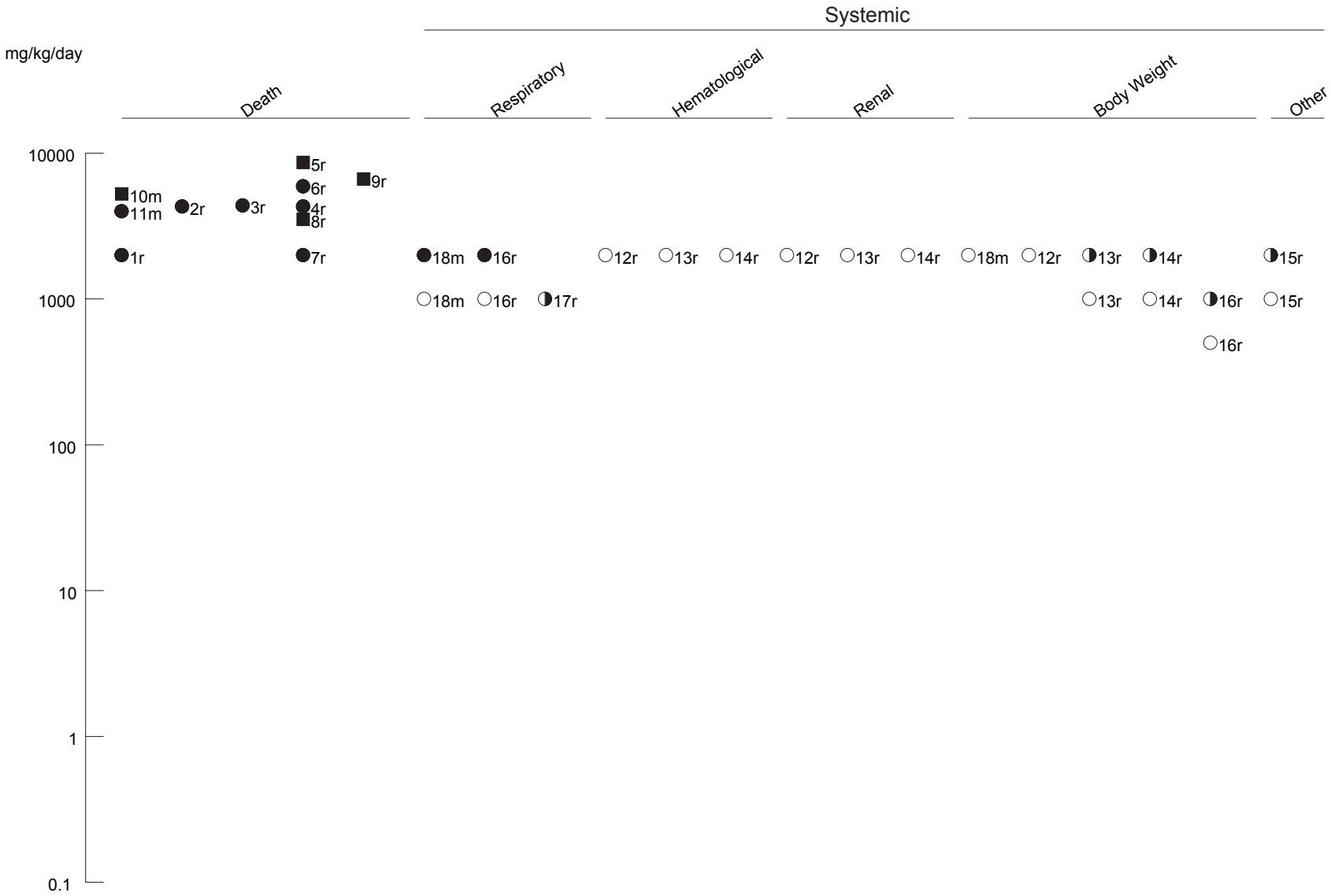


Figure 3-2 Levels of Significant Exposure to Xylene - Oral *(Continued)*

Acute (≤14 days)

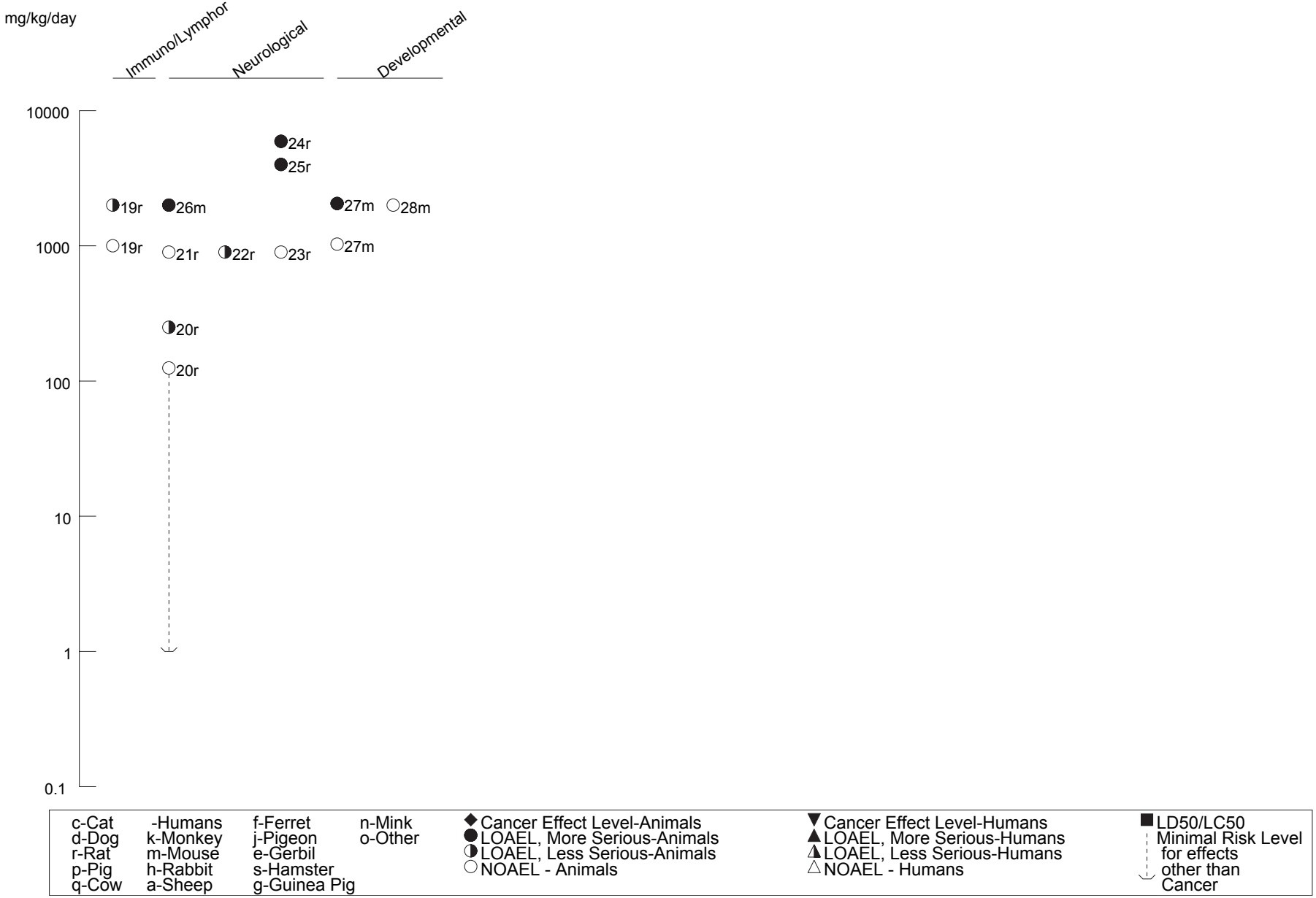


Figure 3-2 Levels of Significant Exposure to Xylene - Oral *(Continued)*

Intermediate (15-364 days)

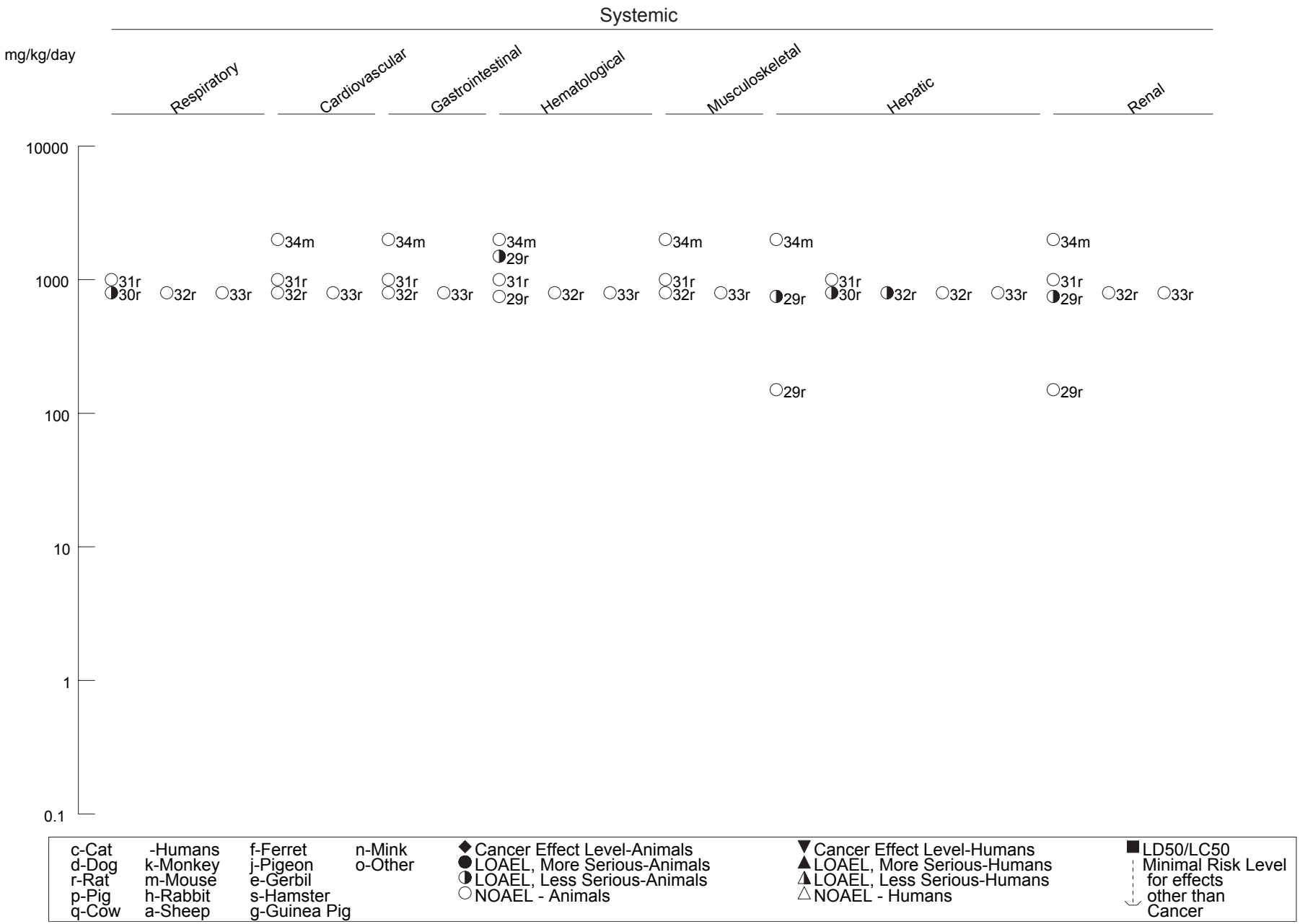


Figure 3-2 Levels of Significant Exposure to Xylene - Oral *(Continued)*

Intermediate (15-364 days)

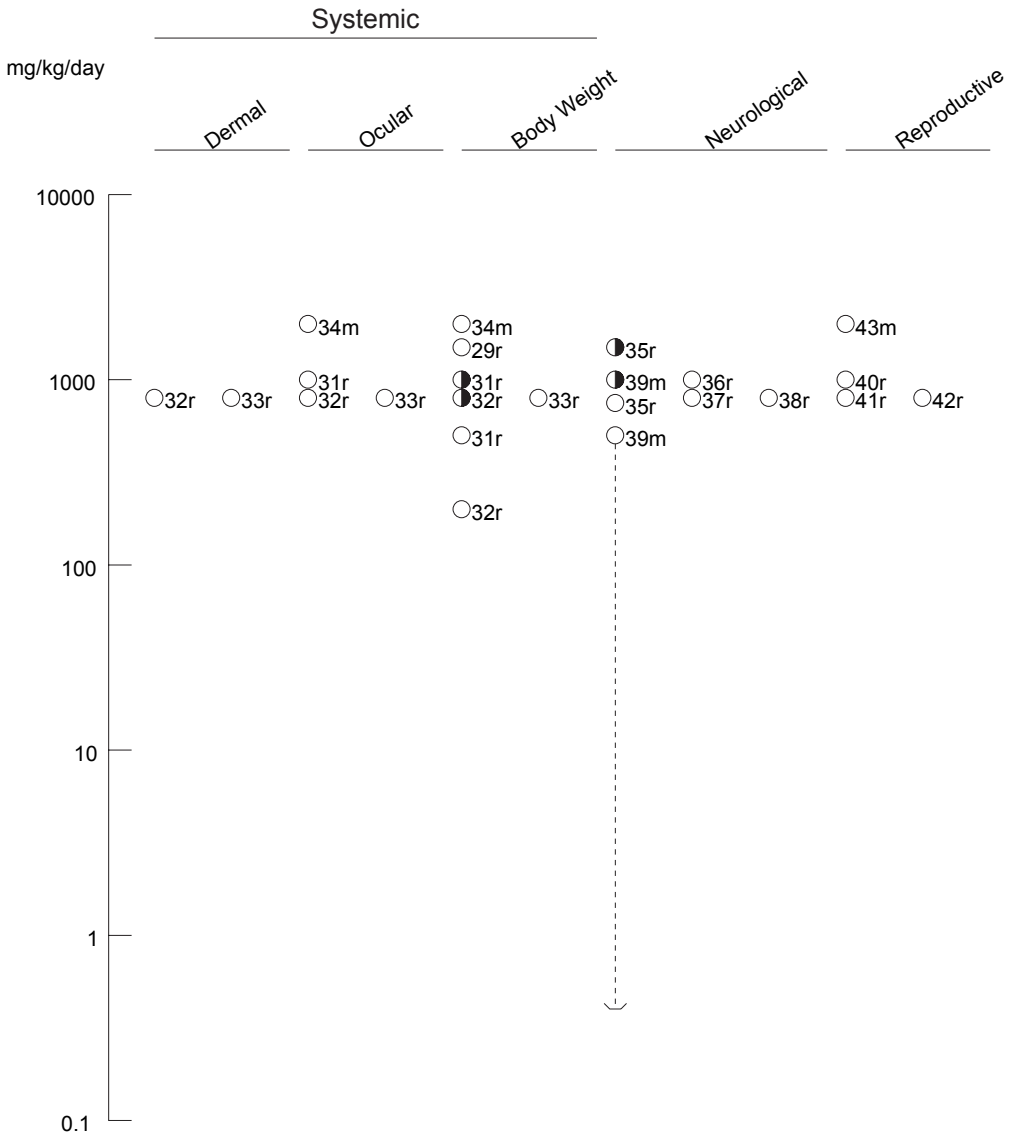
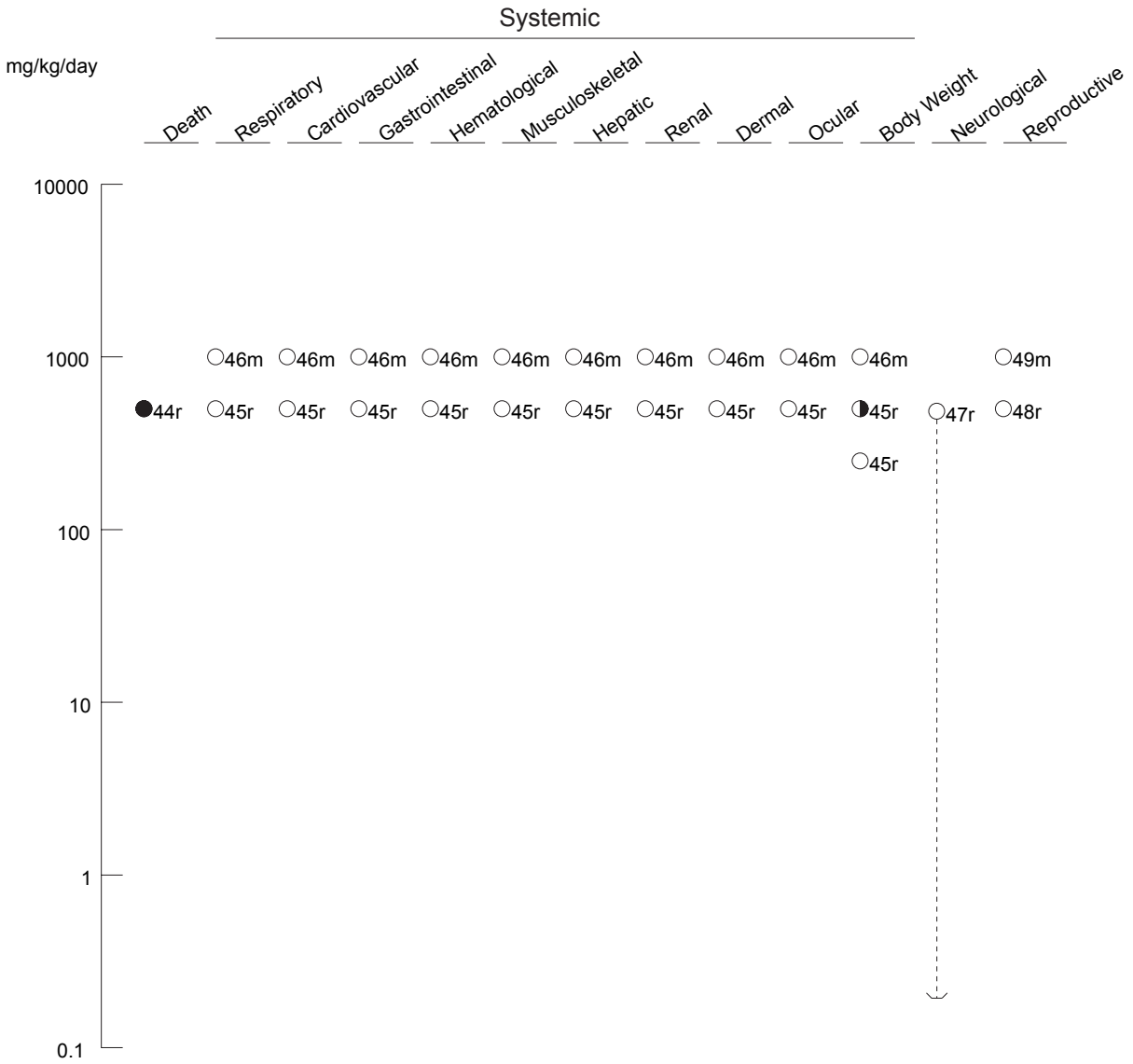


Figure 3-2 Levels of Significant Exposure to Xylene - Oral *(Continued)*

Chronic (≥365 days)



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

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

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

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

daily oral doses of 2,000 mg/kg/day, 5 days/week, for 13 weeks exhibited similar effects 15–60 minutes after dosing (NTP 1986). Histopathological examination of the lungs and mainstem bronchi of rats and mice administered mixed xylene 5 days/week at doses as high as 1,000 mg/kg in rats and 2,000 mg/kg in mice for 13 weeks or 500 mg/kg in rats and 1,000 mg/kg in mice for up to 2 years revealed no adverse effects (NTP 1986). Gross and histopathological examination of rats administered *m*- or *p*-xylene for 13 weeks at doses as high as 800 mg/kg/day revealed no treatment-related effects (Wolfe 1988a, 1988b).

Decreased pulmonary microsomal enzyme activity was observed in rats after a single oral dose of 1,000 mg/kg of *p*-xylene (Patel et al. 1978) and decreased pulmonary cytochrome P-450 content were observed in rats after gavage dosing with 800 mg/kg/day, 5 days/week, for 3 weeks (Elovaara et al. 1989), suggesting some direct toxicity of xylene in the lungs. Selective inactivation of enzymes can result in damage to tissue caused by the toxic metabolite of xylene, a methylbenzaldehyde (Carlone and Fouts 1974; Patel et al. 1978; Smith et al. 1982). The formation of the methylbenzaldehydes has not been confirmed in humans.

Cardiovascular Effects. Limited information was located regarding cardiovascular effects in humans following oral exposure to mixed xylene or its isomers. Postmortem examination showed no adverse effects on the heart or coronary arteries of a man who committed suicide by ingesting a large but unknown quantity of xylene (Abu Al Ragheb et al. 1986). No adverse cardiovascular effects were noted following histopathological examination of the heart in rats and mice exposed 5 days/week to mixed xylene at ≈63–2,000 mg/kg for 13 or 103 weeks (NTP 1986). No treatment-related effects were noted upon gross or histopathological examination of the heart in rats administered *m*- or *p*-xylene at doses as high as 800 mg/kg/day for 13 weeks (Wolfe 1988a, 1988b).

Gastrointestinal Effects. No superficial erosions, deep ulcerations, or other lesions were observed during postmortem examination of the gastric mucosa of a person who died following ingestion of a "large quantity" of xylene (Abu Al Ragheb et al. 1986). Histopathological examination of rats administered doses 5 days/week as high as 1,000 mg/kg of mixed xylene and mice administered doses as high as 2,000 mg/kg of mixed xylene for 13 weeks or in rats and mice administered doses as high as 500 and 1,000 mg/kg, respectively, for 2 years revealed no adverse effects on the stomach, small intestine, or colon (NTP 1986). Administration of *p*-xylene up to 800 mg/kg/day for 13 weeks also had no significant effect on gastrointestinal organs of rats (Wolfe 1988a, 1988b).

Hematological Effects. No studies were located regarding hematological effects in humans following oral exposure to mixed xylene or xylene isomers. Exposure of rats to 2,000 mg/kg/day *p*-xylene for 10 days resulted in no effects detectable in routine hematological analysis (Condie et al. 1988). Exposure to *o*- and *m*-xylene at 2,000 mg/kg/day for 10 days produced a decrease in the spleen weight of male rats (Condie et al. 1988); however, hematological analyses in these rats were normal. Mild polycythemia and leukocytosis in both male and female rats and an increase in spleen weight in females were observed in rats exposed to 1,500 mg/kg/day mixed xylene for 90 days (Condie et al. 1988). No effects were observed upon histopathological examination of the bone marrow following exposure to 800 mg/kg/day of *p*-xylene in rats and mice (Wolfe 1988a), or on administration 5 days/week of 1,000 mg/kg mixed xylene in rats for 13 weeks (NTP 1986), 2,000 mg/kg mixed xylene in mice for 13 weeks (NTP 1986), or 500 mg/kg mixed xylene in rats and 1,000 mg/kg in mice for 2 years (NTP 1986).

Musculoskeletal Effects. No studies were located regarding musculoskeletal effects in humans following oral exposure to mixed xylene or xylene isomers. In two animal bioassays, no musculoskeletal effects were observed in rats and mice upon histopathological examination of the femur, sternebrae, or vertebrae following intermediate or chronic exposure 5 days/week to mixed xylene up to 2,000 mg/kg for mice and 1,000 mg/kg for rats for 13 weeks and 1,000 mg/kg for mice or 500 mg/kg for rats for 103 weeks (NTP 1986). No adverse effects were observed in the sternum (with marrow), thigh musculature, or femur upon histopathological examination of rats administered *m*- or *p*-xylene at doses up to 800 mg/kg/day for 13 weeks (Wolfe 1988a, 1988b).

Hepatic Effects. No studies were located regarding hepatic effects in humans following oral exposure to mixed xylene or xylene isomers. In general, studies in animals have shown mild changes in the liver in response to oral exposure to mixed xylene. These changes included increased activity of liver enzymes and ultrastructural changes indicative of increased metabolic activity, but no evidence of histopathological changes in the liver tissue (Ungvary 1990). In acute and intermediate studies with rats, oral exposure to mixed xylene (Condie et al. 1988; Ungvary 1990) and its isomers (Condie et al. 1988; Elovaara et al. 1989; Pyykko 1980) has been associated with hepatic enzyme induction and increased hepatic weight. In the study by Condie et al. (1988), acute exposure to *p*-xylene at 250 mg/kg/day and *m*- and *o*-xylene at 1,000 mg/kg/day for 10 days caused increases in liver weight. Administration of doses of 1,060 mg/kg/day of all three xylene isomers for 3 days also produced increased cytochrome b_5 content and increased activities of liver enzymes in rats (Pyykko 1980), with the different isomers showing different enzyme induction potencies. Increased liver weight was observed with *m*- and

o-xylene, but not *p*-xylene, and increased cytochrome P-450 was observed only with *m*-xylene. Administration of mixed xylene to rats for 90 days caused 13.8–27% increases in relative liver weight ratios at doses between 750 and 1,500 mg/kg/day (Condie et al. 1988); a statistically significant 5.7% increase in relative liver weight in male rats at 150 mg/kg/day is not biologically significant. No treatment-related histopathological changes were observed in the liver, but mild increases in serum transaminases were observed at 750 mg/kg/day. Similar increases in serum alanine aminotransferase were observed following ingestion of 800 mg/kg/day of *m*-xylene for 3 or 13 weeks (Elovaara et al. 1989; Wolfe 1988a). No effects were noted upon histopathological examination of the liver of rats and mice that were administered mixed xylene 5 days/week for a chronic or intermediate period of time with doses as high as 2,000 mg/kg for mice; 1,000 mg/kg for rats for 13 weeks; and 1,000 mg/kg for mice and 500 mg/kg for rats for 103 weeks (NTP 1986). Administration of doses as high as 800 mg/kg/day of *p*-xylene in rats for 13 weeks produced no adverse hepatic effects (Wolfe 1988b).

Renal Effects. No studies were located regarding renal effects in humans following oral exposure to mixed xylene or xylene isomers. Data are available for renal effects in laboratory animals following acute-duration exposure to individual isomers, but not to mixed xylene (Condie et al. 1988; Pyykko 1980). At 1,060 mg/kg/day for 3 days, increases in kidney weight were observed with *m*-xylene and increases in microsomal enzyme content and activity were observed with all three isomers (Pyykko 1980). No effects on urine parameters were noted after a 10-day exposure to 2,000 mg/kg/day of any of the isomers (Condie et al. 1988). The majority of studies using mixed xylene or its isomers for intermediate or chronic durations also showed no adverse effects on the kidneys. The only toxic change observed was increased hyaline droplet change in males (not relevant to humans) and increased early chronic nephropathy in females at 150–1,500 mg/kg/day mixed xylene for 90 days; the incidence in females at 150 mg/kg/day was not statistically significant. Although urine from these rats were normal (Condie et al. 1988), continued hyaline droplet accumulation can result in cell damage. Increased relative kidney weight was also observed in male rats given mixed xylene at 750 mg/kg/day and in female rats at 1,500 mg/kg/day for 90 days (Condie et al. 1988). Similarly, increased kidney weight and microsomal enzyme activity were observed in rats exposed to 800 mg/kg/day *of m*-xylene, 5 days/week, for 3 weeks (Elovaara et al. 1989). Increased relative kidney weight was also observed in male rats administered 800 mg *m*-xylene/kg/day for 13 weeks (Wolfe 1988a, 1988b). Histopathology of the kidneys and urinary bladder were normal. Also, no adverse effects were noted upon histopathological examination of the kidneys of rats and mice following intermediate or chronic exposure 5 days/week to doses of mixed xylene as high as 2,000 mg/kg (for 13 weeks in mice) and 1,000 mg/kg (for 103 weeks in mice) (NTP 1986).

Endocrine Effects. No studies were located regarding endocrine effects in humans or animals following oral exposure to mixed xylene or xylene isomers.

Dermal Effects. No studies were located regarding dermal effects in humans following oral exposure to mixed xylene or xylene isomers. Limited information was located regarding dermal effects in animals. No adverse effects were noted during microscopic examination of the skin of rats and mice administered mixed xylene 5 days/week at doses as high as 2,000 mg/kg in mice and 1,000 mg/kg for rats for an intermediate (13 weeks) period of time or as high as 1,000 mg/kg for mice and 500 mg/kg for rats for a chronic (103 weeks) period of time (NTP 1986). The skin of rats administered doses as high as 800 mg/kg/day of *m*- or *p*-xylene for 13 weeks appeared normal upon histopathological examination (Wolfe 1988a, 1988b).

Ocular Effects. No studies were located regarding ocular effects in humans following oral exposure to mixed xylenes or xylene isomers. Histopathological examination of the eyes of rats and mice orally exposed to mixed xylenes (NTP 1986) or to *m*- or *p*-xylene (Wolfe 1988a, 1988b) for 13 or 103 weeks showed no effects. No additional data regarding ocular effects in animals following oral exposure to xylenes were available.

Body Weight Effects. Effects on body weight were observed in several acute studies of the effects of mixed xylene and its isomers (Condie et al. 1988; NTP 1986; Pyykko 1980). Exposure to *m*-, *o*-, and *p*-xylene for 3 days resulted in weight losses of between 2.5 and 3 times that observed in control rats (Pyykko 1980). A 14-day exposure of rats and mice to mixed xylene resulted in an 18% decrease in body weight gain in male rats at 1,000 mg/kg/day and an 89% decrease in bodyweight gain in male mice at 2,000 mg/kg/day (NTP 1986). Body weights of male rats given 2,000 mg/kg/day *o*- or *p*-xylene, but not *m*-xylene, for 10 days showed 14% and 13% decreases, respectively, relative to controls (Condie et al. 1988). No significant body weight changes were observed in male or female rats dosed daily with ≤1,500 mg/kg/day mixed xylenes for 13 weeks (Condie et al. 1988), but a 16% decrease was noted in female mice dosed 5 days/week for 13 weeks at 2,000 mg/kg (NTP 1986). Body weights were decreased by 25% in males (with reduced food consumption) and by 15.5% in females (with normal food consumption) treated by daily oral gavage with 800 mg *m*-xylene/kg/day for 13 weeks (Wolfe 1988a). In a parallel study with *p*-xylene, body weights were reduced by 21% in males and by 11% in females treated at 800 mg/kg/day, although food consumption was significantly higher than controls (Wolfe 1988b). Body weight in male rats at 1,000 mg/kg and in female mice at 2,000 mg/kg mixed xylene were

decreased 15% and 16%, respectively, in a 13-week study (NTP 1986). After week 59 in a 2-year study, mean body weights were reduced by 5–8% in male rats exposed by oral gavage 5 days/week to 500 mg/kg mixed xylenes, but this change is not biologically significant (NTP 1986). No body weight effects were observed in female rats exposed at doses as high as 500 mg/kg or in mice exposed at doses as high as 1,000 mg/kg/day (NTP 1986).

3.2.2.3 Immunological and Lymphoreticular Effects

No studies were located regarding immunological or lymphoreticular effects in humans after oral exposure to mixed xylene or xylene isomers. The only information suggesting a possible toxic effect of mixed xylene or its isomers on the immune system was a decrease in spleen and thymus weight observed in rats exposed for 10 days to 2,000 mg/kg/day *p*-xylene (Condie et al. 1988). Organ weight changes were not accompanied by histopathological changes.

The NOAEL and LOAEL values for immunological effects of *p*-xylene in rats are recorded in Table 3-2 and plotted in Figure 3-2.

3.2.2.4 Neurological Effects

Information concerning possible neurological effects associated with the ingestion of xylene is limited. Xylene produced a coma that persisted for more than 26 hours in a person who accidentally ingested an unknown amount (Recchia et al. 1985). The composition of the xylene was also unknown.

Clinical signs consistent with central nervous system toxicity have been observed in rats and mice following oral exposure to mixed xylene. A single oral dose of 4,000 mg/kg caused incoordination, prostration, decreased hindleg movement, and hunched posture in rats and tremors, prostration, and/or slowed breathing in mice (NTP 1986). In 13-week gavage assays in rats, neurotoxic signs included hyperactivity, convulsions, salivation, and epistaxis following exposure to 800 mg/kg/day *m*- or *p*-xylene (Wolfe 1988a, 1988b) and increased aggression following exposure to 1,500 mg/kg/day mixed xylenes (Condie et al. 1988). Clinical signs observed in mice in the hour after gavage dosing with mixed xylenes included weakness, lethargy, unsteadiness, tremors, and partial paralysis of hindlimbs at 2,000 mg/kg in a 13-week assay and hyperactivity at 1,000 mg/kg beginning week 4 in 13-week and 2-year assays (NTP 1986). Mild sedation at 2,000 mg/kg and increases in latency of several peaks in flash-evoked potentials at doses of 250 mg/kg and higher were observed following single doses of *p*-xylene; no effect was seen at

125 mg/kg/day (Dyer et al. 1988). This NOAEL was used to derive an MRL of 1 mg/kg/day for acute oral exposure to mixed xylenes or individual xylene isomers.

Histological damage to the outer hair cells of the organ of Corti provided evidence of ototoxicity in rats exposed by oral gavage to *p*-xylene, but not *m*- or *o*-xylene, at a dose of 900 mg/kg/day, 5 days/week for 2 weeks (Gagnaire and Langlais 2005). The losses of hair cells occurred in the area of the cochlea responsive to medium frequencies (10–25 kHz). No histopathology of the brain or spinal cord was observed in rats or mice administered mixed xylenes 5 days/week at doses as high as 1,000 mg/kg (rats) or 2,000 mg/kg/day (mice) for 13 weeks or 1,000 mg/kg for 2 years (NTP 1986). Similarly, no neurohistopathology was observed in rats administered doses of *m*- or *p*-xylene as high as 800 mg/kg/day for 13 weeks, although the brain-to-body weight ratio was increased in males dosed with 800 mg/kg/day of *m*-xylene (Wolfe 1988a, 1988b).

An acute-duration oral MRL of 1 mg/kg/day for mixed xylenes or individual isomers was calculated based on a NOAEL of 125 mg/kg and a LOAEL of 250 mg/kg for altered visual evoked brain potentials in male Long-Evans rats given single gavage doses of *p*-xylene (Dyer et al. 1988; NTP 1986) as described in the footnote in Table 3-2. An intermediate-duration oral MRL of 0.4 mg/kg/day for mixed xylenes or individual isomers was calculated based on a NOAEL of 500 mg/kg and a LOAEL of 1,000 mg/kg for hyperactivity in male and female B6C3F_1 mice dosed with mixed xylenes by gavage 5 days/week for 4–51 weeks (NTP 1986) as described in the footnote in Table 3-2.

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

3.2.2.5 Reproductive Effects

No studies were located regarding reproductive effects in humans following oral exposure to mixed xylene or individual isomers.

No studies in animals directly examining reproductive function following oral administration of mixed xylene or its isomers were located; however, histological examination of rats and mice administered mixed xylene 5 days/week at doses as high as 1,000 mg/kg in rats and 2,000 mg/kg in mice for 13 weeks revealed no adverse effects on the prostate/testes (male), ovaries/uterus, or mammary glands (female) (NTP 1986). The reproductive system organs of rats administered doses of *m*- or *p*-xylene as high as

800 mg/kg/day appeared comparable to controls after 13 weeks of treatment (Wolfe 1988a, 1988b). In chronic studies, no adverse histopathological changes were observed in the reproductive organs in rats at doses as high as 500 mg/kg and in mice at doses as high as 1,000 mg/kg administered 5 days/week for 103 weeks (NTP 1986). The highest NOAEL value from each reliable study for reproductive effects are recorded in Table 3-2 and plotted in Figure 3-2.

3.2.2.6 Developmental Effects

No studies were located regarding developmental effects in humans following oral exposure to mixed xylene or xylene isomers.

Significantly increased incidences of cleft palate and decreased fetal body weight were reported following maternal oral exposure during gestation days 6–15 to doses of 2,060 mg/kg/day mixed xylene in mice (Marks et al. 1982). Mixed xylene was also toxic to the dams, producing 31.5% mortality at 3,100 mg/kg/day. It is unclear whether the observation of cleft palate in this study is associated with maternal toxicity or a predisposition of mice under stress to give birth to offspring with this birth defect. In a teratology screening study, 2,000 mg/kg/day of *m*-xylene produced no evidence of fetal toxicity in mice (Seidenberg et al. 1986). Given the limited amount of animal data, no conclusion can be made regarding the relationship between oral exposure of xylene and adverse developmental effects. The highest NOAEL value and all LOAEL values from each reliable study for developmental effects are recorded in Table 3-2 and plotted in Figure 3-2.

3.2.2.7 Cancer

No data were located regarding cancer in humans following oral exposure to mixed xylene or xylene isomers.

The carcinogenicity of mixed xylene following oral exposure has been evaluated in chronic studies with rats and mice; however, no animal studies were available on the carcinogenic effects of *m*-, *o*-, or *p*-xylene following oral exposure. Results of the chronic oral studies with mixed xylene have been negative (NTP 1986) or equivocal (Maltoni et al. 1983, 1985). In a chronic bioassay, rats and mice of both sexes received mixed xylene by gavage at 0, 250, or 500 mg/kg and 0, 500, or 1,000 mg/kg, respectively, 5 days/week for 103 weeks. The interpretation of the results of the NTP bioassay was compromised by the large number of gavage-related deaths early in the study in the high-dose male rats. In the other chronic study (Maltoni et al. 1983, 1985), male and female rats were fed xylene (unspecified)

by gavage at 0 or 500 mg/kg, 4–5 days/week for 104 weeks. The Maltoni studies were weakened because of methodological flaws such as failure to report site-specific neoplasia, insufficient toxicity data, and absence of statistical analyses. Therefore, given the limited data, no definitive conclusion can be made regarding the carcinogenicity of mixed xylene in animals following oral exposure.

3.2.3 Dermal Exposure

In addition to studies that have directly examined the health effects of dermal exposure to xylene, a number of reports of health effects resulting from occupational exposure to xylene have been included in this section. Dermal contact with xylene is likely in many occupational situations, and absorption of xylene has been demonstrated in humans (Engstrom et al. 1977; Riihimaki 1979b; Riihimaki and Pfaffli 1978). The results of the occupational studies must be interpreted with caution, however, because of coexposure to other compounds.

3.2.3.1 Death

No reports of death in humans following dermal exposure to xylene were located. Limited animal data suggest that mixed xylene and *m*-xylene can cause death when applied dermally (Hine and Zuidema 1970; Smyth et al. 1962). The acute dermal LD_{50} in rabbits has been determined to be 3,228 mg/kg/day for *m*-xylene and >114 mg/kg/day for mixed xylene for 4 hours or more (Hine and Zuidema 1970; Smyth et al. 1962).

The LD_{50} value for death in rabbits as a result of acute-duration exposure to *m*-xylene is recorded in Table 3-3.

3.2.3.2 Systemic Effects

No studies were located regarding musculoskeletal, endocrine, or body weight effects in humans or animals following dermal exposure to mixed xylenes or xylene isomers. The systemic effects that were observed after dermal exposure to xylene are discussed below. All LOAEL values from each reliable study for systemic effects in each species and duration category are recorded in Table 3-3.

Respiratory Effects. Case reports of dryness of the throat (Goldie 1960) in painters and decreased pulmonary function and dyspnea in histology technicians with chronic exposure to xylene (Hipolito 1980) have been published. It is likely that these effects represent direct effects of xylene or other solvents on

Table 3-3 Levels of Significant Exposure to Xylene - Dermal

Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL	LOAEL				Reference Chemical Form	Comments
				Less Serious			Serious		
ACUTE EXPOSURE									
Death									
Mouse Hall	once					57 mg/kg	(8/120 died)	Pound and Withers 1963 mixed	
Rabbit Albino New Zealand	24 hr					3228 M mg/kg	(LD50)	Smyth et al. 1962 meta	
Systemic									
Rat (hairless)	4 d 5 x/d	Dermal		66 mg/day	(2-fold increase in transepidermal water loss, moderate erythema 5-fold increase in pro-inflammatory cytokine TNF-alpha)			Ahaghotu et al. 2005; Singh 2006	66 mg/day delivered in 5 fractions of 13.2 mg.
Rat (hairless)	1 hr	Dermal		199 M mg	(2-fold higher transepidermal water loss, 30% reduced moisture content, moderate erythema, 2.5-fold increase in pro-inflammatory TNF-alpha)	ortho		Chatterjee et al. 2005 meta	Occlusive exposure.

Table 3-3 Levels of Significant Exposure to Xylene - Dermal (continued)

Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL	LOAEL			Reference Chemical Form	Comments
				Less Serious		Serious		
Rat (hairless)	4 d 5 x/d	Dermal		64.8 M mg/day	(2-fold increase in transepidermal water loss, 22% reduced wate content, moderate erythema, 5.7-fold increase in pro-inflammatory TNF-alpha)		Chatterjee et al. 2005 meta	Unocclusive dermal exposure. 64.8 mg/day delivered in 5 fractions of 12.96 mg.
Rat (Fischer- 344)	1 hr once	Dermal		1200 M mg/kg	(separation of epidermis and dermis)		Gunasekar et al. 2003 meta	
Mouse (BALB/c)	once	Dermal		3136 F µg/kg	(skin edema)		Iyadomi et al. 2000 meta	
Mouse Hall	once	Dermal		57 mg/kg	(edema, irritation, scaliness of skin)		Pound and Withers 1963 mixed	
Gn Pig Dunkin Hartley	3 d 3 x/d	Dermal		2.3 F mg/kg/day	(skin irritation)		Anderson et al. 1986 mixed	
Rabbit NS	once	Ocular		87 mg	(mild irritation of eyes)		Consumer Product Testing 1976	0.1 mL/eye.

Table 3-3 Levels of Significant Exposure to Xylene - Dermal (continued)

Species (Strain)	Exposure/ Duration/ Frequency (Route)	System	NOAEL	LOAEL				Reference Chemical Form	Comments
				Less Serious		Serious			
Rabbit New Zealand white	once	Dermal				114 M mg/kg	(moderate to severe skin irritation)	Hine and Zuidema 1970 mixed	0.1 mL/eye; 0.5 mL to skin.
		Ocular		87 M mg	(moderately irritating to the conjunctiva)				
Rabbit New Zealand white	once	Ocular		87 M mg	(eye irritation)			Hine and Zuidema 1970 mixed	0.1 mL/eye.
Rabbit (New Zealand)	once	Ocular		87 mg	(minimal eye irritation)			Kennah et al. 1989 mixed	0.1 mL/eye.
Rabbit New Zealand albino	once	Ocular		432 mg	(eye irritation)			Smyth et al. 1962 meta	0.5 mL/eye.
Rabbit Albino	once	Dermal		2.3 mg/kg	(skin irritation)			Smyth et al. 1962 meta	

d = day(s); F = Female; hr = hour(s); LD50 = lethal dose, 50% kill;LOAEL = lowest-observed-adverse-effect level; M = male; NOAEL = no-observed-adverse-effect level; TNF = tumor necrosis factor; x = time(s)

the respiratory tissues and they are discussed in more detail in Section 3.2.1.2. No studies were located regarding respiratory effects in animals following dermal exposure to mixed xylene or xylene isomers, although some of the inhalation studies also involved exposure via dermal route as well.

Cardiovascular Effects. Cases of flushing, chest pains, and palpitations in histology technicians have been reported (Hipolito 1980). These studies also involved exposure via inhalation route. It is unclear whether these effects are directly attributable to xylene exposure because of possible exposure to other chemicals. No studies were located regarding cardiovascular effects in animals after dermal exposure to mixed xylene or xylene isomers.

Gastrointestinal Effects. Gastric discomfort in painters (Goldie 1960) and nausea in histology technicians (1980) have been reported; these studies also involved exposure via inhalation route. However, other chemicals in the workplace may have contributed to these effects. No studies were located regarding gastrointestinal effects in animals following dermal exposure to mixed xylene or xylene isomers.

Hematological Effects. Decreased white blood cell count has been observed in histology technicians (Hipolito 1980) and in workers with occupational exposure to benzene, toluene, and xylene (Moszczynski and Lisiewicz 1983, 1984a); these studies also involved exposure via inhalation route. However, chemicals other than xylene may have caused these decreases. No studies were located regarding hematological effects in animals following dermal exposure to mixed xylene or xylene isomers.

Hepatic Effects. When compared with unexposed controls, workers with occupational exposure via dermal and inhalation routes to toluene, xylene, and pigments had significantly increased urinary D-glucaric acid content in the urine indicating hepatic microsomal enzyme induction (Dolara et al. 1982). Serum antipyrine half-life was increased, suggesting possible hepatotoxicity. No effect on serum aminotransferases was observed in workers exposed to a mixture of solvents (Kurppa and Husman 1982). These studies also involved exposure via inhalation route. These studies are limited in that multiple chemical exposures occurred and the effects observed cannot be directly attributed to xylene. No studies were located regarding hepatic effects in animals following dermal exposure to mixed xylene or xylene isomers.

Renal Effects. Occupational exposure to a mixture of mainly xylene and toluene resulted in elevated albumin, erythrocytes, and leukocytes in the urine (Askergren 1981, 1982). In addition, increased

β-glucuronidase was observed in the urine of painters (Franchini et al. 1983). However, these studies are limited in that the effects observed may be attributable to exposure to toluene, which is a known renal toxicant, and that inhalation as well as dermal exposure could have occurred. No studies were located regarding renal effects in animals following dermal exposure to mixed xylene or xylene isomers.

Dermal Effects. Acute dermal exposure of human subjects to undiluted *m*-xylene in hand immersion studies has been associated with transient skin erythema (irritation), vasodilation of the skin, and dryness and scaling of the skin (Engstrom et al. 1977; Riihimaki 1979b). Urticaria was reported in a female cytology worker exposed predominantly to xylene vapors (Palmer and Rycroft 1993). Because this response probably had an immunological component, it is discussed further in the Immunological and Lymphoreticular Effects section.

Mild-to-severe skin irritation was noted in rabbits, guinea pigs, and mice treated topically with mixed xylene (2.3–114 mg/kg/day), *m*-xylene (65–199 mg/day) or *o*-xylene (66 mg/day) in acute studies (Ahaghotu et al. 2005, isomer identified in personal communication by Singh 2006; Anderson et al. 1986; Chatterjee et al. 2005; Consumer Products Testing 1976; Food and Drug Research Labs 1976a; Hine and Zuidema 1970; Pound and Withers 1963; Smyth et al. 1962). The extent of the irritation appeared to increase with duration of exposure; the most severe dermal irritation ratings were obtained in the longest exposures of 10-days (Hine and Zuidema 1970). Application of 0.250 mL (1,200 mg/kg) of *m*-xylene to the skin of rats for 1 hour resulted in a significant increase in oxidative species and DNA fragmentation (increased low-molecular-weight DNA) within 2 hours (Gunasekar et al. 2003; Rogers et al. 2001). Treatment with *m*- or *o*-xylene increased levels of inducible nitric oxide synthetase and tumor necrosis factor-alpha (TNF-alpha), a pro-inflammatory cytokine, in skin, as well as plasma levels of interleukin-1-alpha (IL-1alpha) (Ahaghotu et al. 2005; Chatterjee et al. 2005; Gunasekar et al. 2003). Dermal swelling following application of 69 mg *m*-xylene to the ear of mice resulted in significant increases in the thickness of the ear as measured 0.5–25 hours after application (Iyadomi et al. 2000); peak thickness was observed 6 hours after treatment. Histopathological effects of exposure to *m*- or *o*-xylene included swelling and disruption of the stratum corneum, granulocyte infiltration of the epidermis, and separation of the epidermis and dermis, with evidence of local inflammation (accumulation of mast cells, plasma cells) (Ahaghotu et al. 2005; Chatterjee et al. 2005; Gunasekar et al. 2003). Impairment of skin function was evident in the increase in transepidermal water loss in hairless rats following occlusive exposure (199 mg for 1 hour) or unocclusive exposure (64.8 mg/day in 5 divided daily fractions over 4 days) to *m*-xylene or unocclusive exposure (66 mg/day in 5 divided doses, for 4 days) to *o*-xylene and the reduction in skin moisture content observed in rats treated unocclusively as above with *m*-xylene

(Ahaghotu et al. 2005; Chatterjee et al. 2005). Moderate-to-marked irritation and moderate necrosis were observed in rabbits with a 2–4-week dermal exposure to undiluted xylene (Wolf et al. 1956). No chronic animal studies evaluating the dermal effects of xylene were located.

Ocular Effects. There are a few case reports in humans describing the effects of direct contact of the eye with heated xylene from a pressurized hose in one case or with paint containing xylene as the solvent in two cases (Ansari 1997; Narvaez and Song 2003). Both kinds of exposures resulted in photophobia, redness of the conjunctiva, and partial loss of the conjunctival and corneal epithelia. The effects in the two men exposed to paint were characterized as equivalent to grade II chemical burns and, with medical therapy, largely reversed after about a week (Ansari 1997). The other case also experienced subconjunctival hemorrhage, more extensive loss of the corneal epithelium, and xylene keratopathy (melting of the corneal stroma), the latter of which persisted to 4 weeks following the initial injury (Narvaez and Song 2003). It is possible combined thermal, physical, and chemical injury increased the severity of ocular effects in this case.

Several studies in humans have reported on ocular effects of exposure to xylene vapor. In the controlled exposure situations, eye irritation was reported at concentrations of mixed xylene as low as 200 ppm for 3–5 minutes (Nelson et al. 1943) and of *p*-xylene as low as 100 ppm for 1–7.5 hours/day for 5 days (NIOSH 1981). Case reports have also demonstrated transient eye irritation in humans exposed to vapors of mixed xylene or *p*-xylene (Carpenter et al. 1975a; Hastings et al. 1986; Klaucke et al. 1982; Nelson et al. 1943; Nersesian et al. 1985; NIOSH 1981). Eye irritation was more frequently reported by workers exposed to mixed xylene (geometric mean TWA 14 ppm) than by the controls (Uchida et al. 1993).

Instillation of 0.1 mL (87 mg) of mixed xylene or 0.5 mL (432 mg) of *m*-xylene into the eyes of rabbits resulted in slight-to-moderate eye irritation (Consumer Products Testing 1976; Hine and Zuidema 1970; Kennah et al. 1989; Smyth et al. 1962). No corneal effects were observed in these studies.

3.2.3.3 Immunological and Lymphoreticular Effects

Limited data were located regarding immunological and lymphoreticular effects in humans following dermal exposure to xylene. Occupational exposure to benzene, toluene, and xylene resulted in decreased serum complement (Smolik et al. 1973) and in decreased lymphocytes, but there was no effect on lymphocyte reactions when stimulated with phytohemagglutinin (Moszczynski and Lisiewicz 1983, 1984a). Interpretation of these studies is limited in that chemicals other than xylene may have accounted

for the effects observed. Exposures via inhalation and dermal routes also may have occurred. Contact urticaria was reported in a female cytology worker exposed for several months predominantly to <100 ppm xylene vapors (Palmer and Rycroft 1993). A closed patch test resulting in severe erythema and wealing provides evidence that the effect was a result of direct contact of xylene vapor with the skin and suggests that the reaction was immunological.

No studies were located regarding immunological or lymphoreticular effects in animals after dermal exposure to mixed xylene or xylene isomers.

3.2.3.4 Neurological Effects

Occupational exposure to xylene has been reported to result in headache, dizziness, malaise, a feeling of drunkenness, irritability, fine tremor, dysphasia, hyperreflexia, and/or impaired concentration and memory (Goldie 1960; Hipolito 1980; Kilburn et al. 1985; Roberts et al. 1988). These studies are limited, however, because other chemical exposures in the workplace may have been responsible for the effects observed and that exposures via inhalation and dermal routes may have occurred.

As described in a brief report, pregnant rats dermally exposed to xylene (form not specified) at 2,000 mg/kg/day throughout gestation showed a statistically significant 27% reduction, compared to controls, in motor activity in an open field test, suggesting a neurotoxic effect of xylene (Mirkova et al. 1979). In this study, dosing at 200 or 2,000 mg/kg/day reduced brain cholinesterase activities in dams by 35–38% compared to controls; fetal brain cholinesterase was also reported to be ‘inhibited’ at those doses. The report did not discuss whether any measures were taken to prevent ingestion of the test material.

3.2.3.5 Reproductive Effects

No studies were located regarding reproductive effects in humans or animals after dermal exposure to mixed xylene or xylene isomers.

3.2.3.6 Developmental Effects

The human data regarding the developmental effects of xylene suggest a possible relationship between occupational solvent exposure and developmental toxicity (Holmberg and Nurminen 1980; Kucera 1968; Taskinen et al. 1989; Windham et al. 1991). However, these data are limited for assessing the relationship between dermal exposure to xylene and developmental effects because the available studies

involved concurrent exposure to other chemical agents in addition to xylene in the workplace (Holmberg and Nurminen 1980; Kucera 1968; Taskinen et al. 1989; Windham et al. 1991), because few subjects were tested (Taskinen et al. 1989; Windham et al. 1991), and because it is extremely difficult to have a pure dermal exposure since such exposure in the absence of respiratory protection is accompanied by inhalation exposure.

As described in a brief report, dermal exposure of pregnant rats to doses as low as 200 mg/kg/day of xylene (unspecified concentration and isomer) throughout gestation produced decreases in enzyme activity (cholinesterase, cytochrome oxidase) in fetal and maternal brain tissue (Mirkova et al. 1979).

3.2.3.7 Cancer

Studies of workers occupationally exposed to solvents have examined the cancer and leukemia risks and suggest a possible relationship between coal-based xylene exposure and leukemia (Arp et al. 1983; Wilcosky et al. 1984). Both contain limitations (e.g., small number of subjects, no exposure concentrations, unknown composition of xylene and possible exposure to benzene and other chemicals) that preclude a definitive conclusion regarding dermal exposure to xylene and cancer; development of these studies probably also involved exposure via inhalation.

Limited information was located regarding the carcinogenicity of dermal exposure to xylene in animals (Berenblum 1941; Pound 1970; Pound and Withers 1963). Application of xylene (concentration, purity, and amount unspecified) to the skin for 25 weeks resulted in no increase in skin tumors, and did not potentiate the number of skin tumors produced by benz[a]pyrene (Berenblum 1941). However, two studies showed that a single xylene pretreatment slightly enhanced the number of tumors produced by a combination of ultraviolet light irradiation (initiation) and croton oil (promotion) (Pound 1970) or urethane (initiation) and croton oil (promotion) (Pound and Withers 1963). These findings suggest that xylene may be a promoter for skin cancer and might also act as initiator or cocarcinogen. These studies are limited in that tumors other than skin tumors were not assessed and untreated controls were not used. Furthermore, it is not known whether the xylene was analyzed for the presence of other aromatic hydrocarbons such as benzene that are known to be tumorigenic.

3.3 GENOTOXICITY

The preponderance of data from testing *in vivo* (Table 3-4) and *in vitro* (Table 3-5) indicates that xylenes are not mutagenic and do not induce chromosomal anomalies. A few studies indicate that DNA

Table 3-4. Genotoxicity of Xylene *In Vivo*

Species (test system)	End point	Exposure route; dose	Results	Reference	Isomer
Mammalian cells:					
Human peripheral lymphocytes	Sister chromatid exchange and chromosomal abberations	Inhalation (occupational exposure)	–	Haglund et al. 1980	Not reported
Human peripheral lymphocytes	Sister chromatid exchange	Inhalation (occupational exposure)	–	Pap and Varga 1987	Mixed xylene
Human peripheral lymphocytes	Sister chromatid exchange	Inhalation (three exposures; 40 ppm, 7 hours/day)	–	Richer et al. 1993	Mixed xylene
Mouse reticulocytes	Chromosomal aberrations or Micronuclei formation	Oral (single exposure); ≤1,000 mg/kg	–	Feldt 1986	Mixed xylene
Rat (Fischer 344) skin	DNA damage	Dermal (250 µL; 1 hour)	+	Rogers et al. 2001	*m*-Xylene
Rat bone marrow	Chromosomal aberrations	Intraperitoneal (single exposure)	–	Litton Bionetics 1978b	Mixed xylene (11.4% *o*-xylene, 0.3% *p*-xylene, 36.1% ethylbenzene)
Rat bone marrow	Chromosomal aberrations	Intraperitoneal (five exposures)	–	Litton Bionetics 1978b	Mixed xylene (0.3% p-xylene, 36.1% ethyl-benzene)
Mouse bone marrow polychromatic-erythrocyte assay (micro-nucleus test)	Micronuclei formation	Intraperitoneal (two exposures at ≤650 mg/kg)	–	Mohtashami-pur et al. 1985	*m*-Xylene, *o*-xylene, *p*-xylene
Rat sperm-head morphology assay	Sperm-head abnormalities	Intraperitoneal (two exposures at 435 mg/kg)	–	Washington et al. 1983	*o*-Xylene

– = negative result
+ = positive result

Table 3-5. Genotoxicity of Xylene *In Vitro*

Species (test system)	End point	Results		Reference	Isomer
		With activation	Without activation		
Prokaryotic organisms:					
Salmonella typhimurium TA97, TA98, TA100, TA1535/ plate incorporation assay	Reverse Mutation	–	–	NTP 1986	Mixed xylene
S. typhimurium TA98, TA100, TA1535, TA1537/plate incorporation assay	Reverse Mutation	–	–	Haworth et al. 1983	*m*-Xylene; *o*-xylene; *p*-xylene
S. typhimurium TA98, TA100, UTH8414, UTH8413/plate incorporation assay	Reverse Mutation	–	–	Connor et al. 1985	*m*-Xylene; *o*-xylene; *p*-xylene
S. typhimurium TA98, TA100, TA1535, TA1537, TA1538/ plate incorporation assay	Reverse Mutation	–	–	Bos et al. 1981	*m*-Xylene; *o*-xylene; *p*-xylene
S. typhimurium TA98, TA100, TA1535, TA1537/spot and plate incorporation assays	Reverse Mutation	–	–	Florin et al. 1980	*m*-Xylene; *p*-xylene
S. typhimurium TA98, TA100, TA1535, TA1537, TA1538/ suspension and plate incorporation assays	Reverse Mutation	–	–	Litton Bionetics 1978b	Mixed xylene
S. typhimurium TA98, TA100, TA1535, TA1537, TA1538/ plate incorporation assay	Reverse Mutation	–	–	Shimizu et al. 1985	*p*-Xylene
S. typhimurium TA1535/ pSK1002 (*umu*C-*lac*Z gene expression)	SOS induction	–	–	Nakamura et al. 1987	Mixed xylene
Escherichia coli WP2uvrA/ plate incorporation assay	Mutation	–	–	Shimizu et al. 1985	*p*-Xylene
E. coli WP2 (λ) (Ionii, sulA1, trpE65, uvrA155, IamB+), microscreen prophage-induction assay	Mutation	–	–	DeMarini et al. 1991	Mixed xylene
E. coli WP2, WP2uvrA, WP67, CM611, WP100, W3110polA+, p3478pola-/ DNA repair microsuspension assay	DNA damage	–	–	McCarroll et al. 1981b	Not reported (technical grade)
Bacillus subtilis H17, M45/modified rec assay	DNA damage	–	–	McCarroll et al. 1981a	Not reported (technical grade)
Eukaryotic organisms:					
Saccharomyces cerevisiae D4/suspension and plate incorporation assays	Mitotic gene conversion	–	–	Litton Bionetics 1978b	Mixed xylene

Table 3-5. Genotoxicity of Xylene *In Vitro*

Species (test system)	End point	Results		Reference	Isomer
		With activation	Without activation		
Mammalian cells:					
Cultured mouse lymphoma cells (L5178Y, TK+/-)	Forward mutation	–	–	Litton Bionetics 1978b	Mixed xylene
Cultured human lymphocytes	Sister chromatid exchange and chromosomal aberrations	Not tested	–	Gerner-Smidt and Friedrich 1978	Not reported
Cultured human lymphocytes	Sister chromatid exchange	Not tested	–	Richer et al. 1993	Mixed xylene
Cultured human lymphocytes	DNA-damage (comet assay)	Not tested	+T	Morrozzi et al. 1999	Mixed xylene
Cultured Chinese hamster ovary cells	Sister chromatid exchange and chromosomal aberrations	–	–	Anderson et al. 1990	Mixed xylene
Cultured Syrian hamster embryo cells-Simian adenovirus SA7	Enhanced viral transformation		–	Casto 1981	Mixed xylene

– = negative result; +T = damage associated with cytotoxcity (14% viability)

fragmentation may occur if cells are exposed to levels that cause cytotoxicity, presumably related to the activity of nucleases within dying cells.

Limited human data are available regarding the genotoxic effects of mixed xylene following inhalation exposure. No inhalation studies were located regarding the genotoxicity of *m*-, *o*-, or *p*-xylene in humans and animals. Results of studies by Pap and Varga (1987) and Richer et al. (1993) suggest that inhalation exposure of humans to mixed xylene is not associated with the induction of sister chromatid exchanges or chromosomal aberrations in peripheral lymphocytes. Results of other investigations were also negative for chromosomal aberrations in humans or rats exposed by inhalation to xylene; however, as the isomeric composition of the xylene in these studies was not reported (Haglund et al. 1980; Zhong et al. 1980), it is difficult to assess the contribution of the individual isomers of xylene. The rat study was limited by the lack of details regarding exposure concentrations and duration of exposure.

No studies were located regarding genotoxic effects in humans after oral exposure to mixed xylene or xylene isomers. No chromosomal aberrations or change in the incidence of micronuclei were observed in reticulocytes isolated from mice receiving oral doses of xylenes as high as 1,000 mg/kg within a 24-hour period (Feldt 1986).

No studies were located regarding genotoxic effects in humans after dermal exposure to mixed xylene or xylene isomers. A significant increase in lower molecular weight genomic DNA was detected in the skin of rats that received a dermal application of 250 µL (1,200 mg/kg) *m*-xylene for 1 hour (Rogers et al. 2001). Electrophoretic patterns indicative of DNA fragmentation appeared 2 hours after exposure ended and coincided with the significant increase in oxidative species in skin cells. Although the level of oxidative species subsequently was reduced—the authors speculate as a result of endogenous antioxidant activity—lower molecular weight DNA levels remained significantly elevated at 4 and 6 hours after exposure. DNA fragmentation coincided with histopathological evidence of skin irritation, inflammation, and separation of the epidermis and dermis (Gunasekar et al. 2003).

The absence of genotoxic effects following parenteral *in vivo* exposure to xylene has been reported in the bone marrow chromosomal aberration test with rats (Litton Bionetics 1978b), the bone marrow micronucleus test with mice (Mohtashamipur et al. 1985), and the sperm morphology test with rats (Washington et al. 1983).

Mixed xylene, and the individual xylene isomers, have been tested for genotoxicity in a variety of short-term *in vitro* assays. Results of standard mutagenicity assays indicate that mixed xylene and xylene isomers are not mutagenic in bacteria, yeast, or mammalian cells (Bos et al. 1981; Connor et al. 1985; DeMarini et al. 1991; Florin et al. 1980; Haworth et al. 1983; Litton Bionetics 1978b; McCarroll et al. 1981a, 1981b; NTP 1986; Shimizu et al. 1985). Mixed xylenes did not induce *umu* gene expression, part of the SOS response to DNA damage, in *Salmonella typhimurium* TA1535/pSK1002 (Nakamura et al. 1987). Mixed xylene did not induce chromosomal anomalies (chromosomal aberrations or sister chromatid exchanges) in cultured mammalian cells (Anderson et al. 1990; Gerner-Smidt and Friedrich 1978; Richer et al. 1993). Increased DNA damage in human lymphocytes exposed to a single concentration of mixed xylene (5 μM) was associated with reduced cell viability (14% of controls) and likely caused by nuclease activity in the dying cells (Morozzi et al. 1999). Mixed xylenes did not enhance the adenovirus transformation of hamster embryo cells (Casto 1981).

No mutagenic activity was demonstrated for any of the various metabolites of xylene in bacterial test systems. *S. typhimurium* strains TA98, TA100, TA1535, TA1537, and TA1538, with and without S9 metabolic activation, have been used to test the mutagenic activity of *p*-xylenol (Epler et al. 1979; Florin et al. 1980; Hejtmankova et al. 1979; Pool and Lin 1982), *m*-xylenol (Epler et al. 1979; Florin et al. 1980), and *o*-methylbenzyl alcohol (Bos et al. 1981). 2,4-Dimethylphenol has been evaluated in a gene reversion assay with *Escherichia coli* strain Sd-4-73 (Szybalski 1958).

In summary, genotoxicity studies on mixed xylene and the individual isomers of xylene have provided consistently negative results in a variety of *in vitro* and *in vivo* assays and test systems (bacteria, yeast, insects, cultured mammalian cells, mice, rats, and humans). Thus, there is sufficient evidence to conclude that mixed xylene, *m*-xylene, *o*-xylene, and *p*-xylene are nonmutagenic. Xylenes may cause DNA fragmentation at cytotoxic concentrations because of nucleases released from lysosomes in moribund cells. There is also limited evidence from bacterial test systems that suggests that xylene metabolites, specifically *m*-xylenol, *p*-xylenol, 2,4-dimethylphenol, and *o*-methylbenzyl alcohol, are also nonmutagenic.

3.4 TOXICOKINETICS

Studies in humans and animals have shown that xylenes are well absorbed by the inhalation and oral routes. Approximately 60% of inspired xylene is retained and approximately 90% of ingested xylene is absorbed. Absorption of xylene also occurs by the dermal route, but to a much lesser extent than by the

inhalation and oral routes especially following exposure to xylene vapor. Following absorption, xylene is rapidly distributed throughout the body by way of the systemic circulation. In the blood, xylene is primarily bound to serum proteins. Xylene accumulates primarily in adipose tissue. All three isomers of xylene are primarily metabolized by oxidation of a methyl group and conjugation with glycine to yield the methylhippuric acid. In humans exposed to xylene, >90% of the absorbed xylene is excreted in the urine as the methylhippuric acid. Aromatic hydroxylation of xylene to xylenol occurs to only a limited extent in humans. Less than 2% of an absorbed dose is excreted in the urine as xylenol. Other minor metabolites found in urine include methylbenzyl alcohol and glucuronic acid conjugates of the oxidized xylene. Metabolism in animals is qualitatively similar, but glucuronide conjugates make up a larger proportion of the urinary excretion products (see Figures 3-3 and 3-4). In addition, methylbenzaldehyde (the product of the action of alcohol dehydrogenase on methylbenzyl alcohol) has been detected in animals, where it may exert toxic effects, but its presence has not been confirmed in humans. In humans, about 95% of the absorbed xylene is excreted in the urine, with about 5% excreted unchanged in the exhaled air. Elimination from most tissue compartments is rapid, with slower elimination from muscle and adipose tissue.

3.4.1 Absorption

3.4.1.1 Inhalation Exposure

Xylenes are very soluble in blood and are therefore absorbed easily into the systemic circulation during inhalation exposure (Astrand 1982). Evidence for absorption of xylene by humans following inhalation exposure is provided by the observation that urine metabolites increase in proportion to exposure (Inoue et al. 1993; Jonai and Sato 1988; Kawai et al. 1991; Ogata et al. 1970; Riihimaki and Pfaffli 1978; Riihimaki et al. 1979b; Sedivec and Flek 1976b; Senczuk and Orlowski 1978; Wallen et al. 1985) and in proportion to increased ventilatory rates during exercise (Astrand 1982; Astrand et al. 1978; Bergert and Nestler 1991; Engstrom and Bjurstrom 1978; Riihimaki and Savolainen 1980; Riihimaki et al. 1979b). Absorption of the retained isomers appears to be similar, regardless of exposure duration or dose. The alveolar air concentrations of xylenes in male volunteers exposed at rest to 100 ppm mixed xylenes for 70 minutes reached equilibrium within 10 minutes (Gamberale et al. 1978).

Many authors have measured the retention of xylene in the lungs following inhalation exposure. It is this retained xylene that is available for absorption into the systemic circulation. In experimental studies with human subjects, retention of the various isomers was similar following inhalation of *m*-, *o*-, or *p*-xylene, and averaged 63.6% (Sedivec and Flek 1976b). Other authors have estimated that between 49.8 and

72.8% of inhaled xylene is retained (David et al. 1979; Ogata et al. 1970; Riihimaki and Pfaffli 1978; Riihimaki and Savolainen 1980; Wallen et al. 1985). Pulmonary retention does not appear to differ on the basis of sex (Senczuk and Orlowski 1978). However, one study reported that uptake of *m*-xylene during a 2-hour exposure at 200 mg/m^3 was slightly higher in women than in men (Ernstgard et al. 2003). The enhanced pulmonary ventilation and cardiac output associated with physical exertion can increase the amount of xylene retained and subsequently absorbed into the body (Astrand et al. 1978; Riihimaki et al. 1979b). The study by Astrand et al. (1978) suggests that retention efficiency decreases as exposure duration increases.

In pregnant mice, approximately 30% of an administered inhalation dose of 600 ppm *p*-xylene was absorbed following a 10-minute exposure period (Ghantous and Danielsson 1986). Absorption was not quantified in other animal studies, but absorption can be inferred from the observed effects on pulmonary microsomal enzymes and the appearance of methylhippuric acid in the urine following inhalation of xylene (Carlsson 1981; David et al. 1979; Elovaara 1982; Elovaara et al. 1987; Patel et al. 1978).

3.4.1.2 Oral Exposure

Limited information is available on the absorption of xylene in humans and animals following ingestion. Excretion of urinary metabolites indicated that absorption had occurred following oral doses of either 40 or 80 mg/kg of *o*-xylene or *m*-xylene in humans (Ogata et al. 1979). Although absorption was not fully quantified, recovery of specific metabolites demonstrated that at least 34% of *o*-xylene and 53% of *m*-xylene had been absorbed from the administered doses of 40 mg/kg.

Measurement of urinary metabolites in rats over 24 hours demonstrated almost complete absorption (87–92%) following oral gavage dosing with 1.8 g *m*-xylene, or 1.74 g *o*- or *p*-xylene (Bray et al. 1949). Experiments in which 0.15 mL of radiolabelled *m*-xylene (0.27 mg/kg) was administered by oral gavage in 5% aqueous gum acacia to male and female rats indicated that absorption was rapid (Turkall et al. 1992); peak blood levels of radioactivity were observed within 20 minutes. The half-life of absorption of *m*-xylene was twice as fast in females than in males, although the total amount absorbed over 24 hours—the area under the curve (AUC) for plasma radioactivity—was the same in both sexes (about 0.2% of the initial dose/mL/hour). When *m*-xylene was first adsorbed to 0.5 g of a sandy soil before mixing with the vehicle and gavage administration, the absorption half-life was twice as long in female rats but unaffected in males (Turkall et al. 1992); adsorption to clay soil did not produce a biologically significant difference

from *m*-xylene administered alone. Adsorption to sandy soil doubled the peak blood levels of radioactivity and increased the plasma AUC by 75% in female rats, but had no such effects in males.

3.4.1.3 Dermal Exposure

Results of experimental studies with humans indicate that *m*-xylene is absorbed following dermal exposure; however, the extent of penetration and absorption of *m*-xylene through skin is not nearly as great as that resulting from inhalation (Engstrom et al. 1977; Riihimaki 1979b; Riihimaki and Pfaffli 1978). Dermal absorption may occur via exposure to *m*-xylene vapors, as well as through direct dermal contact with the solvent (Dutkiewicz and Tyras 1968; Engstrom et al. 1977; Kezic et al. 2000; Loizou et al. 1999; Riihimaki 1979b; Riihimaki and Pfaffli 1978). Absorption of *m*-xylene through the skin was measured at a maximal permeation rate, occurring between 15 and 25 minutes at a flux of 46 nmol/cm^2/minute (Kezic et al. 2001). In humans, the estimated absorption rate following immersion of both hands in *m*-xylene for 15 minutes was approximately 2 μg/cm^2/minute (Engstrom et al. 1977). Another study measured a rate of absorption of 75–160 μg/cm^2/minute when xylene was applied to forearm skin (Dutkiewicz and Tyras 1968). Absorption of *m*-xylene vapor through the skin has been estimated at between 0.1 and 2% of the inhaled dose (Kezic et al. 2000; Loizou et al. 1999; Riihimaki and Pfaffli 1978). Dermal permeability constants averaging 0.25 cm/hour were calculated from exposure of subjects to 600 ppm *m*-xylene for 3.5 hours (McDougal et al. 1990; Riihimaki and Pfaffli 1978). In human subjects exposed to 6770 ppm *m*-xylene vapor on the hand and forearm the average flux through the skin was 0.091 mg/L/hour for a 20-minute exposure, falling to 0.061 mg/L/hour for a 180-minute exposure (Kezic et al. 2004). Simultaneously, the maximal flux into the blood was 0.034 mg/L/hour after 20 minutes, rising to 0.063 mg/L/hour after a 180-minute exposure. Maximum permeation rates of *m*-xylene vapor were achieved within 90 minutes (Kezic et al. 2004). Dermal absorption of *m*-xylene following topical administration or exposure to vapor was 3 times greater in subjects with atopic dermatitis compared to normal subjects (Engstrom et al. 1977; Riihimaki and Pfaffli 1978).

Limited information is available regarding the absorption of xylene following dermal exposure in animals. Permeability of rat skin to *m*-xylene was estimated from blood levels obtained during dermal exposure to liquid *m*-xylene (Morgan et al. 1991; Skowronski et al. 1990) or *m*-xylene vapors (McDougal et al. 1990). Peak blood levels of *m*-xylene were reached within 2 hours of topical application from sealed dermal enclosures in rats and slowly declined during the 24-hour observation period (Morgan et al. 1991); an average of 562 mg (0.65 mL) was absorbed across a 3.1 cm^2 area of skin (1% of total surface area) from the initial volume of 2 mL over 24 hours. The total amount absorbed was reduced when

m-xylene was administered in aqueous solution (Morgan et al. 1991). Dermal permeability constants calculated for rats exposed to 5,000 ppm *m*-xylene averaged 0.723 cm/hour, about 3 times greater than those calculated for humans (McDougal et al. 1990); the flux was calculated as 0.0151 mg/cm^2/hour. Skin:air partition values for a series of solvents including *m*-xylene correlated well with permeability constants (McDougal et al. 1990), but not with octanol-water partition coefficients. *m*-Xylene adsorbed on sandy soil or clay soils showed lower peak absorption than for *m*-xylene alone and clay soil significantly prolonged the absorption half-life, but the total amount absorbed over an unspecified period was unchanged (Abdel-Rahman et al. 1993; Skowronski et al. 1990).

Dermal penetration rates of xylene have been estimated using excised skin of rats exposed to liquid or vapor. The absorption of *o*-xylene by excised abdominal skin from rats increased with the time of contact (Tsuruta 1982); the penetration rate was estimated to be 0.006 mg/cm^2/hour. The flux of *o*-xylene through the dorsal skin of hairless rats was measured as 0.22 mg/cm^2/hour (Ahaghotu et al. 2005). The skin:air partition coefficient for *m*-xylene was found to be 50.4±1.7 using rat skin *in vitro* exposed to 203 ppm *m*-xylene vapor (Mattie et al. 1994); the concentration of *m*-xylene in the skin reached equilibrium in 2 hours. Dermal absorption studies using excised skin are not directly comparable to *in vivo* studies because of the lack of an intact blood supply, but diffusion through the skin appears to be the limiting factor rather than removal from the skin by the blood (McDougal et al. 1990).

3.4.2 Distribution

Results of experiments to derive tissue:air partition coefficients for xylene indicate that the isomers are expected to have similar distributions in the body (Table 3-6). Factors influencing the distribution of xylene in blood are discussed in Section 3.5.1.

3.4.2.1 Inhalation Exposure

In human subjects exposed by inhalation to ≤44 ppm deuterium-labeled xylene isomers for 2 hours, peak blood xylene concentrations were highest exactly at the end of exposure and then declined (Adams et al. 2005). Absorbed xylene in venous blood was distributed in cells (12%) and serum (88%), with about 9% of the total amount in blood associated with protein-free serum (Riihimaki et al. 1979b). Following systemic circulation, xylene is distributed primarily to adipose tissue. Estimates of the amount of xylene accumulated in human adipose tissue range from 4 to 10% of the absorbed dose (Astrand 1982; Engstrom and Bjurstrom 1978; Riihimaki et al. 1979b). It has been suggested that following prolonged

Table 3-6. Partition Coefficients for Xylenes

Reference	Species	Partition coefficient[a]		
		m-Xylene	*o*-Xylene	*p*-Xylene
		Blood:Air		
Gargas et al. 1989	Rat (male F-344)	46±1.5	44.3±2.0	41.3±3.5
Kaneko et al. 1991a	Rat (male Wistar)	39.9±7.18[b]	NR	NR
Kumarathasan et al. 1998	Rat (male Sprague-Dawley)	40.3±1.4	NR	37.0±0.4
Thrall et al. 2002[c]	Rat		37.8	
Sato and Nakajima 1979	Human	26.4±0.9[b]	31.1±2.3[b]	37.6±3.5[b]
Pierce et al. 1996	Human	31.9±0.45	35.2±0.45	39.0±0.70
Thrall et al. 2002[c]	Human		34.0	
		Fat:Air		
Gargas et al. 1989	Rat (male F-344)	1,859±93	1,877±132	1,748±65
Kaneko et al. 1991a	Rat (male Wistar)	2,050±459[b]	NR	NR
Pierce et al. 1996	Rat (male F-344)	2,325±194	2,930±260	NR
Kumarathasan et al. 1998	Rat (male Sprague-Dawley)	1,970±34	NR	1,863±37
Pierce et al. 1996	Human	1,919±53	2,460±63	2,019±102
		Liver:Air		
Gargas et al. 1989	Rat (male F-344)	90.9±4.4	108±7	90.0±4.3
Kaneko et al. 1991a	Rat (male Wistar)	79.9±9.42[b]	NR	NR
Kumarathasan et al. 1998	Rat (male Sprague-Dawley)	66.8±6.6	NR	62.1±5.0
		Muscle:Air		
Gargas et al. 1989	Rat (male F-344)	41.9±5.7	51.5±6.7	38.4±4.1
Kaneko et al. 1991a	Rat (male Wistar)	79.7±20.2[b]	NR	NR
Kumarathasan et al. 1998	Rat (male Sprague-Dawley)	41.3±0.1	NR	43.21±3.4
		Milk:Air		
Fisher et al. 1997	Human	133±54.58 for *o-*, *m-*, *p*-xylenes		

[a]Mean ± standard error unless otherwise noted
[b]Mean ± standard deviation
[c]As reported in Thrall and Woodcock 2003

NR = not reported

occupational exposure to xylene, significant amounts of the solvent could accumulate in adipose tissue (Astrand 1982; Engstrom and Bjurstrom 1978).

Studies in mice (Bergman 1983; Ghantous and Danielsson 1986) and rats (Carlsson 1981; Ito et al. 2002) indicate that the distribution of *m*- or *p*-xylene and their metabolites is characterized by high uptake in lipid-rich tissues, such as brain and fat. High uptake also occurs in well-perfused organs, such as the liver and kidney. Using low-temperature (cryogenic) whole-body autoradiography of male mice exposed by inhalation to ^{14}C-labeled *m*-xylene for 10 minutes, Bergman (1983) observed a high level of unmetabolized *m*-xylene (volatile radioactivity) in body fat, bone marrow, white matter of the brain, spinal cord, spinal nerves, liver, and kidney immediately after exposure. High levels of metabolites (nonvolatile radioactivity) were present in the blood, liver, lung, kidney, and adrenal medulla. Unmetabolized *m*-xylene persisted in the central nervous system and spinal nerves for up to 1 hour post-exposure, whereas a considerable amount persisted in body fat at 4–8 hours after exposure. Metabolites were detectable post-exposure in the adrenal medulla for 0.5 hours, in the liver for 2 hours, in the kidney for up to 8 hours, in bile for 2–8 hours, in the intestinal lumen for up to 24 hours, and in the nasal mucosa and bronchi for 2–24 hours. No radioactivity was detectable by 48 hours. In male Sprague-Dawley rats exposed to 2,000 ppm *m*-xylene vapor 4 hours/day for 5 consecutive days, *m*-xylene concentrations were measured in tissues by gas chromatography (Ito et al. 2002). The highest concentrations of *m*-xylene were observed in peri-intestinal fat and subcutaneous fat, whereas concentrations more than 40 times lower were detected in lung, spleen, testis, heart, kidney, liver, and psoas muscle. In the brain subdivided into four parts, the concentration of *m*-xylene was 3–6 times lower than in subcutaneous fat. The uneven distribution of inhaled *m*-xylene in the brain was associated with changes in $GABA_A$ receptor binding (discussed in Section 3.5.2).

According to an intermediate-duration animal study, the level of xylene stored in body fat may decrease as exposure continues due to an increase in metabolic rate possibly by inducing its own metabolism (Savolainen et al. 1979a). Levels of *m*-xylene in perirenal fat of rats exposed to 300 ppm technical xylene decreased from 67.6 to 36.6 μg/g tissue as exposure duration increased from 5 to 18 weeks (Savolainen et al. 1979a).

p-Xylene and *o*-xylene have been shown to readily cross the placenta and were distributed in amniotic fluid and embryonic and fetal tissues (Ghantous and Danielsson 1986; Ungvary et al. 1980b). The level detected in fetal tissues (brain, liver, lung, and kidney), which are low in lipids, was only 2% of that

detected in the maternal brain tissue, which contains large amounts of lipids (Ghantous and Danielsson 1986). Also, higher levels were detected in fetal tissues than in amniotic fluid (Ungvary et al. 1980b).

3.4.2.2 Oral Exposure

No studies were located regarding distribution in humans following oral exposure to mixed xylene or xylene isomers. In rats administered *m*-xylene by gavage, fat contained the highest tissue concentration of radioactivity; approximately 0.3% of the administered dose was found per gram of fat in females and 0.1% per gram of fat in males (Turkall et al. 1992).

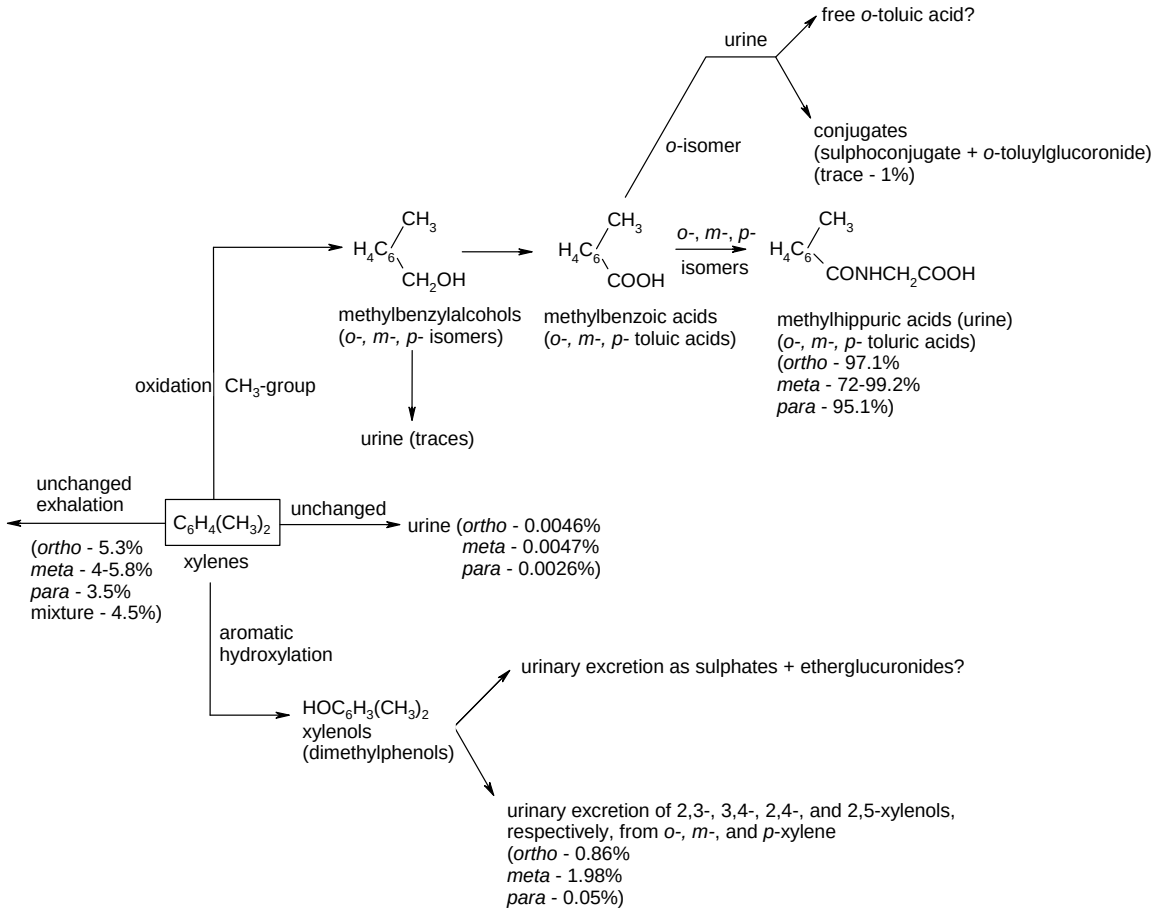
3.4.2.3 Dermal Exposure

No studies were located regarding distribution of xylene in humans following dermal exposure to mixed xylene or individual isomers. Extremely limited information was located regarding distribution in animals following dermal absorption. After topical administration, *m*-xylene was rapidly detectable in plasma, reaching peak concentrations in about 2 hours (Skowronski et al. 1990). About 0.003% of the initial dose was detected per gram of subcutaneous fat 48 hours after exposure.

3.4.3 Metabolism

The biotransformation of xylene in humans proceeds primarily by the oxidation of a side-chain methyl group by microsomal enzymes (mixed function oxidases) in the liver to yield toluic acids (methylbenzoic acids). These toluic acids conjugate with glycine to form toluic acids (methylhippuric acids) that are excreted into the urine (Astrand et al. 1978; Norstrom et al. 1989; Ogata et al. 1970, 1979; Riihimaki et al. 1979a; Sedivec and Flek 1976b; Senczuk and Orlowski 1978). This metabolic pathway accounts for almost all of the absorbed dose of xylene, regardless of the isomer, route of administration, administered dose, or duration of exposure. Minor metabolic pathways that account for <10% of the absorbed dose include the elimination of unchanged compound in the exhaled breath and in the urine, and the urinary elimination of methylbenzyl alcohols, *o*-toluylglucuronides (*o*-toluic acid glucuronide), xylene mercapturic acid (Norstrom et al. 1988), and xylenols (dimethylphenols). Dimethylphenylmercapturic acid (DPMA) was detected in only 9 out of 27 samples of exposed workers and occurred at a ratio of only 0.0003% compared to the main metabolite, methylhippuric acid (Gonzalez-Reche et al. 2003). The metabolism of the various xylene isomers in humans is presented in Figure 3-3.

Figure 3-3. Metabolic Scheme for Xylenes—Humans



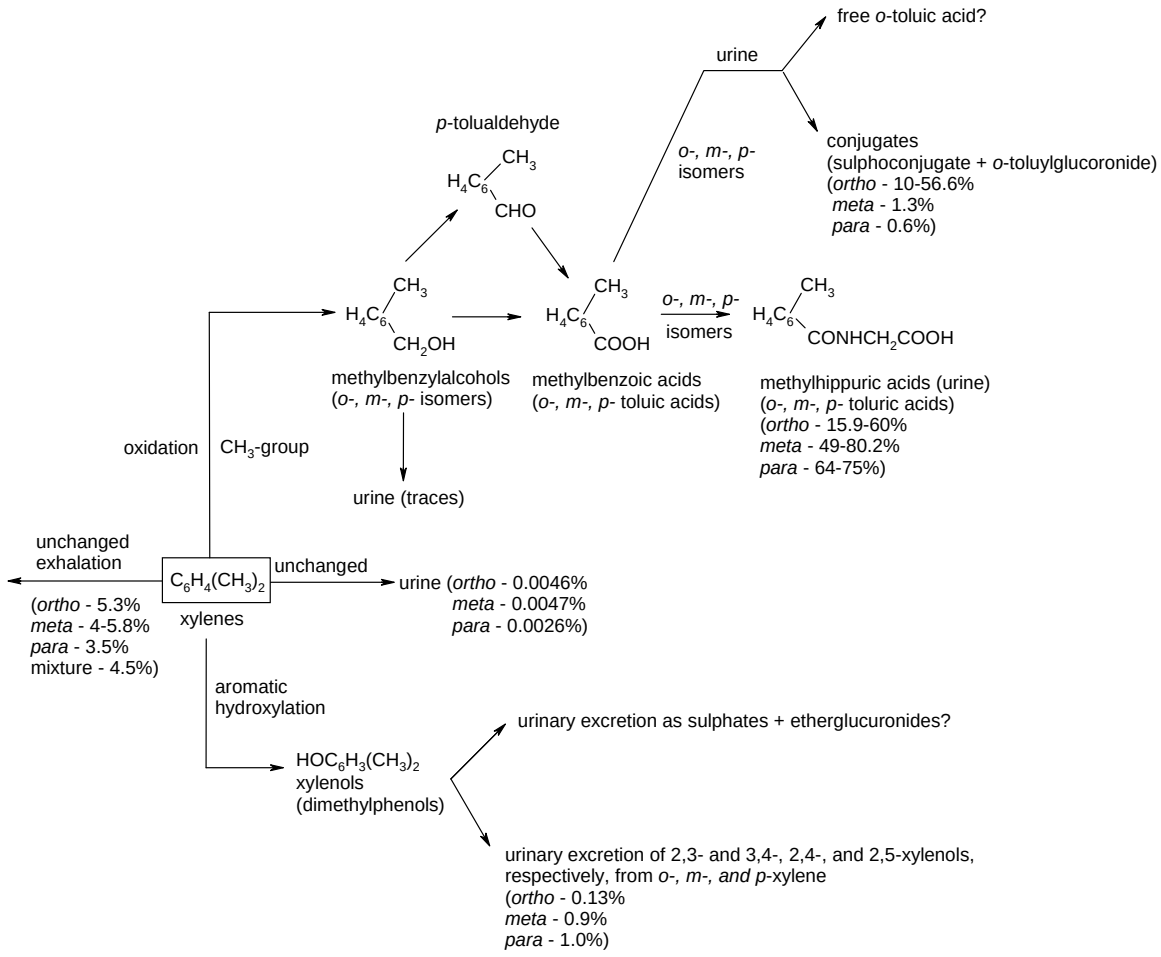
Sources: derived from Astrand et al. 1978; Ogata et al. 1980; Riihimaki et al. 1979a, 1979b; Sedivec and Flek 1976b; Senczuk and Orlowski 1978; Toftgard and Gustafsson 1980

In physiologically based pharmacokinetic (PBPK) modeling of blood concentration data from male subjects exposed by inhalation to 0–44 ppm of deuterium-labeled xylene isomers for 2 hours, the rate of hepatic metabolism was held constant scaled to body weight (Adams et al. 2005). Simulations revealed that apparent systemic clearance of isomers sometimes exceeded that predicted by the hepatic blood flow parameter, suggesting that extrahepatic tissues could play a significant role in xylene metabolism in certain individuals.

The metabolism of xylene in animals is qualitatively similar to that of humans, though quantitative differences do exist (Bakke and Scheline 1970; Bray et al. 1949; Ogata et al. 1979; Sugihara and Ogata 1978; van Doorn et al. 1980). The metabolism of the various isomers in animals is presented in Figure 3-4. The major quantitative difference between animals and humans occurs in the metabolism of the metabolic intermediate methylbenzoic acid (toluic acid). In rats given *m*-, *o*-, or *p*-xylene by intraperitoneal injection, 10–56.6% of the administered dose of *o*-xylene was excreted in the urine as *o*-toluylglucuronide, whereas approximately 1% of the administered doses of *m*- and *p*-xylene was metabolized to the appropriate toluylglucuronide (Ogata et al. 1979; van Doorn et al. 1980). The amounts of *m*-methylhippuric acid and *p*-methylhippuric acid excreted in the urine accounted for 49–63 and 64–75% of the administered dose, respectively (Ogata et al. 1979; Sugihara and Ogata 1978). Similar results were seen in rats administered *m*-xylene by gavage (Turkall et al. 1992). In studies with rabbits, 60% of an administered *o*-xylene dose, 81% of an *m*-xylene dose, and 88% of a *p*-xylene dose were excreted in the urine as methylhippuric acids (Bray et al. 1949). Minor quantities of methylbenzyl alcohols and xylenols have also been detected in the urine of experimental animals administered xylene isomers (Bakke and Scheline 1970; Ogata et al. 1979; Turkall et al. 1992; van Doorn et al. 1980). In a study of metabolites formed by aromatic hydroxylation, rats administered 100 mg/kg doses of xylene isomers eliminated 0.1% of a dose of *o*-xylene in the urine as 3,4-xylenol and 0.03% as 2,3-xylenol, 0.9% of a dose of *m*-xylene as 2,4-xylenol, and 1% of a dose of *p*-xylene as 2,5-xylenol (Bakke and Scheline 1970); A trace of the methylbenzyl alcohol was also detected in the urine of rats given *o*- and *m*-xylene, but not in rats given *p*-xylene. In rats administered *m*-xylene by the dermal route, the major metabolite in the urine over a 24-hour period was identified as methylhippuric acid (82.3%), with xylenol comprising 7.2% and unchanged *m*-xylene comprising 3.8% of the urinary products (Skowronski et al. 1990). In rats given *m*-xylene adsorbed onto sandy soil, the proportion of xylenol present in the urine over the first 12 hours of excretion was significantly increased.

Studies in animals have also shown that the metabolism of xylene may be influenced by prior exposures to xylene (Elovaara et al. 1989). Pretreatment of rats with *m*-xylene increased the percentage of

Figure 3-4. Metabolic Scheme for Xylenes—Animals



Sources: derived from Bakke and Scheline 1970; Bray et al. 1949; Ogata et al. 1980; Sugihara and Ogata 1978; Toftgard and Gustafsson 1980; van Doorn et al. 1980

methylhippuric acid and thioethers in the urine by approximately 10%. A toxic metabolite of xylene in rats and rabbits appears to be methylbenzaldehyde (tolualdehyde) (Carlone and Fouts 1974; Patel et al. 1978; Smith et al. 1982). It is formed by the action of alcohol dehydrogenase on methylbenzyl alcohol in lung and liver tissues (Elovaara et al. 1987); the presence of methylbenzaldehyde has not been confirmed in humans. Lung tissue can be damaged by this intermediate because of its selective inactivation of enzymes involved in microsomal electron transport (mixed function oxidases, cytochrome P-450) (Elovaara et al. 1987).

The differences in xylene metabolism observed between humans and animals may, in part, be explained by differences in the size of the doses given to humans and animals in experimental studies (David et al. 1979; Ogata et al. 1979; van Doorn et al. 1980). The formation of glucuronic acid derivatives may be an emergency mechanism that is activated when the organism can no longer conjugate all acids with glycine (Ogata et al. 1979; Sedivec and Flek 1976b; van Doorn et al. 1980). Humans dosed with 19 mg/kg xylene excreted only methylhippuric acids in the urine, whereas rabbits exposed to 600 mg/kg excreted both methylhippuric acids and derivatives of glucuronic acid (Sedivec and Flek 1976b). The second-phase conjugation of the main oxidized intermediate (methylbenzoic acid with glycine to form methylhippuric acid) may be the rate-limiting step in humans. The maximum rate of glycine mobilization limits the rate of conjugation to a level below 200 µmol/minute (Riihimaki et al. 1979a, 1979b). If this limit is approached, other elimination pathways may be activated, such as conjugation with glucuronic acid or aromatic hydroxylation to form xylenols. The capacity of the first-phase oxidation reaction, encompassing both side-chain and aromatic oxidation, is not known. Aromatic oxidation of xylene could possibly produce toxic intermediates and phenolic end-metabolites (Riihimaki et al. 1979b); however, this is a minor metabolic pathway.

Using a low-temperature (cryogenic), whole-body autoradiography technique for mice exposed by inhalation to radiolabeled *m*-xylene for 10 minutes, Bergman (1983) observed high levels of metabolites (non-volatile radioactivity) in the blood, liver, lung, kidney, and adrenal medulla immediately after exposure. Metabolites were detectable post-exposure in the adrenal medulla for 0.5 hours, in the liver for 2 hours, in the kidney for up to 8 hours, in bile for 2–8 hours, in the intestinal lumen for up to 24 hours, and in the nasal mucosa and bronchi for 2–24 hours. No radioactivity was detectable 48 hours after exposure, indicating that there was no significant binding of xylene metabolites to tissue macromolecules.

3.4.4 Elimination and Excretion

3.4.4.1 Inhalation Exposure

In humans, about 95% of absorbed xylene is biotransformed and excreted as urinary metabolites, almost exclusively as methylhippuric acids; the remaining 5% is eliminated unchanged in the exhaled breath (Astrand et al. 1978; Ogata et al. 1979; Pellizzari et al. 1992; Riihimaki et al. 1979b; Sedivec and Flek 1976b; Senczuk and Orlowski 1978). Less than 0.005% of the absorbed dose of xylene isomers is eliminated unchanged in the urine, and <2% is eliminated as xylenols (Sedivec and Flek 1976b). The excretion of methylhippuric acids is rapid and a significant amount is detected in the urine within 2 hours of exposure. The amount of methylhippuric acid increases with time. Differences in the amount of the metabolites excreted depend on the interpersonal differences in lung ventilation and retention, not on the isomer of xylene (Sedivec and Flek 1976b).

There appear to be at least two distinct phases of elimination, a relatively rapid one (half-life, 1 hour) and a slower one (half-life, 20 hours). These phases of elimination are consistent with the distribution of xylene into three main tissue compartments; the rapid and slower elimination phases correspond to elimination from the muscles and the adipose tissue, respectively, whereas the elimination of xylene from the parenchymal organs is so rapid that the available studies could not monitor it (Ogata et al. 1970; Riihimaki et al. 1979a, 1979b). It is also possible that the renal excretion of the most common xylene metabolite, methylhippuric acid, takes place via the tubular active secretion mechanism of organic acids. Renal excretion is not a rate-limiting step in the elimination of absorbed xylene under normal physiological conditions (Riihimaki et al. 1979b). Physiologically based pharmacokinetic modeling suggests that the urinary excretion of *m*-methylhippuric acid following *m*-xylene exposure of humans is linear at concentrations up to 500 ppm, and that elimination of *m*-methylhippuric acid is slower in individuals with a greater percentage of body fat (e.g., women) (Kaneko et al. 1991a, 1991b). Systemic clearance rates in male subjects following inhalation of ≤44 ppm deuterium-labeled xylene isomers for 2 hours ranged between 116 and 129 L/hour (Adams et al. 2005). The terminal half-life averaged 34 hours for all three isomers among all subjects, but varied widely among individuals from 12 to 108 hours.

Volunteers acutely exposed by inhalation to 100 or 200 ppm *m*-xylene for 7 hours excreted 54 and 61%, respectively, of the administered dose by 18 hours after exposure ended (Ogata et al. 1970). Following intermittent acute exposure of men and women to 23, 69, or 138 ppm *m*-xylene, excretion of *m*-methylhippuric acid peaked 6–8 hours after exposure began. It decreased rapidly, regardless of

exposure level or sex, after exposure had ended. Almost no xylene or *m*-methylhippuric was detected 24 hours later (Senczuk and Orlowski 1978). Following exposure to 200 mg/m^3 *m*-xylene for 2 hours, small differences in elimination were noted between genders (Ernstgard et al. 2003). Women excreted 31% more (AUC) unmetabolized *m*-xylene in exhaled air than men, whereas men excreted 37% more (AUC) *m*-methylhippuric acid in urine than women.

Exercise increased the amount of xylene absorbed and thus increased the amount of *m*-methylhippuric acid and 2,4-xylenol eliminated in the urine of men exposed to *m*-xylene (Riihimaki et al. 1979b). No evidence of saturation of metabolism of xylene was observed at exposures as high as 200 ppm (Riihimaki et al. 1979b). The excretion of *m*-methylhippuric acid appeared to correspond very closely to the estimated xylene uptake and expired xylene represented about 4–5% of the absorbed xylene in all exposure groups (Riihimaki et al. 1979b). Urinary excretion of methylhippuric acid correlated well with exposure (Kawai et al. 1992; Lapare et al. 1993; Skender et al. 1993), and based on a study of workers occupationally exposed to mixed xylenes (geometric mean TWA 14 ppm), Inoue et al. (1993) estimated a slope of 13 mg methylhippuric acid/L/ppm (11.1 mg/g creatinine/ppm) for all three isomers. A sex-related difference in the urinary excretion of methylhippuric acids was not observed (Inoue et al. 1993).

A few studies have evaluated the kinetics of elimination of the metabolites of xylene following inhalation exposure of experimental animals. The primary urinary metabolite is methylhippuric acid (David et al. 1979). A dose-relationship in the urinary elimination of *m*-methylhippuric acid in rats was observed immediately after exposure to 50 or 500 ppm *m*-xylene for 6 hours (Kaneko et al. 2000). Another study found the increase of urinary elimination of methylhippuric acid metabolites to be linear in rats exposed for 5 hours to mixed xylene between 75 and 150 ppm, but non-linear between exposures at 150 and 225 ppm (Tardif et al. 1992). Concomitantly, there were non-linear increases in the amount of xylene excreted unchanged in exhaled air, indicating possible saturation of metabolism at 225 ppm.

Using a low-temperature (cryogenic), whole-body autoradiography technique for mice exposed by inhalation to radiolabeled *m*-xylene for 10 minutes, Bergman (1983) observed elimination of metabolites (non-volatile radioactivity) in bile and feces. Clearance of metabolites was completed in the adrenal medulla by 0.5 hours, in the liver after a little over 2 hours, in the kidney after 8 hours, in bile after 8 hours, in the intestinal lumen after 24 hours, and in the nasal mucosa and bronchi after 24 hours. No radioactivity was detectable 48 hours after exposure.

3.4.4.2 Oral Exposure

Limited information is available on the elimination of the metabolites of xylene following ingestion in humans. In an unspecified number of male volunteers given oral doses of 40 mg/kg/day of *o*-xylene or *m*-xylene, the molar (mol) excretion ratios (total excretion [mol] in urine during appropriate interval/dose administered [mol] x 100[%]) for *o*-methylhippuric acid and *m*-methylhippuric acid were 33.1 and 53.1, respectively (Ogata et al. 1979). More of the *m*-xylene is eliminated as methylhippuric acid than the ortho derivative for *o*-xylene. The molar excretion ratio for *o*-toluic acid glucuronide (*o*-toluyl-glucuronide) was 1.0 in men given *o*-xylene as an oral dose of 40 mg/kg/day. The amounts of *o*-methylhippuric acid (*o*-toluic acid) and of *o*-toluic acid glucuronide excreted in the urine attained maximal levels in 3–6 hours of exposure, while that of *m*-methylhippuric acid attained peak levels in 1–3 hours (Ogata et al. 1979). These results indicate that the major elimination pathway of *o*-xylene is the formation of *o*-methylhippuric acid in humans. The formation of *o*-toluic acid glucuronide is a minor pathway for the elimination of *o*-toluic acid, but would be available in the event of saturation of the major pathway.

Excretion of radioactivity by rats following an oral dose of *m*-xylene showed most excretion occurred in the urine during the first 12 hours after dosing (50–59% of the dose), with a total of 96.2% eliminated in urine in males and 73.7% in urine in females over 48 hours (Turkall et al. 1992). Approximately 8 and 22% of the total dose was excreted in exhaled air by males and females, respectively, during the first 12 hours. *m*-Methylhippuric acid comprised 67–75% of the urinary radioactivity, with xylenol comprising 2–18%, and unchanged xylene comprising approximately 1%. When *m*-xylene adsorbed onto sandy soil matrix was administered, the excretion of radioactivity in urine and exhaled air in female rats was significantly decreased compared to *m*-xylene during the first 12 hours (Turkall et al. 1992). In males given xylene in a sandy or clay soil matrix, the excretion of radioactivity was increased in exhaled air during the first 12 hours. In a study of aromatic hydroxylation products of xylene metabolism, rats administered 100 mg/kg doses of xylene isomers eliminated 1% or less of the administered dose as xylenols in the urine, along with trace amounts of methylbenzyl alcohols for *o*- and *m*-xylene only (Bakke and Scheline 1970).

3.4.4.3 Dermal Exposure

The elimination of *m*-xylene has been studied in human subjects exposed dermally either to liquid or to vapor. The elimination of liquid *m*-xylene absorbed dermally in humans following a 15-minute exposure was through the exhaled breath and urine (Engstrom et al. 1977; Riihimaki and Pfaffli 1978). Elimination

in the exhaled breath followed a two-phase elimination curve with a rapid half-life of 1 hour and a longer half-life of 10 hours. In human subjects who immersed both hands in *m*-xylene for 15 minutes, the rate of excretion of *m*-methylhippuric acid was approximately 50 µmol/hour at 2 hours and 2 µmol/hour 3 hours later (Riihimaki 1979b). In male subjects whose hand and forearm were exposed dermally to 6,770 ppm *m*-xylene vapor for up to 3 hours, elimination of xylene in the exhaled breath was detected within 10 minutes and continued up to several hours after the exposure ended (Kezic et al. 2004).

In rats receiving a dermal application of 15 mg of radiolabeled *m*-xylene per cm^2, elimination over 48 hours was primarily in unmetabolized expired air (61.9% of the initial dose), with 42.7% excreted as metabolites in the urine and 0.1% in feces (Skowronski et al. 1990). The majority of the excretion in expired air occurred within the first 12 hours, with excretion in the urine occurring primarily during the first 24 hours; fecal excretion was detected the second day. If *m*-xylene was applied to the skin in the form of a sandy soil matrix, the excretion was similar to that seen with *m*-xylene alone, but if the *m*-xylene was applied adsorbed onto clay soil matrix, approximately equal amounts were excreted in exhaled air and in the urine (46 and 53%, respectively).

3.4.4.4 Other Routes of Exposure

Limited information was available on the elimination of xylene metabolites in rats following intraperitoneal injection (Ogata et al. 1979; Sugihara and Ogata 1978; van Doorn et al. 1980). The urinary metabolites of xylene are similar regardless of route of exposure; however, the amounts of the various metabolites differ. The elimination profile of xylene isomers is related more to absorption than it is with dose or duration of exposure. In rats, 49–62.6% of various doses of *m*-xylene or 64–75% of various doses of *p*-xylene were excreted in the urine as *m*-methylhippuric acid or *p*-methylhippuric acid, respectively (Sugihara and Ogata 1978). Urinary excretion of *o*-toluic acid glucuronide and *o*-methylhippuric acid accounted for 57 and 16%, respectively, of single intraperitoneal dose of 1,240 mg *o*-xylene/kg give to rats (Ogata et al. 1979). The amount of *o*-toluic acid glucuronide and *o*-methylhippuric acid excreted reached a maximum 8–24 hours after dosing. Mercapturic acid derivatives were present in the urine of rats following an intraperitoneal dose of *m*-, *o*-, or *p*-xylene (Tanaka et al. 1990; van Doorn et al. 1980). The percentages ranged from 0.6% (*p*-xylene) to 10–29% (*o*-xylene).

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

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

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

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

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

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

PBPK models improve the pharmacokinetic extrapolations used in risk assessments that identify the maximal (i.e., the safe) levels for human exposure to chemical substances (Andersen and Krishnan 1994). PBPK models provide a scientifically sound means to predict the target tissue dose of chemicals in humans who are exposed to environmental levels (for example, levels that might occur at hazardous waste sites) based on the results of studies where doses were higher or were administered in different species. Figure 3-5 shows a conceptualized representation of a PBPK model.

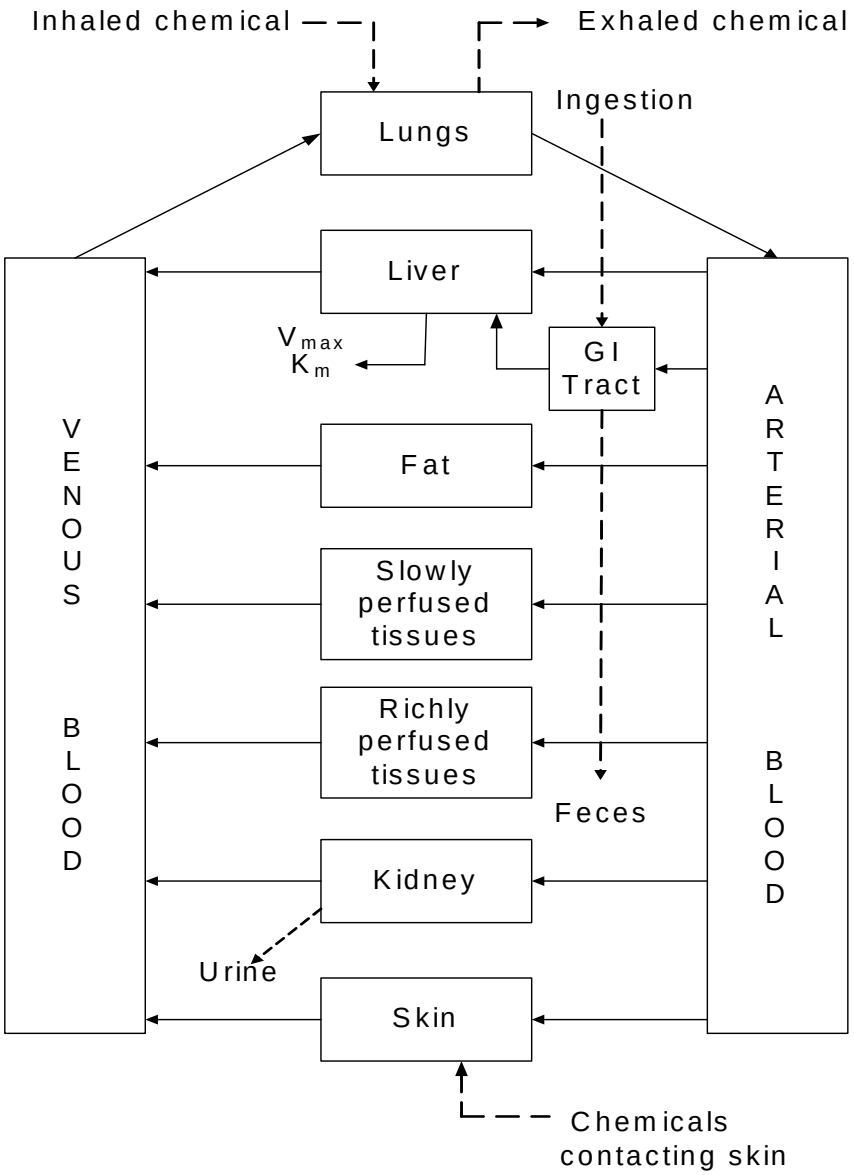
If PBPK models for xylene exist, the overall results and individual models are discussed in this section in terms of their use in risk assessment, tissue dosimetry, and dose, route, and species extrapolations.

Kaneko et al. (1991a) Inhalation Exposure Model

Kaneko et al. (1991a) developed a PBPK model for inhalation of *m*-xylene by rats and humans, based on the trichloroethylene model of Endoh et al. (1989). The model simulates concentrations of *m*-xylene in seven modeled tissue compartments and the amount of metabolites formed (Figure 3-6 and Table 3-7).

Description of the Model. The model consists of compartments for lung (lung arterial blood and one-third of the tidal volume), liver, fat (adipose tissues and yellow bone marrow), slowly-perfused tissues (red bone marrow), richly-perfused tissues (brain, heart, kidneys and glandular tissues), muscle (muscle and skin), and gastrointestinal tract (portal system excluding the liver), plus an arteriovenous shunt. Model parameters included tissue volumes and blood flows, calculated for each compartment based on values by Davis and Mapleson (1981), and several values that were determined experimentally for adult male Wistar rats: partition coefficients, metabolic constants (V_{max} and K_m), and the rate constant for urinary excretion of *m*-methyl hippuric acid. The concentration flowing out of a given compartment (in venous blood) was set as equilibrated with the concentration in that compartment. All metabolism of *m*-xylene was assumed to occur in the liver and was expressed as a Michaelis-Menten process. The metabolism of *m*-xylene considered only the main pathway, (i.e., the conversion to *m*-toluic acid, which is conjugated to glycine and excreted in the urine as *m*-methylhippuric acid); the pathway from *m*-xylene to coefficients for rat blood, lung, brain, heart, kidney, testis, intestine, spleen, liver, fat, and muscle were determined by vial equilibration, and tissue/blood values were calculated by dividing the tissue/air

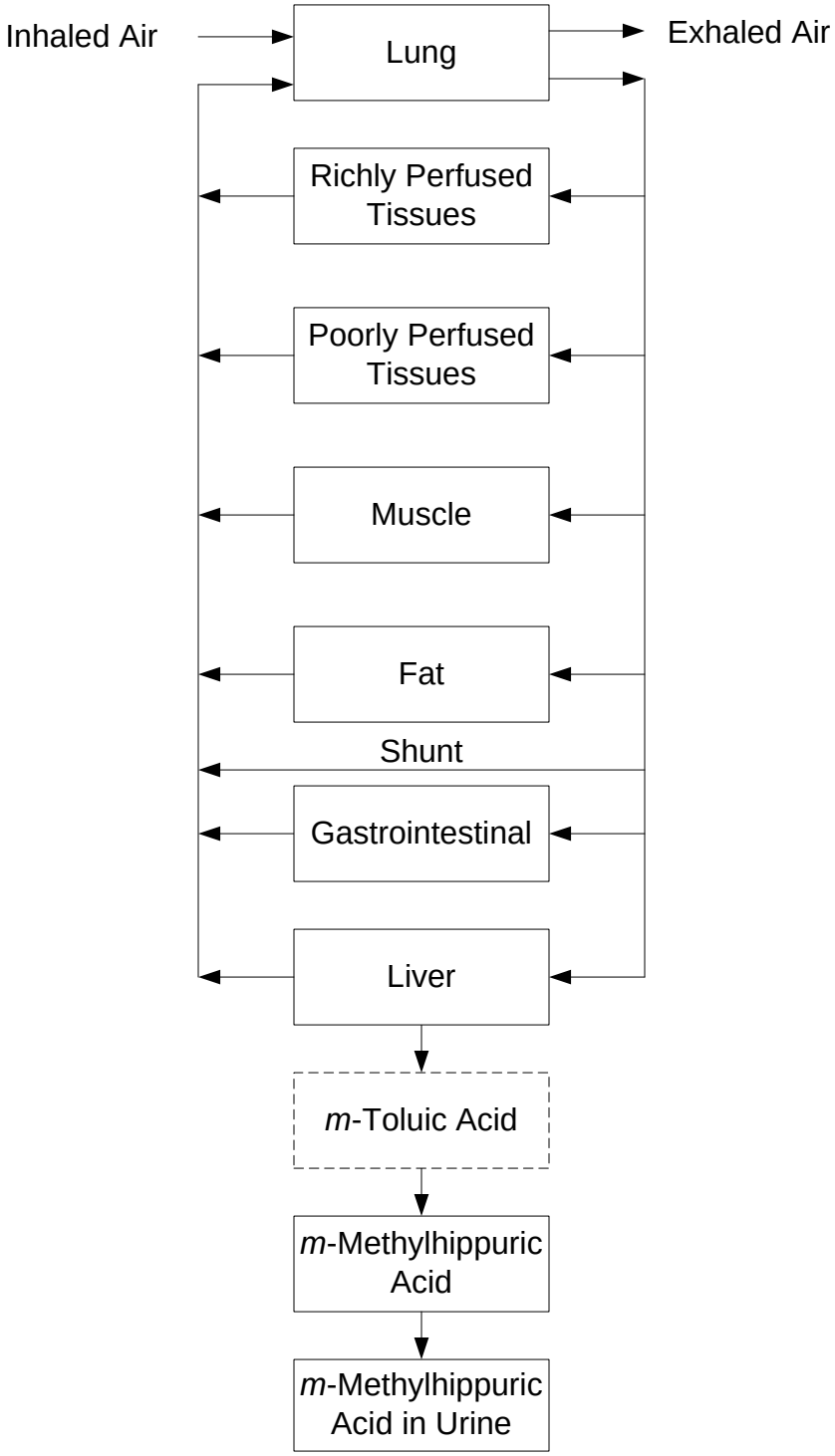
Figure 3-5. Conceptual Representation of a Physiologically Based Pharmacokinetic (PBPK) Model for a Hypothetical Chemical Substance



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

Source: adapted from Krishnan and Andersen 1994

Figure 3-6. PBPK Model for Inhaled *m*-Xylene



Source: adapted from Kaneko et al. (1991a)

Table 3-7. Human Parameters Used in the PBPK Inhalation Model for *m*-Xylene

Tissue volume (percent body weight)		
Lung	calculated[a]	
Richly perfused	3	
Slowly perfused	8.5	
Muscle	41.5	
Fat	21.1	
Gastrointestinal tract	1.9	
Liver	2.3	
Cardiac output (L/minute)	$0.296\ (bw)^{0.7}$	
Blood flow (percent cardiac output)		
Lung	100	
Richly perfused	37.9	
Slowly perfused	6.3	
Muscle	11.4	
Fat	5.3	
Gastrointestinal tract	17.1	
Liver	6.9	
Shunt	15.1	
Air flow		
Alveolar ventilation (L/hour)[b]	$0.296\ (bw)^{0.7}$	
Partition coefficients		
Lung:blood	4.09	
Richly perfused:blood	4.42	
Slowly perfused:blood	2.01	
Muscle:blood	3.01	
Fat:blood	77.8	
Gastrointestinal tract:blood	4.67	
Liver:blood	3.02	
Blood:air	26.4	
Metabolic constants		
V_{max}[c] (mmol/minute)	$V_{max1} = 1.394 \times 10^{-3}(bw)^{0.7}$	$V_{max2} = 1.115 \times 10^{-2}(bw)^{0.7}$
K_m[d] (mmol/L)	$K_{m1} = 0.033$	$K_{m2} = 0.330$
K_{ex}[d] (per minute)	0.012	

Source: Adapted from Kaneko et al. (1991a)

[a]Lung volume = functional residual capacity + one third tidal volume + (volume arterial blood x blood:air partition coefficient + volume of lung tissue x lung:air partition coefficient.
[b]Set equal to cardiac output.
[c]Rat V_{max} and K_m were determined experimentally for two different exposure concentrations. Rat V_{max1} (0.6×10^{-3} mmol/minute) and V_{max2} (4.8×10^{-3} mmol/minute) values were corrected for body surface area $(body\ weight)^{0.7}$.
[d]Set equal to experimentally determined rat value.

bw = body weight

coefficient by the blood/air coefficient. The richly perfused coefficient was set equal to that of the liver. Human coefficients were calculated as the rat tissue/air partition coefficients divided by the human blood/air coefficient reported by Sato and Nakajima (1979). Metabolic rate constants were experimentally determined for high and low concentrations. For the human model, cardiac output and metabolic rate constants were estimated from rat values using a (body weight)$^{0.70}$ scalar, but the rat K_m was used directly. Simulations were performed for a 70-kg man inhaling 100 ppm *m*-xylene for 6 hours as reported by Riihimaki et al. (1979b). The blood concentration was set equivalent to the concentration flowing out of the richly-perfused compartment.

Validation of the Model. The model parameters were optimized against the human data of Riihimaki et al. (1979b). Comparisons of model simulations against data from studies other than those used in model development were not presented.

Additional Predictions. Kaneko et al. (1991b) used the model to evaluate the relationship of external and internal doses of *m*-xylene and determine the effects of body weight, body fat content, sex, and physical activity on blood concentrations and urinary excretion of metabolite. Several different exposure scenarios were simulated: continuous exposure of a 70-kg male to 50 ppm for 8 hours, intermittent exposure of a 70-kg male to 100 ppm for 1 hour 4 times at 1-hour intervals, continuous exposure of a 70-kg male for 8 hours to various concentrations between 0 and 4,000 ppm. In the simulation, larger bodies absorbed more *m*-xylene, and excreted more metabolite, but there was no significant change in blood concentration. Increased physical activity increased both the blood concentration of *m*-xylene and the rate of urinary excretion of metabolite. The concentration of *m*-xylene in the blood during exposure was lower in women than in men, but was higher in women 10 hours after exposure. The rate of urinary excretion of metabolite was lower in women both during and after exposure.

Adams et al. (2005) Inhalation Exposure Model

Adams et al. (2005) developed a PBPK model for inhalation of deuterium-labeled xylene isomers in humans, using biomonitoring data (isomer levels in blood and exhaled air) to quantify the kinetics of each isomer in adult Caucasian men. The model simulates concentrations in six modeled tissue compartments and assumes systematic clearance of xylenes by metabolism primarily in the liver, but also in the lung.

Description of the Model. The model consists of compartments for lung blood, adipose tissue, slowly perfused tissue, rapidly perfused tissue, and liver. The model used subject-specific values for

body weight, adipose tissue fraction, and exposure concentration. Cardiac output and all blood flows were scaled to (body weight)$^{0.74}$. Published values were used for fractional tissue compartment volumes, fractional blood flows, tissue/blood partition coefficients, and hepatic metabolic constants. Alveolar ventilation, blood flows, and blood/air partition coefficients were assigned as Bayesian parameters to be optimized during model fitting. As xylene was assumed to diffuse rapidly into tissues, the transfer of xylene from arterial blood to tissues was assumed to be flow-limited. Changes in the amount of xylenes in nonmetabolizing tissues, in the metabolizing hepatic compartment, and the mass balance of xylene in the lung (including a metabolic component varying from 0 to 25% of hepatic metabolism) were defined by a series of simultaneous differential equations. Extrahepatic metabolism was indicated when the apparent systematic clearance of xylenes exceeded hepatic blood flow.

Validation of the Model. The model parameters were optimized against human data collected by the investigators (Adams et al. 2005). Twenty-seven male Caucasian volunteers were exposed at rest to atmospheres containing 0–40 ppm each of $^{2}H_{10}$-*meta*-, $^{2}H_{10}$-*ortho*-, and $^{2}H_{10}$-*para*-xylene for 2 hours through a gated mouthpiece. Eight subjects experienced replicate exposures (at a minimal 2-week interval), bringing the total number of exposures to 37. Inhaled and exhaled breath concentrations of deuterium-labeled isomers were measured continuously during exposure. Doses were estimated for all subjects as the product of exposure concentration, exposure duration, and model-fitted subject-specific alveolar ventilation rates. Blood concentrations of isomers were measured in 1 venous blood sample collected before exposure and in 15–20 samples collected at the end of exposure for 4 days; collection frequency was every 15 minutes for the first several hours to every 12 hours after day 1. Model-fitted values for systemic clearance, terminal half-lives, and blood:air partition coefficients were similar to values reported by other investigators (Pierce et al. 1996; Sato 1988; Wallen et al. 1985). Subjects exposed during multiple sessions showed significant intra-individual variability in respiratory and blood physiology parameters as well as metabolism rates. The study authors suggested that differences in diet, alcohol consumption, and stress could affect variation in these parameters.

Thrall and Woodstock (2003) Dermal Exposure Model

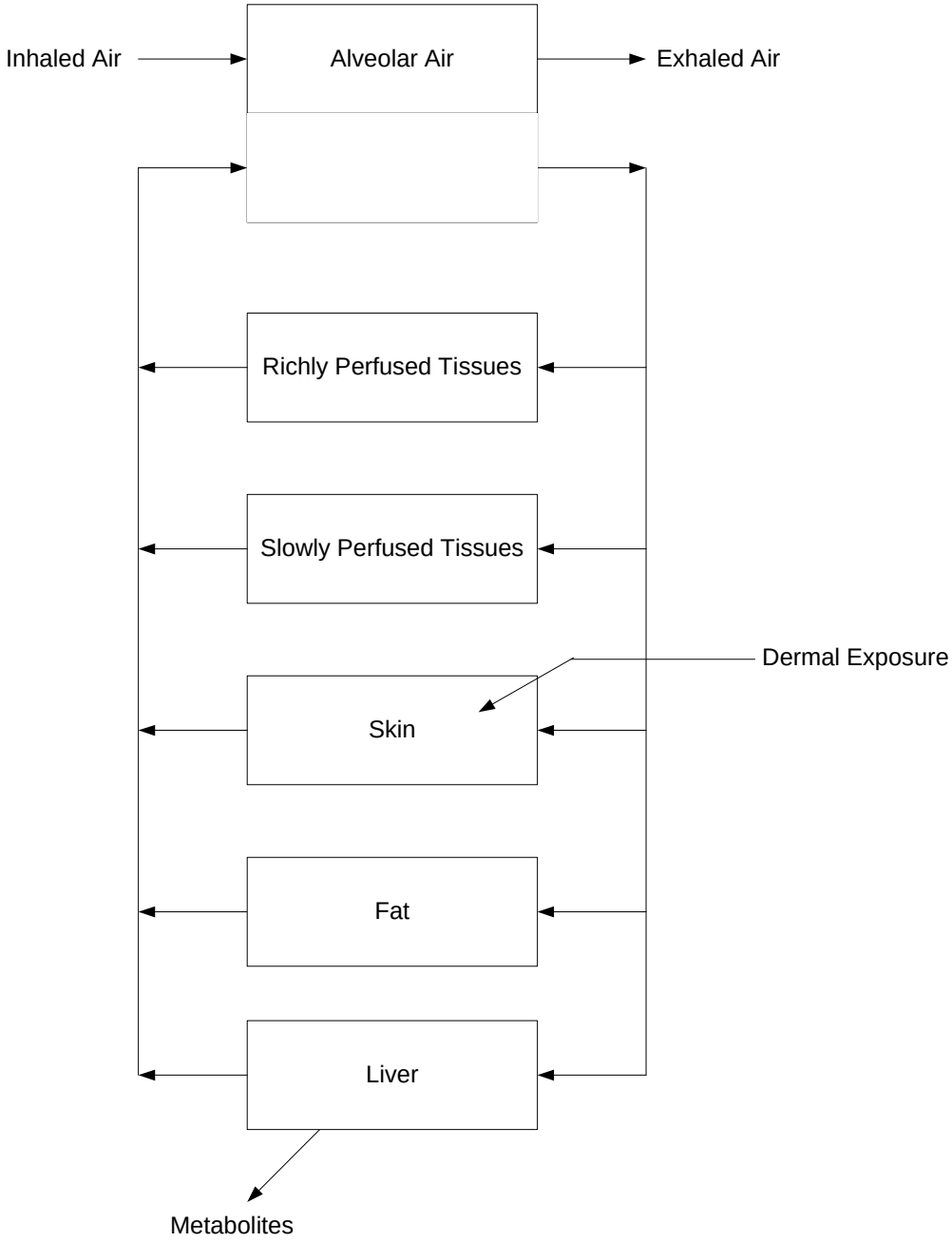
Thrall and Woodstock (2003) adapted a PBPK model for binary inhalation exposures to toluene and *m*-xylene by Tardif et al. (1993a) to construct a PBPK model for dermal exposures to *o*-xylene in rats and humans (Table 3-8 and Figure 3-7).

Table 3-8. Rat and Human Parameters Used in the PBPK Dermal Model for *o*-Xylene

	Rat	Human
Body weight (kg)	0.2–0.25	83.9, 48.5, 80.7
Tissue volume (percent body weight)		
Liver	4	4
Fat	8	30.7, 22.3, 26.4
Richly perfused	5	5
Slowly perfused	64	Variable
Total skin	10	10
Cardiac output (L/hour)	5.4	348
Blood flow (percent cardiac output)		
Liver	25	25
Fat	5	6
Richly perfused	51	49
Slowly perfused	15	15
Total skin	5	5
Air flow		
Alveolar ventilation (L/hour)	5.4	348
Partition coefficients		
Saline:air	2.6	2.6
Blood:air	37.8	34.0
Liver:blood	3.4	3.8
Fat:blood	55.7	61.9
Muscle:blood	2.0	2.2
Skin:air	65.4	65.4
Metabolic constants		
V_{max} (mg/kg/hour)	8.4	8.4
K_m (mg/L)	0.2	0.2

Source: Adapted from Thrall and Woodstock (2003)

Figure 3-7. PBPK Model to Describe Dermal Absorption of *o*-Xylene



Source: adapted from Thrall and Woodstock (2003)

Description of the Model. The model consists of compartments for lung, blood, richly perfused tissues, slowly perfused tissues, skin, fat, and liver. The skin compartment represents exposed skin, whereas nonexposed skin is incorporated into the slowly perfused compartment. Total skin volume (10% of body weight) was assumed to receive 5% of the total cardiac output. The volume and blood flow for exposed skin were calculated according to the methods of Jepson and McDougal (1997). Model parameters were as presented in Table 3-8. Metabolism was assumed to occur in the liver. Dermal exposure was simulated by relating the rate of change in the concentration of xylene in the skin compartment to the rate of penetration through the skin (the flux) and the rate of delivery because of blood flow and the arterial concentration (the perfusion) (Thrall et al. 2000). The model also considered the amount of xylene lost through evaporation from nonoccluded skin. The kinetics of dermal absorption was determined empirically in experiments in rats and humans based on the appearance of xylene in exhaled air. A total of 400 μg of *o*-xylene in aqueous solution (0.02%) was applied to the skin of rats under an occlusive covering for 4 hours. Human subjects submerged their lower legs into a hydrotherapy tub containing 500 μg/L *o*-xylene (a concentration 400 times lower than for rats) for 10–30 minutes and were monitored unexposed for an additional 30 minutes. An algorithm was used to vary the skin permeability coefficient to achieve an optimal fit to the time course data for each subject and then averaged. The calculated skin permeability rate constants of 0.058±0.009 cm/hour for rat and 0.005±0.001 cm/hour for human adequately described the respective individual dermal exposure data sets. Both species rapidly absorbed aqueous xylene through the skin as shown by the chemical's appearance in exhaled air within a few seconds of exposure.

Validation of the Model. The dermal PBPK model was used to simulate the exhaled breath data from experiments in humans dermally exposed for 3 minutes to undiluted *m*-xylene (Kezic et al. 2001). The calculated skin permeability coefficient was identical to the average value of 0.005 cm/hour derived by Thrall and Woodstock (2003). Simulation of blood concentration data from F344 rats dermally exposed to saturated *m*-xylene for 24 hours (Morgan et al. 1991) resulted in a calculated skin permeability coefficient of 0.0727 cm/hour, which was in the range of values determined by Thrall and Woodstock (2003).

Fisher et al. (1997) Lactational Model for Inhalation Exposure

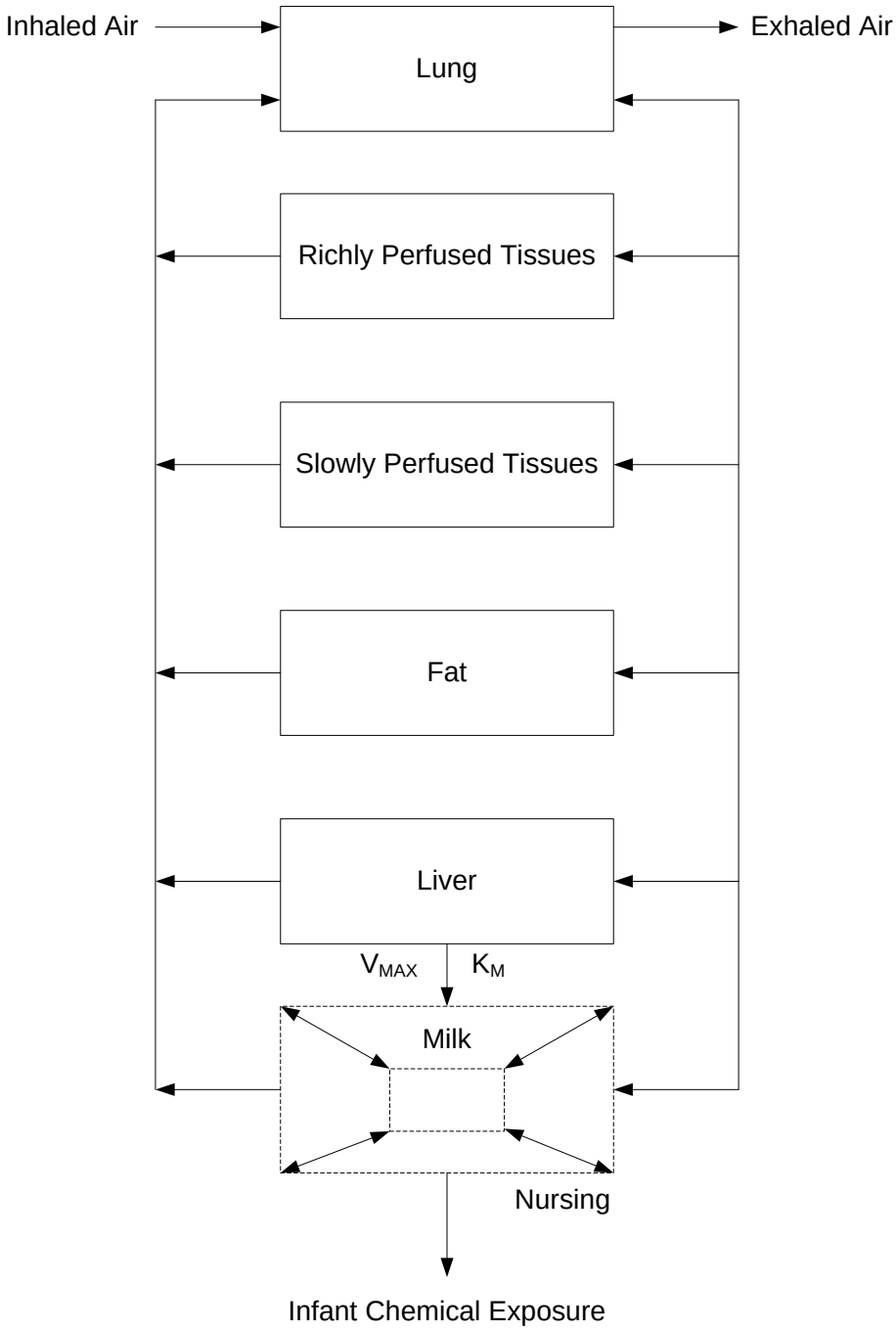
Fisher et al. (1997) adapted the PBPK model of Ramsey and Andersen (1984) to create a lactational model for maternal occupational exposure to volatile organic solvents, including *o*-, *m*-, and *p*-xylenes (Table 3-9 and Figure 3-8).

Table 3-9. Human Parameters Used in the PBPK Lactational Model for *o*-, *m*-, *p*-Xylene

	Human
Body weight (kg)	60
Tissue volume (percent body weight)	
Liver	1.5
Fat	25
Richly perfused	10
Slowly perfused	54
Milk (mL)	10–125
Cardiac output (L/hour)	15 x body weight$^{0.74}$
Blood flow (percent cardiac output)	
Liver	29
Fat	10
Richly perfused	35
Slowly perfused	19
Milk	7
Air flow	
Alveolar ventilation (L/hour)	24 x body weight$^{0.74}$
Milk compartment	
Rate of ingestion of breast milk by nursing infant (per hour)	20
Rate of breast milk production at 1.5–3.0 months lactation (L/hour)	0.06
Partition coefficients	
Liver:blood	2.14
Fat:blood	41.28
Milk:blood	2.98
Richly perfused:blood	2.14
Slowly perfused:blood	0.97
Metabolic constants	
V_{max}[a]	Not reported
K_m[a]	Not reported

Source: Adapted from Fisher et al. (1997)

Figure 3-8. PBPK Model for Lactational Exposure to Volatile Organic Compounds (Including *o*-, *m*-, *p*-Xylenes) Following Maternal Occupational Inhalation Exposure



Source: adapted from Fisher et al. (1997)

Description of the Model. In addition to the standard compartments for lung, richly- and slowly-perfused tissues, fat, and lung (the site of metabolism), the model contained a milk compartment. Milk production was described as a zero-order process at 0.06 L/hour. Milk letdown from nursing (infant nursing rate) was described as a first order process (20/hour). The milk volume in mammary tissue was 0.125 L before nursing and 0.010 L at the end of nursing. The equation describing the rate of change in the amount of chemical ingested by the nursing infant included terms for the chemical concentration in milk, the amount of milk in the mammary tissue lumen, the infant nursing rate, and a switch to turn on or off nursing over 24 hours. Integration of this equation provided the cumulative amount of chemical ingested by a nursing infant in 24 hours. Partition coefficients for blood:air and milk:air were experimentally derived to calculate the milk:blood partition coefficient (Table 3-9). The human tissue:blood coefficient for xylenes was calculated by dividing the rat tissue:air coefficients of Gargas et al. (1989) by the human blood:air coefficient. The published paper did not report the constants (V_{max} and K_m) used to describe the rate of metabolism of xylenes in humans, so it appears that the authors may have assumed zero metabolism for xylenes.

The lactational model was used to simulate lactational transfer of xylenes following maternal exposure for 8 hours at the threshold limit value (TLV) of 100 ppm. The model predicted that approximately 1–2% of inhaled xylenes would be transferred to milk and that the daily intake for a 10 kg infant would be 6.59 mg.

Interaction Models

In addition to those described above, a number of PBPK models have been developed for exposures to xylene in the presence of other chemicals. Tardif et al. (1993a, 1995) first developed a PBPK model for inhalation exposures to binary mixtures of *m*-xylene and toluene in the rat and later adapted the model for humans. The models based on parameters for *m*-xylene were found to adequately simulate the kinetics of mixed xylenes as well. A later model was developed for inhalation of a ternary mixture of alkyl benzenes (*m*-xylene, toluene, and ethylbenzene) in rats and humans (Tardif et al. 1997). Haddad et al. (2000) validated a PBPK model for inhalation of mixtures of benzene, toluene, ethyl benzene, and *m*-xylene (BTEX), with the addition of a model for dichloromethane. Kaneko et al. (1995) modeled the effect of phenobarbital pretreatment with exposure to *m*-xylene orally or intraperitoneally at the same doses or by inhalation. The interactions between xylene and the other chemicals are presented in Section 3.9.

MacDonald et al. (2002) used PBPK modeling to estimate central nervous system effects following exposure to a time-weighted-average 100 ppm of *m*-xylene for 8 hours. The model incorporated the inhalation PBPK model for *m*-xylene by Kaneko et al. (1991a) and a model for ingested ethanol by Derr (1993). Other components included a competitive enxyme (CYP2E1) inhibition model for the two chemicals, as well as a population PBPK simulation model derived from statistical distributions of the PBPK model parameters with Monte Carlo sampling. Finally, since central nervous system effects were assumed to be dependent on blood levels of *m*-xylene, a pharmacodynamic (PD) threshold model was included for peak venous concentrations of *m*-xylene. The deterministic PBPK model for *m*-xylene was validated against available human data (Loizou et al. 1999; Riihimaki et al. 1982b). The population PBPK-PD model for *m*-xylene was run to establish the population distribution for peak venous blood *m*-xylene concentrations ($C_{V,max}$) following exposure at the U.K. occupational exposure standard of 100 ppm over 8 hours and for a 4-hour exposure at 200 ppm that elicited central nervous system disturbances in healthy volunteers (Savolainen et al. 1985a). The corresponding 80th percentile on the $C_{V,max}$ cumulative frequency distribution, 20.35 μmol/L, was selected as the pharmacokinetic/ pharmcodynamic threshold for central nervous system depression elicited by exposure to *m*-xylene. Simulations were also reported for blood levels of ethanol alone, combined *m*-xylene and ethanol, and population PBPK-PD modeling for the combined exposure scenario. In the model simulations, increasing exposure to ethanol increased the probability of central nervous system effects from exposure to *m*-xylene.

PBPK modeling has been employed to assess possible toxicological interactions between chemicals, such as inhalation exposure to *m*-xylene and methylchloroform (Tardif and Charest-Tardif 1999).

Haddad et al. (2001) developed a PBPK modeling-based risk assessment approach for mixtures of volatile chemicals that would incorporate pharmacokinetic interactions, such as changes in hepatic metabolism, among components of the mixture.

Model Applications

In deriving a chronic-duration inhalation reference concentration (RfC) for xylenes, the EPA (2003) examined the application of the inhalation PBPK models for *m*-xylene developed by Tardif and colleagues (Haddad et al. 1999; Tardif et al. 1991, 1992, 1993a, 1993b, 1995) to extrapolate rat exposure levels from the intermediate-duration inhalation study by Korsak et al. (1994) to human equivalent exposure levels. Using a time-weighted-average steady-state concentration of *m*-xylene in blood as the

internal dose surrogate, it was found that model-predicted human equivalent concentrations (46.5 mg/m^3) were similar, although not identical, to human equivalent concentrations (39 mg/m^3) calculated from rat experimental exposure levels using the 1994 EPA default cross-species dosimetric equations (EPA 2003). EPA (2003) noted that, as neurobehavioral effects were the targets in the study by Korsak et al. (1994), the brain concentration of xylene would have been a more optimal dose surrogate, but the model did not include such a compartment.

Pelekis et al. (2001) used the PBPK models of Tardif et al. (1993a, 1995) to examine possible differences between children and adults with respect to estimated blood and tissue doses following inhalation exposure to xylene. The model simulations incorporated published physiological parameters for a 10-kg 1-year-old child and high and low values for adults. The simulations revealed that the parameters having a significant effect on within-species variability of tissue and blood doses following inhalation exposures were the body weight, ventilation rate, fraction of cardiac output flowing to the liver, blood:air partition coefficient, and hepatic extraction ratio.

Jang and Droz (1997) used PBPK modeling to evaluate possible ethnic differences in metabolism in individuals exposed by inhalation to 100 ppm *m*-xylene for 6 hours (Jang et al. 1997). No significant differences were noted between six Caucasians and six Asians in metabolic clearance of methylhippuric acid.

3.5 MECHANISMS OF ACTION

3.5.1 Pharmacokinetic Mechanisms

Absorption. The isomers of xylene are expected to have similar absorption behavior because of their relatively small molecular size and similar chemical properties. Penetration of xylenes is rapid by all routes of exposure because of the lipophilic nature of the chemicals, as indicated by the log K_{ow} values (see Table 4-2). The similar blood-air partition coefficients for the xylene isomers also reflect the expected similarities in absorption from inhaled air through the lung (Table 3-6). PBPK modeling of human data revealed little inter-subject variability with respect to blood:air partition coefficients (Adams et al. 2005).

Distribution. Distribution of xylenes is rapid throughout the body although their water solubility is low (Table 4-2). Xylenes, because of their lipophilic properties, are readily distributed to fat and tissues rich in cell membranes, such as the brain. The lipophilic nature of xylenes is generally responsible for

their penetration across the placenta to fetal tissues and for distribution into milk (Fisher et al. 1997). Distribution of xylenes is a simple diffusion process. As shown by PBPK modeling, the factors affecting tissue and blood distributions of inhaled xylenes include the body weight (since several physiological parameters are scaled to body weight), ventilation rate, fraction of cardiac output flowing to the liver, blood:air partition coefficient, and hepatic extraction ratio (Pelekis et al. 2001). In addition, hepatic blood flow can be significantly affected by changes in posture, exercise, and thermal stress (Adams et al. 2005). All of these factors may contribute to intra- and inter-individual variations in tissue distributions of xylenes (Adams et al. 2005; Pelekis et al. 2001).

In developing a tissue composition-based algorithm for predicting tissue:air partition coefficients for volatile organic compounds, including isomers of xylene, Poulin and Krishnan (1996a, 1996b) uncovered an anomaly regarding blood compared to other tissues. The algorithm was based on tissue composition with respect to water and total lipid content by weight, and phospholipids and neutral lipids as fractions of total lipids. For liver, muscle, and adipose tissue, the algorithm, which used the solubilities of chemicals in water, neutral lipid, and phospholipid, predicted tissue:air partition coefficients that were similar to values in the published literature (ratios of predicted vs experimental between 0.93 and 1.57 for xylene isomers on average). For blood, however, the algorithm seriously underpredicted the partition coefficients of hydrophobic organic compounds, for xylenes by a factor of 5, indicating that solubility characteristics alone could not account for partitioning. Poulin and Krishnan (1996b) suggested that retention of hydrophobic organic compounds in blood was partly the result of reversible binding to hydrophobic holes in hemoglobin molecules. Association constants (K_a) of 1,512, 1,853, and 1,609 per mole were calculated for reversible binding to hemoglobin by *o*-, *m*-, and *p*-xylene, respectively (Poulin and Krishnan 1996b).

In PBPK modeling of data from humans exposed by inhalation, fitted values for partitioning between blood and slowly perfused tissues exhibited significant inter-individual variability, ranging from 0.68 to 13.78 for *m*-xylene (mean 3.05±3.77), 0.65–17.44 for *o*-xylene (mean 2.88±3.37), and 0.55–12.27 for *p*-xylene (2.25±2.58) (Adams et al. 2005). The basis for this variability is not understood.

Metabolism. Phase I metabolism of xylenes is primarily by the oxidation of side-chain methyl groups by mixed function oxidases in the liver, forming methylbenzoic (toluic) acids. Phase II metabolism is primarily through conjugation with glycine, which is rate limiting in humans (Riihimaki et al. 1979a, 1979b). In the absence of glycine, minor pathways are employed, such as conjugation with glucuronic acid or aromatic hydroxylation to form xylenols. In humans, hepatic microsomal CYP2E1 is the primary

enzyme involved with the metabolism of xylenes to methylbenzylalcohols, the dominant pathway leading to the formation of methylhippuric acid isomers (Tassaneeyakul et al. 1996); human CYP1A2 causes the ring hydroxylation of *m*-xylene to form 2,4-dimethylphenol.

Inter-individual variations in rates of metabolism of xylenes may be affected by lifestyle differences. Treatment with ethanol prior to xylene exposure increases rates of xylene metabolism by the liver, presumably by inducing hepatic microsomal enzymes (Tardif et al. 1994; Wisniewska-Knypl et al. 1989).

Excretion. The lipophilic properties of unmetabolized xylene are responsible for the diffusion out of the lungs into exhaled breath. The water solubility of conjugated metabolites of xylene is responsible for their transported out of the body in urine. As estimated by PBPK modeling of human data, systemic clearance of *m*-xylene (fitted average of 131 L/hour) proceeds more rapidly than for *o*-xylene (116 L/hour) or *p*-xylene (119 L/hour) (Adams et al. 2005).

3.5.2 Mechanisms of Toxicity

The lipophilic effects of xylenes, which dissolve lipid membranes, is responsible for the irritant effects on eyes, mucous membranes and skin (Riihimaki 1979b). In addition, the lipophilicity of xylenes is responsible for their narcotic and anaesthetic properties, which are similar for the three isomers (Fang et al. 1996). The *para* isomer differs in also eliciting excitation reactions in rats (Fang et al. 1996). The mechanism of anesthetics in general is not well understood, but probably relates to intercalation of the chemical into neuronal cell membranes, changing membrane properties that affect transmission of nerve impulses (Desi et al. 1967; EPA 1985a; Gerarde 1959; Savolainen and Pfaffli 1980; Tahti 1992). The mechanism could be either by a disruption of the lipid environment in which membrane proteins function or by direct interaction with the hydrophobic/hydrophilic conformation of proteins in the neuronal membrane. A statistically significant 28% increase in binding (fmol/mg tissue) of radiolabeled *t*-butyl-bicyclophosphorothionate, a ligand to the picrotoxin/convulsant binding stite of $GABA_A$ receptors, was observed in the molecular layer of the cerebellum of rats exposed by inhalation to *m*-xylene at a high concentration (Ito et al. 2002). The results of this study suggest that the high concentration of *m*-xylene in the cerebellum (see Section 3.2.2.1) increased GABA release and/or enhanced $GABA_A$ receptor function, consequently increasing $GABA_A$ receptor binding activity. The increased inhibitory effect of GABAergic neurotransmission in the cerebellum is consistent with the adverse effect of *m*-xylene on motor coordination (Ito et al. 2002).

Changes in levels of various neurotransmitters and lipid composition have been observed in several brain areas following acute- and intermediate-duration exposure to xylene (Andersson et al. 1981; Honma et al. 1983; Savolainen and Seppalainen 1979). It is unclear whether these represent direct effects of xylene or are secondary changes resulting from nonspecific central nervous system depression. Some authors have also suggested that metabolic intermediates, such as arene oxides or methylbenzaldehyde, may be responsible for the toxic effects of xylene (Savolainen and Pfaffli 1980). Oxidation of xylene to these intermediates by microsomal enzyme systems may occur within brain cells (Savolainen and Pfaffli 1980). Acute-duration oral or intermediate-duration inhalation exposures to high concentrations of *p*-xylene, but not the other isomers, results in death of cochlear hair cells and hearing loss in rats (Gagnaire and Langlais 2005; Gagnaire et al. 2001). The reason for the isomer-specificity of the effect is not known, but Gagnaire and Langlais (2005) have indicated that ototoxicity from exposure to aromatic solvents is generally associated with structural constraints: the presence of a single short side chain on the benzene ring. They imply that the *meta*- and *ortho*- placements of a second short side chain in xylene interfere with ototoxic potency, whereas the *para*- position does not. Direct effects of xylene on the brain may be responsible for degraded performance of rats in tests of motor coordination, but the precise brain areas affected have not been identified (Gralewicz and Wiaderna 2001; Gralewicz et al. 1995; Korsak et al. 1990, 1992, 1993, 1994).

Inhibition of pulmonary microsomal enzymes has been observed by several investigators (Elovaara et al. 1987; Foy and Schatz 2004; Foy et al. 1996; Park et al. 1994; Patel et al. 1978; Silverman and Schatz 1991; Smith et al. 1982; Stickney et al. 1989). The exact mechanism of the enzyme inhibition is unknown but has been attributed to the formation of a toxic reactive metabolite (such as methylbenzaldehyde) that binds directly to microsomal protein and inactivates the microsomal enzymes (Patel et al. 1978; Smith et al. 1982). Direct effects on microsomal membrane fluidity and/or lipid content do not appear to be involved (Stickney et al. 1989). Inhalation exposure to *m*-xylene for 6 hours caused a condensation-dependent reduction in cytochromes CYP 2B1, 2E1, and 4B1 in the lung and 2B1 and 2E1 in the nasal mucosa (Vaidyanathan et al. 2003).

The cytotoxicity of *m*-xylene has been associated with reductions in cellular antioxidant status (Coleman et al. 2003). In rat dermal fibroblasts cultured in a collagen matrix (dermal equivalents), exposure to *m*-xylene caused duration- and concentration-related reductions in viability, endogenous catalase activity, and endogenous thiol levels (thiols detected with 5,5-dithiobis[2-nitrobenzoic acid], DTNB). Supplementation with the antioxidant *N*-acetylcysteine, a hydroxyl radical scavenger as well as a

precursor to glutathione, increased survival of cultured fibroblasts exposed to *m*-xylene. These results suggest that hydroxyl radicals may play a role in xylene-induced cytotoxicity.

The mechanism for xylene's toxic effects on the kidneys is also unknown, but may be related to formation of reactive metabolites and subsequent irritation or direct membrane fluidization (EPA 1985a). Support for the former idea is provided in several experiments using porcine proximal tubular cells (LLC-PK1, an established cell line) cultured in the presence of *p*-xylene (Al-Ghamdi et al. 2003a, 2003b, 2004). Treatment for up to 48 hours with 5 mM *p*-xylene reduced cell viability, increased the activity of CYP2E1, and increased the release of malondialdehyde (indicative of lipid peroxidation), but did not cause an increase in DNA fragmentation (Al-Ghamdi et al. 2003a). Disulfuram, an inhibitor of CYP2E1, reduced the activity of this enzyme, as well as the release of malondialdehyde occurring from treatment with *p*-xylene. Experiments using a free radical scavenger (catalase) also counteracted the effect of *p*-xylene. These experiments suggest that renal toxicity of *p*-xylene may be related to its metabolism by CYP2E1, which generates the production of oxidative intermediates and subsequent necrosis; the authors suggest that this mechanism would be relevant to acute high-level exposures to xylene. Results of additional experiments with LLC-PK1 cells treated for 96 hours with 1 mM *p*-xylene suggested that nephrotoxicity from xylene may involve induction of apoptosis through the activation of mitochondrial caspase-9 and caspase-3 (Al-Ghamdi et al. 2003b, 2004); typical features of apoptosis, DNA fragmentation and upregulation of Bax protein compared to Bcl-2 protein (increasing the ratio of Bax/Bcl-2), were observed with xylene treatment. In humans exposed to solvent mixtures containing xylene and a substantial proportion of toluene, a number of effects were noted (lysozymuria and increased urinary excretion of beta-glucuronidase, albumen, erythrocytes and leukocytes) that are attributable to renal injury (Askergren, 1982; Franchini et al. 1983). However, these effects are likely caused by toluene, which is a known renal toxicant.

Developmental toxicity of *m*- and *p*-xylenes in standard animal bioassays is largely related to maternal toxicity (Saillenfait et al. 2003). Effects on fetal body weight and delay of skeletal ossification are both growth related and impaired by compound-induced reductions in maternal body weight. The reason that fetal body weight effects occur at doses lower than the maternal LOAEL for *o*-xylene and mixed xylenes is not known. The fact that mixed xylenes did not elicit the skeletal variants or delayed ossification seen with the individual isomers at the same concentration suggests that the effects of the isomers are not additive. Thus, it is possible that there may be isomer-specific targets relating to skeletal development, as indicated by the slightly different pattern of effects; for example, the incidence of incomplete ossification

in thoracic vertebral centra was significantly elevated in fetuses exposed to *m*-xylene, but not the other isomers.

The mechanism of neurobehavioral deficits observed in rats exposed gestationally following maternal inhalation to *m*-xylene is not known (Hass and Jakobsen 1993; Hass et al. 1995, 1997). Toxicity to vestibular structures involved with balance or hippocampal neurons involved with sensory processing have been suggested as targets associated with impaired motor coordination and spatial navigation following xylene exposure (Hass et al. 1997).

Xylenes have not been shown to be mutagenic in short-term bioassays, but DNA degradation was reported in the skin of rats exposed dermally to *m*-xylene (Rogers et al. 2001). Penetration of liquid xylene may impair the integrity of cell membranes, resulting in release of nucleases from membrane-bound lysosomal stores and concomitant fragmentation of DNA. This process appears to be associated with cell death.

3.5.3 Animal-to-Human Extrapolations

In general, toxic effects of xylenes observed in humans, such as depression of the central nervous system and irritant effects, are also observed in animals. These similarities are to be expected given that they are a consequence of the chemical properties of unmetabolized xylenes and are not likely to be affected by minor species differences in cell type. Toxicokinetic processes, including metabolic pathways for xylenes, are similar in animals and humans, but would be limited by physiological differences in respiration. EPA (1994) has provided guidance for extrapolating from inhalation exposure in animals to human equivalent exposure levels in humans. PBPK models (see Section 3.4.5) have been employed to estimate human equivalent exposures for toxicity in exposed rats.

3.6 TOXICITIES MEDIATED THROUGH THE NEUROENDOCRINE AXIS

Recently, attention has focused on the potential hazardous effects of certain chemicals on the endocrine system because of the ability of these chemicals to mimic or block endogenous hormones. Chemicals with this type of activity are most commonly referred to as *endocrine disruptors*. However, appropriate terminology to describe such effects remains controversial. The terminology *endocrine disruptors*, initially used by Thomas and Colborn (1992), was also used in 1996 when Congress mandated the EPA to develop a screening program for "...certain substances [which] may have an effect produced by a naturally occurring estrogen, or other such endocrine effect[s]...". To meet this mandate, EPA convened a

panel called the Endocrine Disruptors Screening and Testing Advisory Committee (EDSTAC), and in 1998, the EDSTAC completed its deliberations and made recommendations to EPA concerning *endocrine disruptors*. In 1999, the National Academy of Sciences released a report that referred to these same types of chemicals as *hormonally active agents*. The terminology *endocrine modulators* has also been used to convey the fact that effects caused by such chemicals may not necessarily be adverse. Many scientists agree that chemicals with the ability to disrupt or modulate the endocrine system are a potential threat to the health of humans, aquatic animals, and wildlife. However, others think that endocrine-active chemicals do not pose a significant health risk, particularly in view of the fact that hormone mimics exist in the natural environment. Examples of natural hormone mimics are the isoflavinoid phytoestrogens (Adlercreutz 1995; Livingston 1978; Mayr et al. 1992). These chemicals are derived from plants and are similar in structure and action to endogenous estrogen. Although the public health significance and descriptive terminology of substances capable of affecting the endocrine system remains controversial, scientists agree that these chemicals may affect the synthesis, secretion, transport, binding, action, or elimination of natural hormones in the body responsible for maintaining homeostasis, reproduction, development, and/or behavior (EPA 1997b). Stated differently, such compounds may cause toxicities that are mediated through the neuroendocrine axis. As a result, these chemicals may play a role in altering, for example, metabolic, sexual, immune, and neurobehavioral function. Such chemicals are also thought to be involved in inducing breast, testicular, and prostate cancers, as well as endometriosis (Berger 1994; Giwercman et al. 1993; Hoel et al. 1992).

No *in vivo* or *in vitro* studies were located regarding endocrine disruption in human and/or animals after exposure to mixed xylenes or individual isomers of xylene. Evidence for endocrine effects has not been seen in studies on reproductive, developmental, or chronic toxicity of xylenes.

3.7 CHILDREN'S SUSCEPTIBILITY

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

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

Children sometimes differ from adults in their susceptibility to hazardous chemicals, but whether there is a difference depends on the chemical (Guzelian et al. 1992; NRC 1993). Children may be more or less susceptible than adults to health effects, and the relationship may change with developmental age (Guzelian et al. 1992; NRC 1993). Vulnerability often depends on developmental stage. There are critical periods of structural and functional development during both prenatal and postnatal life, and a particular structure or function will be most sensitive to disruption during its critical period(s). Damage may not be evident until a later stage of development. There are often differences in pharmacokinetics and metabolism between children and adults. For example, absorption may be different in neonates because of the immaturity of their gastrointestinal tract and their larger skin surface area in proportion to body weight (Morselli et al. 1980; NRC 1993); the gastrointestinal absorption of lead is greatest in infants and young children (Ziegler et al. 1978). Distribution of xenobiotics may be different; for example, infants have a larger proportion of their bodies as extracellular water, and their brains and livers are proportionately larger (Altman and Dittmer 1974; Fomon 1966; Fomon et al. 1982; Owen and Brozek 1966; Widdowson and Dickerson 1964). The infant also has an immature blood-brain barrier (Adinolfi 1985; Johanson 1980) and probably an immature blood-testis barrier (Setchell and Waites 1975). Many xenobiotic metabolizing enzymes have distinctive developmental patterns. At various stages of growth and development, levels of particular enzymes may be higher or lower than those of adults, and sometimes unique enzymes may exist at particular developmental stages (Komori et al. 1990; Leeder and Kearns 1997; NRC 1993; Vieira et al. 1996). Whether differences in xenobiotic metabolism make the child more or less susceptible also depends on whether the relevant enzymes are involved in activation of the parent compound to its toxic form or in detoxification. There may also be differences in excretion, particularly in newborns who all have a low glomerular filtration rate and have not developed efficient tubular secretion and resorption capacities (Altman and Dittmer 1974; NRC 1993; West et al. 1948). Children and adults may differ in their capacity to repair damage from chemical insults. Children also have a longer remaining lifetime in which to express damage from chemicals; this potential is particularly relevant to cancer.

Certain characteristics of the developing human may increase exposure or susceptibility, whereas others may decrease susceptibility to the same chemical. For example, although infants breathe more air per kilogram of body weight than adults breathe, this difference might be somewhat counterbalanced by their

alveoli being less developed, which results in a disproportionately smaller surface area for alveolar absorption (NRC 1993).

No data are available regarding the effects of exposure to xylenes in children, but it is expected that children would experience the same effects as exposed adults. In controlled acute inhalation experiments on *m*-xylene, one reason offered for the greater impairment in respiratory function (reduced forced vital capacity) in exposed women to compared to exposed men was that the airways in women are narrower and therefore more sensitive to swelling (Ernstgard et al. 2002). On this basis, it is likely that children exposed to xylenes by inhalation would be more sensitive to respiratory impairment than exposed adults.

A few epidemiological studies on laboratory workers suggested that maternal exposure to xylenes might increase the frequency of spontaneous abortion, but the validity of the association is confounded by simultaneous exposure to other chemicals (Lindbohm et al. 1990; Taskinen et al. 1994).

The lipophilic properties of xylenes suggest that the absorption and distribution in children are likely to be similar to those of adults. PBPK modeling estimated that about 1–2% of xylene absorbed from maternal occupational inhalation exposure at 100 ppm would be transferred to breast milk (Fisher et al. 1997). The daily intake of xylene by a 10-kg nursing infant under this scenario was estimated to be 6.59 mg. Studies in mice have demonstrated that xylene absorbed by pregnant females is transferred to the fetus, but that uptake by fetal tissues was lower than in maternal tissues (Ghantous and Danielsson 1986). The most reliable developmental toxicity assay in rats exposed by inhalation indicated that fetotoxicity (reduced fetal weight and delayed ossification, a skeletal variation that is affected by impaired growth) from maternal exposure to *m*- and *p*-xylene only occurs at maternally toxic exposure levels (Saillenfait et al. 2003); exposure to *o*-xylene or mixed xylenes resulted in fetal body weight effects at concentrations below the LOAEL for maternal effects. The pattern of skeletal variations (i.e., the bones affected) was slightly different for each isomer and no serious malformations were observed at concentrations as high as 2,000 ppm. No significant increase in skeletal variations was noted for gestational exposure to mixed xylenes, suggesting that the effect of the isomers was not additive (Saillenfait et al. 2003). Other developmental inhalation studies reported skeletal variations and delayed ossification at lower concentrations, but the results were not considered for determining NOAEL or LOAEL values because fetal effects were not reported on a per-litter basis and little information on maternal toxicity was provided (see Section 3.2.1.6). Fetal toxicity in oral rodent studies occurred at relatively high doses near the maternal effect level (Marks et al. 1982). A potentially serious developmental effect of xylene is impaired neurobehavioral function, which has been observed in rats

gestationally exposed at a level (500 ppm) that was not toxic to dams (Hass and Jakobson 1995; Hass et al. 1997). In these studies, motor coordination was impaired and deficiencies in a water maze test for spatial navigation persisted to postnatal week 28 in the offspring of exposed dams. These results suggest that in humans, excess maternal exposure to xylene might result in postnatal neurobehavioral deficits even if no overt structural anomalies occurred.

The main phase I metabolizing enzyme for xylene, CYP2E1, is present in the fetus and in children. The presence of CYP2E1 facilitates the elimination of xylene from the body. Total fetal liver CYP content is a relatively constant 30% of the adult level from the end of the first trimester of gestation up to 1 year of age (EPA 2001b). mRNA for CYP2E1 has been detected in human first-trimester placentas (Hakkola and Saarinen 1996). Low levels of CYP2E1 protein have been detected in human fetal brain as early as gestational day 46, substantially increasing around day 50 (Boutelet-Bochan et al. 1997; Brzezinski et al. 1999). In the fetal liver, CYP2E1 protein was not detectable at 10 weeks of gestation, but was present at 16 weeks (Carpenter et al. 1996). Additionally, there is some evidence that maternal alcohol consumption induces placental CYP2E1 in humans (Rasheed et al. 1997). Hepatic levels of CYP2E1 mRNA increase significantly during the first 24 hours after birth, largely resulting from demethylation that allows transcription to proceed (Vieira et al. 1996). Major accumulations of CYP2E1 occur between 1 and 3 months of age and values comparable to those of adults are achieved sometime between 1 and 10 years of age (EPA 2001b; Vieira et al. 1996). Thus, elimination of xylene metabolites in exposed children would be expected to be similar to exposed adults.

Aside from the model described above (and in Section 3.4.5) for lactational transfer to nursing infants by Fisher et al. (1997), no PBPK models have been developed for children or for fetuses or pregnant women exposed to xylenes.

Ginsberg et al. (2004) considered developmental and pharmacokinetic differences between adults and children that could impact risk assessments of children exposed to toxicants. The higher inhalation rate per respiratory surface area in children compared to adults might result in an apparent higher dose to the lungs in children for the first several years compared to adults exposed to xylene at the same atmospheric concentration. As the stratum corneum of the epidermis is well-developed in newborn infants, dermal absorption of xylene by newborns would be generally expected to be similar to adults. The one exception is for the infants born prematurely, in which the stratum corneum attains a normal thickness only by the second postnatal week. The relatively larger ratio of liver weight to body weight in children, especially during the first 2 years postnatally, and the lower body lipid content for the first 3 months postnatally,

compared to adults, would suggest that retention of absorbed xylene is lower and systemic clearance of absorbed xylene is faster in young children than in adults. These factors are countered by the possibly lower rates of metabolism of xylenes during the first two postnatal months (see above). The larger brain weight to body weight ratios in children, especially during the first two postnatal years, suggests that proportionally more xylene may be delivered to the brain of children, rendering them more vulnerable to the neurotoxic effects of xylene than similarly exposed adults. No compound-specific experimental data are available to confirm these suppositions regarding the exposure of children to xylenes.

3.8 BIOMARKERS OF EXPOSURE AND EFFECT

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

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

Biomarkers of effect are defined as any measurable biochemical, physiologic, or other alteration within an organism that, depending on magnitude, can be recognized as an established or potential health impairment or disease (NAS/NRC 1989). This definition encompasses biochemical or cellular signals of tissue dysfunction (e.g., increased liver enzyme activity or pathologic changes in female genital epithelial cells), as well as physiologic signs of dysfunction such as increased blood pressure or decreased lung

capacity. Note that these markers are not often substance specific. They also may not be directly adverse, but can indicate potential health impairment (e.g., DNA adducts). Biomarkers of effects caused by xylene are discussed in Section 3.8.2.

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

3.8.1 Biomarkers Used to Identify or Quantify Exposure to Xylene

Xylene levels in the blood and levels of its metabolite, methylhippuric acid, in the urine are the primary markers used to detect exposure to xylene. Xylene is very soluble in the blood and is readily absorbed into the circulation during exposure (Astrand 1982). Measurement of blood levels of xylene is limited by the rapid metabolism of xylene. Moreover, there are no data on background concentrations of xylene in blood or urine. Xylene and xylene isomers have been detected at concentrations of up to 400 nM in urine of workers exposed to xylene at geometric mean concentration of 1.9 ppm with a maximum concentration of 27.3 ppm (Takeuchi et al. 2002). Xylenes are metabolized almost exclusively to methylhippuric acids in humans. Detection of methylhippuric acid in the urine is the most widely used indicator of xylene exposure (ACGIH 1986). A strong association has been shown between urinary methylhippuric acid concentrations and exposure to xylene (Daniell et al. 1992; Jonai and Sato 1988; Kawai et al. 1991); during an 8-hour workshift, a concentration of 57.8 mg/L of methylhippuric acid isomers (i.e., all isomers combined) was found to correlate with exposure to 3.8 ppm (geometric mean concentration) of total xylenes (Kawai et al. 1991). In a study of Chinese men and women occupationally exposed to mixed xylenes, Inoue et al. (1993) estimated that 13 mg of methylhippuric acid would be excreted in a liter of urine for each ppm of xylene exposure (or 11.1 mg/g creatinine/ppm). This relationship was true for both men and women as well as for mixed and individual isomers. Within 2 hours of an inhalation exposure, methylhippuric acid may be detected in the urine (Sedivec and Flek 1976b). The excretion of methylhippuric acid is complete within 1 or 2 days of exposure to xylene, limiting the utility of this biomarker to the detection of only very recent exposures. With chronic exposure to xylene, the metabolism is enhanced, further limiting the time following exposure that xylene levels may be measured in the blood (Savolainen et al. 1979a). Since the methylhippuric acid background levels in persons not exposed to xylenes are very low, methylhippuric acids are specific markers for xylenes, except for

exposure to alkyl toluenes in which the number of carbon atoms in the alkyl group is odd. A minor metabolite of xylene, *N*-acetyl-*S*-xylyl cysteine (a trioether), may also be detected in the urine (Tanaka et al. 1990; van Doorn et al. 1980); however, it is at such low levels in the urine during experimental exposures that it is ineffective as a biomarker (Norstrom et al. 1988). For additional information on the kinetics of xylene absorption, distribution, metabolism, or excretion, see Section 3-4.

3.8.2 Biomarkers Used to Characterize Effects Caused by Xylene

The following changes are potential biomarkers of effect for xylenes; however, none of the changes are unique to xylene exposure. Xylenes have been observed to enhance the activity of a variety of microsomal enzymes and increase hepatic cytochrome P-450 content (Elovaara 1982; Elovaara et al. 1980; Patel et al. 1979; Savolainen et al. 1978; Tatrai et al. 1981; Toftgard and Nilsen 1981, 1982; Toftgard et al. 1981). Scores consistent with memory impairment and decreased reaction time have been observed using standard intelligence tests and measures of reaction time (Gamberale et al. 1978; Riihimaki and Savolainen 1980; Savolainen and Riihimaki 1981a; Savolainen et al. 1979b, 1984, 1985a). These effects are likely to be elicited by any solvent that has anaesthetic properties. Erythema of the skin is a biomarker of dermal exposure to xylene, but this is a common reaction to compounds with irritant properties (Ahaghotu et al. 2005; Anderson et al. 1986; Chatterjee et al. 2005; Hine and Zuidema 1970; Pound and Withers 1963; Smyth et al. 1962). Increases in blood levels of interleukin 1-alpha, as demonstrated in rats, may represent a biomarker of inflammatory processes in skin following dermal exposure to xylene, but might apply to any chemical that elicits dermal irritation (Ahaghotu et al. 2005; Chatterjee et al. 2005).

3.9 INTERACTIONS WITH OTHER CHEMICALS

The interaction of xylene with alcohol, drugs (aspirin, phenobarbitol), and various solvents (1,1,1-trichlorethane, benzene, toluene, ethylbenzene, methyl ethyl ketone) has been evaluated in experimental studies with humans and animals. Xylene has a high potential to interact with numerous substances because the isomers induce microsomal enzymes in the liver (Blanchard and Morris 1994; Liira et al. 1991), while microsomal enzymes in the lungs are inhibited by xylene exposure (Blanchard and Morris 1994; Elovaara et al. 1987; Patel et al. 1978; Silverman and Schatz 1991; Toftgard and Nilsen 1982). Which enzymes will be affected is isomer dependent. For example, *m*-xylene is a more potent inducer of P-450 2B enzymes than *p*-xylene (Backes et al. 1993). The isomer differences, as well as organ differences in effects on xenobiotic metabolizing enzymes, make it difficult to predict the interaction of xylene with other substances.

The effects from combined exposure to xylene and ethanol have been studied most extensively because of the reasonable expectancy that some workers will consume alcoholic beverages and subsequently might be exposed to xylene occupationally by inhalation. Results of studies with humans and animals indicate that metabolic interaction between xylene and ethanol occurs. Co-administered ethanol appears to inhibit the metabolism of xylene, probably by competition for mixed function oxidases, resulting in elevated blood levels of xylene and decreased excretion of methylhippuric acid (Elovaara et al. 1980; Riihimaki et al. 1982a, 1982b; Romer et al. 1986; Savolainen 1980; Savolainen et al. 1978, 1979b, 1980b). A kinetic study in rats (Kaneko et al. 1993) suggests that ethanol inhibition of xylene metabolism occurs only at high airborne concentrations (500 ppm). Paradoxically, ethanol pretreatment has a synergistic effect on metabolism of xylene as a result of the induction of microsomal enzymes in the liver (Wisniewska-Knypl et al. 1989). Ethanol pretreatment (137 g taken orally) enhanced the metabolism of *m*-xylene in human subjects exposed at 400 ppm, but had no effect at 100 ppm (Tardif et al. 1994). This would enhance the metabolic capacity of the liver and modify biological effects of other chemicals that are either detoxified or converted to toxic metabolites by the microsomal enzymes. In summary, it cannot be stated with certainty whether alcohol and xylene would interact to produce synergistic or antagonistic effects in humans and animals because there are reasons why both would occur. MacDonald et al. (2002) used PBPK modeling to evaluate the likelihood of central nervous system effects in people exposed to a time-weighted-average concentration of 100 ppm *m*-xylene for 8 hours and also exposed to ethanol. In the model simulations, increasing exposure to ethanol increased the probability of central nervous system effects from exposure to *m*-xylene. Combined exposure to *m*-xylene and ethanol increased lipid peroxidation in the rat liver (Jajte et al. 2003).

Kaneko et al. (1995) modeled the effect of phenobarbital pretreatment (80 mg/kg/day for 3 days) with exposure to *m*-xylene orally or intraperitoneally at 8.6 or 86.4 mg/kg or by inhalation for 6 hours at 40 or 400 ppm. Phenobarbital treatment increased the metabolism of inhaled *m*-xylene at 400 ppm. This was related to a decrease in xylene blood concentrations and increased urinary excretion. Phenobarbital had no significant effect on metabolism of ingested xylene. The metabolism of *m*-xylene was not affected by coexposure to *n*-butyl alcohol in rats exposed by inhalation for 7 hours to 100 ppm of each chemical (Swiercz et al. 1995).

Combined exposure to ethanol and xylene results in macrocytosis and decreased erythrocyte membrane fluidity (Wronska-Nofer et al. 1991). These effects were not observed when either chemical was

administered alone. It is unclear whether this interaction is pharmacological or pharmacokinetic in nature.

Acute inhalation exposure to a mixture of toluene and xylene resulted in more-than-additive respiratory and central nervous system toxicity (Korsak et al. 1988, 1992). Elevated blood levels of xylene and toluene and decreased excretion of the major metabolites of xylene and toluene in the urine (Tardif et al. 1992) suggest mutual metabolic inhibition. However, simultaneous exposures in humans indicate that a threshold exists for this interaction (Tardif et al. 1991). No increase in blood levels of these substances was observed during combined exposures to 50 ppm toluene and 40 ppm xylene over 3 consecutive days, whereas increases in blood levels and levels in exhaled air were observed during a combined 4-hour exposure to 95 ppm toluene and 80 ppm xylene. Thus, combined exposures at below threshold level are unlikely to produce greater than additive toxicity (Tardif et al. 1991). Physiologically based toxicokinetic modeling studies using rat data suggests that the interaction between toluene and xylene is competitive, with toluene a more potent inhibitor of xylene metabolism than xylene is of toluene metabolism (Tardif et al. 1993a, 1993b, 1995). PBPK modeling studies of ternary mixtures predicted that humans exposed to permissible levels for the mixture (33 ppm *m*-xylene, 17 ppm toluene, and 33 ppm ethylbenzene), would not exhibit biologically significant metabolic inhibition of the components (Tardif et al. 1997). In these ternary simulations, after 0.5–8 hours of exposure, the concentration of xylene in alveolar air was slightly, but significantly higher than expected compared to exposure of xylene alone; the blood concentration of xylene following exposure to the ternary mixture was slightly elevated in venous blood after 5.5–8 hours of exposure. Studies in rats exposed to binary or ternary mixtures (*m*-xylene and toluene with or without ethylbenzene) at component concentrations of 100 or 200 ppm demonstrated that the kinetics of metabolism was less affected by metabolic interactions when metabolism was near saturation (Tardif et al. 1996).

Exposure to xylene combined with benzene or ethylbenzene may also produce mutual inhibition of the metabolism of both solvents (Engstrom et al. 1984; Nakajima and Sato 1979b). Ethylbenzene is found in commercial xylene. In contrast, ethyl acetate exposure in combination with exposure to *m*-xylene caused a reduction in blood xylene levels (Freundt et al. 1989).

Combined exposures to *m*-xylene and methyl ethyl ketone (2-butanone) produced a synergistic induction of microsomal enzymes in rats (Liira et al. 1991), but in humans, inhibited the metabolism of *m*-xylene to methylhippuric acid, resulting in corresponding increases in levels of xylene in blood and fat (Liira et al. 1988, 1991). While the side-chain oxidation of xylene to methylhippuric acid was inhibited, an increase

in ring oxidation (xylenol production) was observed (Liira et al. 1991), indicating that the inhibition was specific to a particular oxidation reaction. Thus, it is not known as to whether 2-butanone and *m*-xylene would interact to produce additive or antagonist effects in humans and animals.

Inhalation of *m*-xylene following pretreatment with phenobarbital was associated with both increased pulmonary retention of *m*-xylene and increased urinary excretion of *m*-methylbenzoic acid (David et al. 1979). Combined inhalation of 1,1,1-trichloroethane (methylchloroform) and *m*-xylene had no effect on the metabolism of *m*-xylene, as indicated by blood concentrations of the parent compound and urinary concentrations of *m*-methylhippuric acid (Tardif and Charest-Tardif 1999). Surprisingly, inhalation of *m*-xylene and 1,1,1-trichloroethane has been associated with slight improvements in certain psychophysiological parameters, including reaction time and equilibrium in humans as compared with pre-exposure measurements (Savolainen et al. 1982a, 1982b), and impairment in others such as visual evoked potentials and equilibrium (Savolainen et al. 1982a; Seppalainen et al. 1983). Also, a protective effect of xylene on *n*-hexane-induced testicular atrophy and peripheral nerve effects were observed when rats were exposed to *n*-hexane and xylene simultaneously (Nylen and Hagman 1994; Nylen et al. 1989), although combined exposure to xylene and *n*-hexane increased loss of auditory sensitivity (Nylen and Hagman 1994; Nylen 1996). Bromobenzene, which requires metabolic activation, showed greater toxicity to the liver in *p*-xylene exposed rats, while lung toxicity was not affected (Day et al. 1992).

Following concurrent exposure to 100 ppm *m*-xylene and 1,500 mg aspirin orally in human subjects, the urinary excretion of metabolites conjugated to glycine (*m*-methylhippuric acid and salicyluric acid, respectively) was reduced by 50% for both compounds (Campbell et al. 1988). The authors speculate that the cause was competition for the enzymes involved in conjugation with glycine (acyl-CoA synthetase and glycine N-acylase), resulting in saturation of the conjugation pathway for both chemicals (Campbell et al. 1988). Administration of aspirin to pregnant rats during inhalation exposure to xylene caused greater-than-additive potentiation of maternal and fetal toxic effects (Ungvary 1985). This was postulated to be due to the interference with metabolism of aspirin by xylene and vice versa.

Oral exposure to a relatively high dose of sodium benzoate, a food additive, 3 hours before intraperitoneal injection of *o*-xylene altered the metabolism of *o*-xylene in rats (Moriwaki et al. 2005). Pre-treatment with sodium benzoate (more than twice the amount of *o*-xylene on a molar basis) reduced the excretion of *o*-methylhippuric acid but increased the excretion of *o*-toluic acid conjugates and unmodified *o*-xylene in urine. These changes were attributed to competition for the enzymes involved in glycine conjugation (benzoyl-Coenzyme A synthetase and benzoyl-CoA:glycine N-acyltransferase).

Exposure to xylene has been shown to inhibit several microsomal enzymes in the lung (Blanchard and Morris 1994; Elovaara et al. 1987; Patel et al. 1978; Silverman and Schatz 1991; Toftgard and Nilsen 1982). *m*-Xylene administered intraperitoneally or exposure to *o*-xylene by inhalation altered the pulmonary microsomal metabolism of benzo[a]pyrene in rats, resulting in inhibition of its detoxification and increased production of toxic, mutagenic metabolites (bay region diols) (Park and Schatz 1999; Stickney et al. 1991). One study suggested that xylene acts as a promotor or cocarcinogen for the induction of skin tumors in mice, but the purity of the tested material was not reported (Pound 1970). If verified, these findings could be relevant in combined human exposures to xylene and polyaromatic hydrocarbons present in cigarette smoke and combustion emissions and especially to petrochemical workers who could be exposed to xylene, crude oils (promotor), and ultraviolet light (initiator).

Dilution in polyethylene glycol 600 (PEG 600) increased the severity of eye irritation elicited by xylene and other aromatic compounds in rabbits compared to the undiluted compounds (Kennah et al. 1989). Undiluted xylene (not specified but presumed to be mixed isomers) elicited only minimal eye irritation whereas dilutions in PEG 600 resulted in slight or moderate irritation (3–5-fold increases in Draize scores); similar increases were observed with toluene and styrene. Because other diluents (cellosolve acetate or methyl isobutyl ketone) did not have the same effect, the study authors proposed that the aromatic compounds interacted with PEG 600 to form a more potent ocular irritant.

In addition to interacting with other chemicals, exposure to xylene at high concentrations has also been shown to increase the effects of a virus. Acute exposure of mice to 1,208 ppm (but not 595 ppm) *p*-xylene (4 days, 4 hours/day) increased the mortality resulting from the murine cytomegalovirus (Selgrade et al. 1993). This effect was a result of potentiation of the liver damage cause by the virus rather than an immunological effect.

3.10 POPULATIONS THAT ARE UNUSUALLY SUSCEPTIBLE

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

Available data indicate that subsets of the human population may be unusually susceptible to the toxic effects of xylene. Pregnant women, fetuses, and very young children may be at greater risk of adverse health effects from xylene exposure than the population in general (Barlow and Sullivan 1982; Holmberg and Nurminen 1980; Hudak and Ungvary 1978; Kucera 1968; Marks et al. 1982; Mirkova et al. 1983; Ungvary et al. 1980b, 1981). Although no human studies were located indicating maternal or fetal toxicity following mixed xylene exposure, animal studies that involved exposure to *m*-xylene and aspirin or xylene alone suggest there may be a relationship between exposure to the agents and developmental effects (Hudak and Ungvary 1978; Marks et al. 1982; Ungvary 1985; Ungvary et al. 1980b, 1981). In summary, although it is not clear how toxic xylene might be to fetuses and infants, for safety's sake caution is urged. The ability of fetuses and very young children to metabolize certain xenobiotics, including possibly xylene, is reduced because of their immature enzyme detoxification systems (Calabrese 1978). Thus, for pregnant women exposed to xylene, ingestion of aspirin is likely to potentiate adverse effects of xylene in both the mother and the offspring.

People with subclinical and clinical epilepsy are at increased risk of seizures if exposed to xylene because of its excitatory central nervous system effects (Arthur and Curnock 1982; Goldie 1960; Riihimaki and Hanninen 1987). It has also been demonstrated in human (Goldie 1960; Riihimaki et al. 1982a; Savolainen 1980; Savolainen et al. 1978, 1980b) and animal studies (Elovaara et al. 1980; Savolainen et al. 1979b) that alcohol consumption potentiates xylene toxicity. Some people appear particularly susceptible to the interaction and may develop dizziness, nausea, and dermal flush (Riihimaki et al. 1982b; Savolainen et al. 1980b).

People with clinical or subclinical renal, hepatic, or cardiac disease may be more susceptible to the effects of xylene. Evidence from occupational and case studies indicates that exposure to high levels of xylene might cause renal impairment and some hepatic effects, as well as cardiac manifestations, including tachycardia and ECG abnormalities (Goldie 1960; Hipolito 1980; Morley et al. 1970; NIOSH 1975; Von Burg 1982). However, exposure to xylene in these studies was confounded with exposure to other chemical agents.

Limited human data suggest that people with respiratory diseases, such as asthma, could potentially be at risk with regard to the adverse effects of xylene following inhalation exposure (Hipolito 1980; Morley et al. 1970).

3.11 METHODS FOR REDUCING TOXIC EFFECTS

This section will describe clinical practice and research concerning methods for reducing toxic effects of exposure to xylene. 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 xylene. When specific exposures have occurred, poison control centers and medical toxicologists should be consulted for medical advice. The following texts provide specific information about treatment following exposures to xylene:

Ellenhorn MJ, Barceloux DG, eds. 1988. Medical toxicology: Diagnosis and treatment of human poisoning. New York, NY: Elsevier, 1422-1426, 1730-1731.

IPCS. 1992. Xylene. Poisons information. Monograph 565. International Programme on Chemical Safety. http://www.inchem.org/documents/pims/chemical/xylene.htm. April 7, 2005.

3.11.1 Reducing Peak Absorption Following Exposure

General recommendations reported for reducing absorption following acute high-dose exposure to xylene include removal of the patient from the source of exposure to fresh air and decontamination of the skin with mild soap and water (Bronstein and Currance 1988; Ellenhorn and Barceloux 1988; Goldfrank et al. 1990; Stutz and Janusz 1988). When the eyes have been involved, copious rinsing with tepid water or normal saline has been used for decontamination (Bronstein and Currance 1988; Stutz and Janusz 1988).

Strategies for reducing xylene absorption following ingestion are limited because light petroleum products such as xylene cause severe aspiration pneumonitis (Ellenhorn and Barceloux 1988; IPCS 1992). Gastric lavage is the preferred method recommended for emptying the stomach contents, as long as a cuffed endotracheal tube is used to limit the possibility of aspiration (Goldfrank et al. 1990). Induction of emesis (vomiting) with syrup of ipecac is generally not recommended, especially in pediatric cases (Goldfrank et al. 1990); emesis is contraindicated if unprovoked emesis has already occurred or if the patient is not alert or has an impaired gag reflex (Ellenhorn and Barceloux 1988; Goldfrank et al. 1990). The only circumstances in which emesis is recommended are when very large quantities have been ingested, such as in a suicide attempt (Ellenhorn and Barceloux 1988; Goldfrank et al. 1990), or another highly toxic substance has been ingested together with xylene (Goldfrank et al. 1990). In summary, gastric lavage is recommended to reduce xylene absorption from the gastrointestinal tract only when one is certain that aspiration is not likely to occur.

Although the use of activated charcoal and/or cathartics to limit intestinal absorption is recommended in some treatment protocols (Stutz and Janusz 1988), their use has been reported to be equivocal (Ellenhorn and Barceloux 1988). No studies have shown that activated charcoal is effective in adsorbing petroleum distillates or that cathartics are effective in speeding excretion (Goldfrank et al. 1990). Furthermore, because of low viscosity, oil-based cathartics may increase aspiration pneumonitis and absorption (Goldfrank et al. 1990).

3.11.2 Reducing Body Burden

In acute exposure situations, most xylene absorbed by the body is excreted in the urine or exhaled air within a day after exposure (see Section 3.4.4). However, charcoal hemoperfusion has been used to speed the removal of xylene from the body and to reverse its acute toxicity (Recchia et al. 1985). Sevcik et al. (1992) also used hemoperfusion and hemodialysis in an attempt to speed removal of xylene. Whether the relative gain from these treatment methods is worth the potential risks of invasive treatment remains to be established. A small percentage of absorbed xylene is retained in body fat. It has been suggested that over a prolonged period of exposure, significant amounts of xylene could accumulate in adipose tissue (Astrand 1982; Engstrom and Bjurstrom 1978). However, xylene has been shown to induce its own metabolism with the result that greater amounts of metabolites are excreted and less is available for storage (Elovaara et al. 1989; Savolainen et al. 1979a). No information was located regarding methods for reducing adipose stores of xylene; removal of xylene from adipose tissue is slow, limited by the lipophilic nature of the chemical and the poor vascular perfusion of adipose tissue (Riihimaki et al. 1979b). Use of agents known to induce microsomal enzyme activity is a possible experimental method for enhancing excretion of xylene released from adipose stores by increasing the gradient of diffusion.

3.11.3 Interfering with the Mechanism of Action for Toxic Effects

No information was located on established therapies designed to interfere with the mechanism of action of xylene. However, some speculation is possible regarding areas for future research in this regard. For example, the central nervous system toxicity of xylene is believed to be similar to that produced by other nonspecific central nervous system depressants (Desi et al. 1967; EPA 1985a; Gerarde 1959; Savolainen and Pfaffli 1980; Tahti 1992). If circulating xylene levels could be reduced, then the central nervous system toxicity may likewise be reduced (see Section 3.11.2).

Since the exact metabolite responsible for the pulmonary toxicity of xylene has not been identified, it is difficult to speculate on steps to avert its synthesis or speed its excretion. In animals, selective

inactivation of enzymes can result in damage to tissue caused by the toxic metabolite of xylene, methylbenzaldehyde. This effect has not been confirmed in humans. Decreased pulmonary microsomal enzyme activity was seen in rats administered a single dose or repeated doses of *p*-xylene for 3 weeks (Elovaara et al. 1989; Patel et al. 1978). However, the inhibition of pulmonary microsomal enzymes decreases to some extent with continued exposure to xylene (Silverman and Schatz 1991); this may indicate that xylene-induced activation of metabolizing enzymes and thereby acceleration of its own metabolism (Elovaara et al. 1989) may be limiting the production of the toxic metabolite.

At least one *in vitro* study indicated that cytotoxicity caused by *m*-xylene is associated with reductions in cellular antioxidants and that supplementation with *N*-acetylcysteine partially ameliorates these effects (Coleman et al. 2003). *N*-acetylcysteine has been employed as a therapeutic antagonist for acetaminophen poisoning, but its effectiveness for reducing toxicity of xylenes *in vivo* has not been assessed.

The available information on the mechanisms of renal and fetotoxicity is insufficient to allow speculation on potential means for blocking these effects.

3.12 ADEQUACY OF THE DATABASE

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

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

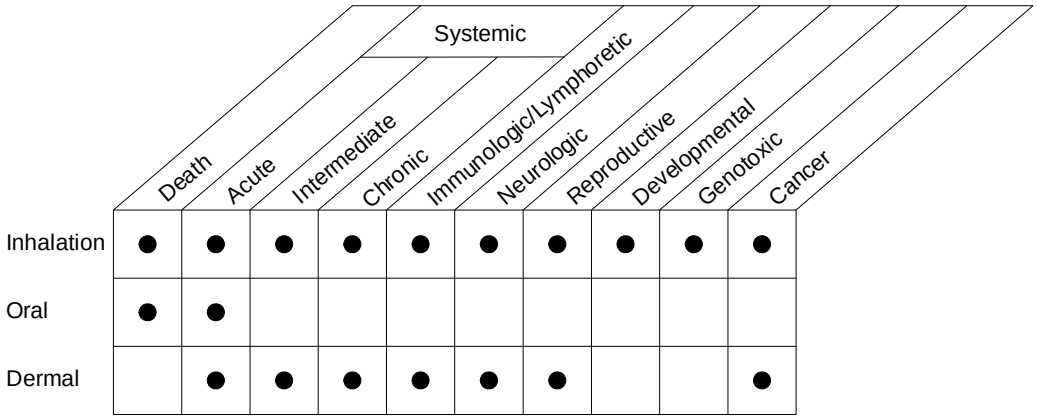
3.12.1 Existing Information on Health Effects of Xylene

The existing data on health effects of inhalation, oral, and dermal exposure of humans and animals to xylene are summarized in Figure 3-9. The purpose of this figure is to illustrate the existing information concerning the health effects of xylene. Each dot in the figure indicates that one or more studies provide information associated with that particular effect. The dot does not necessarily imply anything about the quality of the study or studies, nor should missing information in this figure be interpreted as a "data need". A data need, as defined in ATSDR's *Decision Guide for Identifying Substance-Specific Data Needs Related to Toxicological Profiles* (Agency for Toxic Substances and Disease Registry 1989), is substance-specific information necessary to conduct comprehensive public health assessments. Generally, ATSDR defines a data gap more broadly as any substance-specific information missing from the scientific literature.

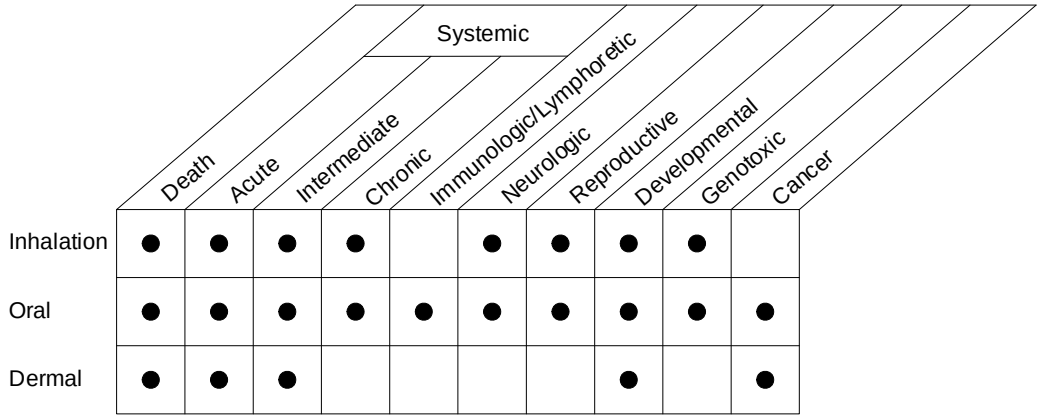
People may be exposed to xylene at hazardous waste sites by inhalation of contaminated air, drinking contaminated water, or dermal contact with contaminated water or subsurface soils and sediments. Volatilization of xylene from surface water and soil occurs rapidly; therefore, inhalation is the most likely route of exposure to xylene at these sites. The human health effects of xylene by inhalation exposure have been studied to the greatest extent. There is little information available regarding health effects in humans following oral or dermal exposure to xylene. The bulk of the information on health effects in humans associated with dermal exposure comes from reports of occupational exposures, which are likely to be combined inhalation and dermal exposures. As noted above, ingestion of xylene may be of concern because of the potential for xylene to contaminate sources of drinking water (groundwater) and certain soils. Dermal exposure to xylene is of concern not only because of potential workplace exposures, but also because members of the general public are potentially exposed to xylene contained in paints, glues, and other household products. As noted above, dermal exposure to soils and water contaminated with xylene at waste sites could also occur.

Human fatalities following both inhalation and ingestion of xylene have been reported in the literature. Acute inhalation exposure of humans to xylene has resulted in hepatic and cardiovascular effects as well as neurologic effects. Very limited data regarding the systemic health effects of intermediate-duration human exposure to xylene were located in the literature. Also, very limited human carcinogenicity data were reported in the literature. Very little information is available on the chronic systemic, immunologic, developmental, reproductive, and genotoxic health effects of xylene exposure in humans. Interpretation of the large number of human studies examining the health effects of inhaled xylene vapor is difficult

Figure 3-9. Existing Information on Health Effects of Total Xylenes



Human



Animal

● Existing Studies

because of study design limitations, such as inadequate characterization of exposure and concurrent exposure to other solvents such as toluene and benzene.

Studies conducted on experimental animals have been fairly extensive (Figure 3-9) and have focused on the adverse health effects following inhalation and oral exposure to xylene. Data are comprehensive on neurological and systemic effects. There are several developmental studies in animals, although most have limitations. Limited information exists on the carcinogenicity of mixed xylenes. A large number of studies on the genotoxicity of xylene are available, with the majority reporting negative results.

3.12.2 Identification of Data Needs

Acute-Duration Exposure. There are acute exposure data in humans and/or animals that indicate that the central nervous system (Andersson et al. 1981; Arthur and Curnock 1982; Bushnell 1989; Carpenter et al. 1975a; De Ceaurriz et al. 1983; Dudek et al. 1990; Dyer et al. 1988; Ernstgard et al. 2002; Furnas and Hine 1958; Gamberale et al. 1978; Ghosh et al. 1987; Klaucke et al. 1982; Korsak et al. 1988; Martinez et al. 1989; Molnar et al. 1986; Morley et al. 1970; Muralidhara and Krishnakumari 1980; Nersesian et al. 1985; NIOSH 1981; NTP 1986; Padilla and Lyerly 1989; Pryor et al. 1987; Savolainen and Linnavuo 1979; Savolainen et al. 1978, 1979b, 1984, 1985a; Seppalainen et al. 1989; Wimolwattanapun et al. 1987) and possibly the developing fetus (Balogh et al. 1982; Hudak and Ungvary 1978; Marks et al. 1982; Ungvary 1985; Ungvary and Tatrai 1985; Ungvary et al. 1980b, 1981) are the major targets of acute xylene toxicity by the inhalation and oral routes. Limited information is available on the nervous system effects of dermal exposure to xylenes (Goldie 1960; Hipolito 1980; Kilburn et al. 1985; Roberts et al. 1988). Death has been observed to occur as a result of exposure by inhalation, oral, and dermal exposure, and lethal and nonlethal levels of total xylenes have been determined (Abu Al Ragheb et al. 1986; Bonnet et al. 1979; Cameron et al. 1938; Carpenter et al. 1975a; Condie et al. 1988; Dyer et al. 1988; Furnas and Hine 1958; Gerarde 1959; Harper et al. 1975; Hine and Zuidema 1970; Morley et al. 1970; Muralidhara and Krishnakumari 1980; NTP 1986; Pound and Withers 1963; Smyth et al. 1962; Ungvary et al. 1980b; Wolf et al. 1956). Acute studies have demonstrated that xylene is irritating to the skin and eyes (Anderson et al. 1986; Carpenter et al. 1975a; Consumer Products Testing 1976; De Ceaurriz et al. 1981; Engstrom et al. 1977; Ernstgard et al. 2002; Food and Drug Research Labs 1976a; Hine and Zuidema 1970; Klaucke et al. 1982; Nelson et al. 1943; Nersesian et al. 1985; NIOSH 1981; Pound and Withers 1963; Riihimaki 1979b; Smyth et al. 1962; Wolf et al. 1956). Inhalation of xylenes has also been shown to cause irritation of the respiratory tract and dyspnea (Carpenter et al.

1975a; De Ceaurriz et al. 1981; Ernstgard et al. 2002; Furnas and Hine 1958; Klaucke et al. 1982; Korsak et al. 1988; Morvai et al. 1976; Nelson et al. 1943; Nersesian et al. 1985; NIOSH 1981).

Data were sufficient to determine an acute-duration inhalation MRL for mixed xylenes based on increased respiratory effects (slight irritation and impaired lung function) and subjective neurological symptoms (headache, dizziness, feeling of intoxication) in humans exposed to *m*-xylene (Ernstgard et al. 2002). The acute oral MRL for mixed xylenes is based on a NOAEL for neurological effects in rats exposed to *p*-xylene (Dyer et al. 1988). Additional information on the effects observed after acute dermal exposure is needed due to the likelihood that acute duration skin contact with xylenes could occur in the home, workplace, and possibly at hazardous waste sites. Pharmacokinetic data and toxicity data indicate that xylene is absorbed through the skin (Dutkiewicz and Tyras 1968; Engstrom et al. 1977; Kezic et al. 2001; McDougal et al. 1990; Morgan et al. 1991; Riihimaki 1979b; Riihimaki and Pfaffli 1978; Skowronski et al. 1990), although the relative absorption by this route is difficult to ascertain because of the rapid evaporation of xylenes from the skin. A PBPK model has been developed for dermal exposure (Thrall and Woodstock 2003). Additional acute-duration inhalation and oral studies clarifying which nervous system effects are the most sensitive are needed to provide critical, reliable guidance values for acute exposure.

Intermediate-Duration Exposure. Intermediate-duration inhalation, oral, and dermal studies have identified the central nervous system (Condie et al. 1988; Goldie 1960; Honma et al. 1983; Jenkins et al. 1970; NTP 1986; Pryor et al. 1987; Rank 1985; Savolainen and Seppalainen 1979; Savolainen et al. 1979a), liver (Condie et al. 1988; Elovaara et al. 1989; Ungvary 1990), kidneys (Condie et al. 1988), and possibly the developing fetus (Bio/dynamics 1983; Mirkova et al. 1979, 1983; Taskinen et al. 1989) as the primary targets of intermediate-duration xylene exposure. Very few studies were located that examined the effects associated with intermediate-duration dermal exposure to xylenes (Mirkova et al. 1979; Wolf et al. 1956). Pharmacokinetic data indicate that absorption of xylenes occurs through the skin; however, it is difficult to determine whether similar end points would be expected after repeated dermal exposure to xylenes. Human skin may be repeatedly exposed to xylene as a result of occupational and home use. Repeated exposure of the skin to contaminated media at hazardous waste sites may also occur. Therefore, a well-designed and well-conducted intermediate-duration dermal study may help to better characterize the potential health hazards associated with repeated dermal exposure to xylenes.

An intermediate-duration inhalation MRL was derived based on a NOAEL for decreased rotarod performance in rats discontinuously exposed to *m*-xylene for 3 months (Korsak et al. 1994). Data were

sufficient to determine an intermediate-duration oral MRL for mixed xylenes based on a LOAEL for hyperactivity in animals (Condie et al. 1988). Deficiencies of the intermediate-duration oral toxicity database include the lack of testing for neurobehavioral effects, which appear to be the most sensitive end points in acute-duration oral toxicity studies, and lack of developmental and multi-generational data. Additional intermediate-duration oral studies that identify a NOAEL and LOAEL for sensitive neurobehavioral effects are needed to provide reliable guidance values for intermediate-duration exposure.

Chronic-Duration Exposure and Cancer. Few human (Arp et al. 1983; Askergren 1981, 1982; Askergren et al. 1981b, 1981c; Brasington and Thorpe-Swenson 1991; Dolara et al. 1982; Franchini et al. 1983; Gupta et al. 1990; Hipolito 1980; Holmberg and Nurminen 1980; Kilburn et al. 1985; Kucera 1968; Kurppa and Husman 1982; Moszczynski and Lisiewicz 1983, 1984a; Roberts et al. 1988; Smolik et al. 1973; Triebig et al. 1992a, 1992b; Uchida et al. 1993; Wilcosky et al. 1984) or animal studies (Tatrai et al. 1981; Maltoni et al. 1983, 1985; NTP 1986) were available regarding the health effects associated with chronic exposure to xylenes. The central nervous system (Gupta et al. 1990; Hipolito 1980; NTP 1986; Roberts et al. 1988) and the kidney (Askergren 1981, 1982; Askergren et al. 1981b, 1981c; Franchini et al. 1983) appear to be the primary targets of chronic xylene exposure. However, the study by Uchida et al. (1993) suggests that in healthy individuals, kidney effects are unlikely to occur at concentrations below those that cause neurological effects and eye and respiratory tract irritation. A chronic-duration inhalation MRL was derived based on the subjective neurological and respiratory effects noted in the Uchida et al. (1993) study. It is not clear if the effects noted in this study were a result of exposure at the TWA (14 ppm) or a result of short-term exposure at higher concentrations. Studies that focus on neurological effects with different exposure scenarios resulting in the same TWA may help to distinguish between effects caused by transient exposure to higher concentrations and those caused by stable low-level exposure. A chronic-duration oral MRL was calculated based on a NOAEL for decreased body weight at a dose that reduced survival in rats exposed to mixed xylenes for 2 years (NTP 1986). No specific target organ was identified in this study and there is uncertainty as to the NOAEL because there is no information on sensitive neurobehavioral effects. No chronic dermal studies of xylenes were identified. Since the inhalation and oral routes of exposure are the most important for individuals living near hazardous waste sites or in occupational settings, additional inhalation and oral studies are needed to provide critical, reliable guidance values for chronic exposure to xylenes.

Few epidemiological studies were available regarding the development of cancer in humans following inhalation, oral, or dermal exposure to mixed xylene or xylene isomers (Arp et al. 1983; Wilcosky et al.

1984). Several oral carcinogenicity bioassays involving lifetime exposure have been conducted with mixed xylene in rats and mice (Maltoni et al. 1983, 1985; NTP 1986); however, all of these bioassays contained limitations that preclude a definitive conclusion regarding the carcinogenicity of xylene. Several dermal studies are available in which xylene (unspecified isomeric content) was evaluated for its ability to enhance tumor induction by tumor-initiating and tumor-promoting agents (Berenblum 1941; Pound 1970; Pound and Withers 1963); however, these studies are less than lifetime and have often involved exposures to more than one chemical agent. No animal cancer bioassays involving inhalation exposure to mixed xylene or isomers of xylene have been conducted. Because the issue of the potential carcinogenicity of xylenes has not been resolved, additional bioassays are needed. Chronic inhalation exposure to low levels is needed in these studies because chronic exposure by this route may be encountered in the workplace, home, or in the vicinity of hazardous waste sites.

Genotoxicity. Limited data are available regarding the genotoxicity of inhalation of xylenes in humans (Haglund et al. 1980; Pap and Varga 1987; Richer et al. 1993). No data are available regarding the potential genotoxicity of xylenes in humans following oral or dermal exposure. Animal studies examining the genotoxicity of inhalation (Zhong et al. 1980) or oral (Feldt 1986) exposure to xylenes have been almost uniformly negative. The only positive assay was for DNA fragmentation in skin of rats exposed dermally (Rogers et al. 2001); this effect appears to be related to cell death. Also, a variety of *in vitro* assays (Anderson et al. 1990; Bos et al. 1981; Connor et al. 1985; DeMarini et al. 1991; Epler et al. 1979; Florin et al. 1980; Gerner-Smidt and Friedrich 1978; Haworth et al. 1983; Hejtmankova et al. 1979; Litton Bionetics 1978b; McCarroll et al. 1981a, 1981b; NTP 1986; Pool and Lin 1982; Richer et al. 1993; Shimizu et al. 1985) produced negative results. Because of the large number of negative studies that exist, additional *in vivo* or *in vitro* assays of the genotoxicity potential of xylenes are not needed.

Reproductive Toxicity. One epidemiological study suggested that paternal exposure to xylenes in the workplace may increase the likelihood of abortions; however, this study was limited by the size of the sample population (Taskinen et al. 1989). Only one animal inhalation study has been conducted to test the potential reproductive toxicity of mixed xylene (Bio/dynamics 1983). No studies of reproductive function have been conducted on either mixed xylene or the individual xylene isomers in animals following exposure via oral or dermal routes. Histopathological examination of reproductive organs of rats and mice following intermediate (NTP 1986; Wolfe 1988a, 1988b) and chronic (NTP 1986) oral bioassays revealed no adverse effects; however, given the high potential for human exposure to xylene and its isomers and their ability to cross the placenta (Ghantous and Danielsson 1986; Ungvary et al. 1980b), additional studies in animals and epidemiological studies in humans are needed to assess more

fully the reproductive toxicity of xylene and its isomers. A multi-generation reproductive toxicity study in animals is needed, possibly combined with a study examining developmental neurotoxicity end points.

Developmental Toxicity. Congenital defects of the central nervous system in children whose mothers were exposed occupationally to mixed xylene vapors were reported in two case studies (Holmberg and Nurminen 1980; Kucera 1968). However, the studies have many limitations, and no conclusion can be made. Animal inhalation, oral, and dermal studies have provided some information on the developmental effects of xylene and its isomers (Balogh et al. 1982; Bio/dynamics 1983; Hudak and Ungvary 1978; Litton Bionetics 1978a; Marks et al. 1982; Mirkova et al. 1979, 1983; Rosen et al. 1986; Seidenberg et al. 1986; Ungvary 1985; Ungvary and Tatrai 1985; Ungvary et al. 1980b, 1981); however, the quality of many of these studies precludes drawing conclusions. An inhalation developmental study in rats evaluated all three isomers and mixed xylenes using standard contemporary protocols, and found all tested xylenes could produce developmental toxicity (Saillenfait et al. 2003). Ingestion of aspirin by pregnant rats exposed to xylene have been shown to potentiate adverse maternal and fetal effects (Ungvary 1985). The most critical developmental studies in animals relate to neurobehavioral deficits observed in offspring following inhalation exposure of dams to xylenes, since these effects persist for months (Hass and Jakobsen 1993, 1995; Hass et al. 1997). No developmental neurobehavioral studies of xylenes have been conducted in animals exposed orally. Such studies are needed to better assess the potential of developmental toxicity and dose-response relationships associated with oral exposure to xylene. More information is needed on the mechanism of xylene-induced developmental toxicity, particularly of the developing nervous system, because this appears to be a sensitive target of xylene exposure. Such a study could be combined with a multi-generational reproductive toxicity study.

Immunotoxicity. Several occupational studies have been conducted to evaluate the immunological effects of xylene (Moszczynski and Lisiewicz 1983, 1984a; Smolik et al. 1973); however, workers in these studies were exposed to other chemical agents in addition to xylene. No animal studies involving exposure by any route have been conducted to examine directly the immunotoxicity of mixed xylene or the xylene isomers, although a decrease in thymus weight was observed in one oral study (Condie et al. 1988). Inhalation exposure studies in animals employing only xylene or its isomers may remove uncertainties about the immunotoxicity potential of xylene. One case report indicates that dermal sensitization to xylene is possible (Palmer and Rycroft 1993). Dermal sensitization tests would provide additional information on whether an allergic response to xylene is likely, since the potential for skin contact by humans occurs in occupational settings and in soil and water at hazardous waste sites.

Neurotoxicity. Human and animal studies regarding neurologic effects have been conducted following inhalation, oral, and dermal exposures to xylene (Andersson et al. 1981; Carpenter et al. 1975a; Condie et al. 1988; Dyer et al. 1988; Ernstgard et al. 2002; Gagnaire and Langlais 2005; Gagnaire et al. 2001, 2006; Gamberale et al. 1978; Gralewicz and Wiaderna 2001; Gralewicz et al. 1995; Klaucke et al. 1982; Korsak et al. 1990, 1991, 1992, 1993, 1994; Morley et al. 1970; NIOSH 1981; NTP 1986; Nylen and Hagman 1994; Ogata et al. 1970; Savolainen et al. 1984, 1985a; Wolfe 1988a, 1988b) (see Sections 3.2.1.4 and 3.2.2.4 for additional data). Data from such studies indicate that xylene adversely affects the nervous system; all inhalation MRLs and the acute oral MRL are based, at least in part, on neurological effects of xylene. The majority of studies in humans and animals concentrated on the neurobehavioral effects of xylene. Further studies attempting to elucidate the mechanism of action of xylenes on the nervous system are needed to increase understanding of the neurotoxic effects produced by high concentrations of xylenes. An occupational study of workers exposed to low concentrations of mixed solvents including xylenes for 10–44 years found no significant effects on CAT-scan measures of brain atrophy (Triebig et al. 1992a), but specific information on xylene is not available. Additional well-conducted studies in animals on the histopathologic changes of the central nervous system following intermediate or chronic exposure are needed to provide useful information on permanent structural alterations induced by xylene. More epidemiological and/or animal data are needed to better assess sensitive neurological end points (subjective effects, behavior, performance) for intermediate and chronic exposures by both inhalation and oral routes. The priorities in descending order of need are chronic oral > intermediate oral = chronic inhalation > intermediate inhalation.

Epidemiological and Human Dosimetry Studies. Limited epidemiological studies (Arp et al. 1983; Askergren 1981, 1982; Askergren et al. 1981b, 1981c; Dolara et al. 1982; Franchini et al. 1983; Gupta et al. 1990; Holmberg and Nurminen 1980; Kilburn et al. 1985; Kucera 1968; Kurppa and Husman 1982; Moszczynski and Lisiewicz 1983, 1984a; Smolik et al. 1973; Taskinen et al. 1989; Uchida et al. 1993; Wilcosky et al. 1984) and no human dosimetry studies on any of the xylenes have been conducted. Much of the available information on the effects of xylene in humans comes from case reports (Abu Al Ragheb et al. 1986; Arthur and Curnock 1982; Brasington and Thorpe-Swenson 1991; Goldie 1960; Hipolito 1980; Klaucke et al. 1982; Martinez et al. 1989; Morley et al. 1970; Nersesian et al. 1985; Roberts et al. 1988) and occupational studies in which subjects were exposed to other chemical agents in addition to xylene (Arp et al. 1983; Askergren 1981, 1982; Askergren et al. 1981b, 1981c; Dolara et al. 1982; Franchini et al. 1983; Gupta et al. 1990; Holmberg and Nurminen 1980; Jovanovic et al. 2004; Kilburn et al. 1985; Kucera 1968; Kurppa and Husman 1982; Moszczynski and Lisiewicz 1983, 1984a; Smolik et al. 1973; Taskinen et al. 1989; Uchida et al. 1993; Wilcosky et al. 1984). The best

characterization of exposure, verified with urinary metabolite measurements, was conducted in the occupational study by Uchida et al. (1993); subjects were exposed to 70% xylene with little involvement of other chemicals, besides ethylbenzene, a component of mixed xylene. Many of the case reports and occupational studies were also limited because exposure conditions were not well characterized. Additional well-designed and well-controlled epidemiological studies of people living near waste sites or industries using xylene, or occupational studies in which xylene exposure conditions are better characterized, may be useful to better describe potential neurotoxic effects and their dose-response relationships. Epidemiological studies examining the nervous system, reproductive outcome, and renal effects associated with xylene exposure would be particularly useful since these have been shown to be sensitive end points.

Human Sensory Studies on Oral Exposure. A sensory study is needed using human subjects on the detection of taste and smell thresholds for drinking water containing mixed xylenes. This data need is prompted by the observation that the guidance values for xylene (0.6–1 mg/kg/day) translated to typical daily water consumption (2 L/day) are only slightly below the maximum solubility of xylene in water (~145 mg/L). Information on the detectability of xylene at maximum solubility would be a useful rule of thumb for public health communications. This information might allow individuals to rule out water contamination at levels of concern in many situations.

Biomarkers of Exposure and Effect.

Exposure. Methods are available for determining xylene and its metabolite, methylhippuric acid, in biological tissues and fluids (Daniell et al. 1992; Jonai and Sato 1988; Kawai et al. 1991; Sedivec and Flek 1976b). These biomarkers of exposure are specific for xylene exposure and are sufficient for determining recent exposure to xylenes but are incapable of distinguishing short-term from intermediate- and chronic-duration exposures. It would be useful to determine if a biomarker of longer-term exposure could be derived, although it is not known whether one could be found.

Effect. No specific biomarkers of effects have been identified for xylenes. Xylenes have been demonstrated to cause a number of adverse health effects including central nervous system depression (Gamberale et al. 1978; Riihimaki and Savolainen 1980; Savolainen and Linnavuo 1979; Savolainen and Riihimaki 1981b; Savolainen et al. 1979b, 1984, 1985a). A number of neurological and cognitive function tests exist and have been used to identify central nervous system changes produced by xylenes. However, until the mechanism for nervous system disruption is identified, it is unlikely that a specific test

could predict xylene-specific intoxication. Similarly, xylene treatment of the skin elicits increases in pro-inflammatory cytokines in blood (interleukin 1-alpha) and skin (tumor necrosis factor-alpha) (Ahaghotu et al. 2005; Chatterjee et al. 2005; Gunasekar et al. 2003), but these responses are not specific to xylene exposure. Assessment of hepatic enzyme induction is difficult without obtaining liver tissue. Demonstration of enhanced metabolism of substances by the microsomal enzyme system could be interpreted as microsomal induction; however, a large number of substances other than xylenes also induce enhanced enzyme activity. Renal impairment also has been associated with high levels of xylene exposure. Increased excretion of albumin, leukocytes, and erythrocytes demonstrates kidney damage of the type ascribed to xylene exposure, but these effects are not specific for xylenes. However, limited data are available associating levels of xylene in human tissues and fluids with adverse health effects. Available human studies have focused on the blood concentrations of *m*-xylene associated with central nervous system effects. Additional animal studies evaluating the association between xylene (or xylene metabolite) levels in other human tissues or fluids and adverse health effects are needed.

Absorption, Distribution, Metabolism, and Excretion. The absorption, metabolism, and excretion of xylenes following inhalation, oral, and dermal exposures in humans and/or animals have been well characterized (Astrand 1982; Engstrom et al. 1977; Inoue et al. 1993; Jonai and Sato 1988; Kawai et al. 1991; Kezic et al. 2001; Ogata et al. 1970, 1979; Riihimaki 1979b; Riihimaki and Pfaffli 1978; Riihimaki et al. 1979a, 1979b; Skowronski et al. 1990). The distribution of xylene has been well characterized in animals and identified to a small extent in humans. The database for absorption, distribution, and excretion of xylene isomers in humans and/or animals after inhalation exposure is most extensive. The database for oral and dermal exposures is not as extensive but has been well described. Differences in the rate of metabolism of xylenes after short-term or chronic exposure have been identified. Differences in the toxicokinetics of xylene seen when exposure occurs with xylene adsorbed to sandy or clay soil have also been examined. Dermal penetration and resulting doses of xylene could be better characterized.

Although a number of route-specific PBPK models are available, a model is needed that would enable route-to-route extrapolation from intermediate- or chronic-duration inhalation data for oral exposure. Such a model would help address uncertainties in existing oral exposure data resulting from experimental bolus delivery to animals. Since environmental oral exposures involve consumption of contaminated drinking water during the course of a day, toxicity values based on bolus dosing would tend to overestimate risk.

Comparative Toxicokinetics. The target organs and adverse health effects of xylenes are similar across species. Toxicokinetic studies have been performed in humans, rats, mice, rabbits, and monkeys (Adams et al. 2005; Astrand et al. 1978; Bakke and Scheline 1970; Bray et al. 1949; Ogata et al. 1979; Patel et al. 1978; Smith et al. 1982; Sugihara and Ogata 1978; van Doorn et al. 1980). There is reasonable correlation between the end points examined in these studies. The metabolism of *m*- and *p*-xylenes is similar in rats and humans. However, a difference in the metabolism of *o*-xylene in rats and in humans exists. Whereas *o*-xylene is almost exclusively metabolized to *o*-methylhippuric acid in humans, 10–56% of *o*-xylene is also conjugated by glucuronide and glutathione in rats. Toxic metabolic intermediates of xylene such as benzaldehyde found in rats have not been found in humans. Additional studies are needed to determine whether other differences exist in the metabolism of xylenes among species. Although Inoue et al. (1993) did not observe a sex-related difference in excretion in men and women occupationally exposed to xylenes, sex-related differences in the toxicokinetics of xylene have been identified in animals. Additional studies concerning sex/genetic factors controlling xylene metabolism in humans might by useful.

Methods for Reducing Toxic Effects. Current methods used for reducing toxic effects of xylenes after acute exposures concentrate on decreasing absorption (HSDB 2007). Additional research on speeding excretion of xylene and reducing its concentration at its target organs are needed. As future research identifies the mechanisms underlying the toxic effects of xylenes, additional methods may be developed for combating the effects of xylene at the molecular level.

Children's Susceptibility. Data needs relating to both prenatal and childhood exposures, and developmental effects expressed either prenatally or during childhood, are discussed in detail in the Developmental Toxicity subsection above. There are no studies examining health effects in children exposed to xylenes, but the effects are likely to be the same as those observed in exposed adults. As mentioned in the Developmental Toxicity and Neurotoxicity subsections, there is a need for more data on neurological (neurobehavioral) effects of xylenes in laboratory animals. Studies in rats indicated that inhalation exposure to xylenes during gestation can affect neurobehavioral performance of offspring (Hass and Jakobson 1995; Hass et al. 1997). Studies in laboratory animals exposed by the oral route could identify dose-response relationships for neurological effects that would help in the assessment of health risks for children exposed to drinking water containing xylene.

There is currently insufficient age-specific information relevant to the absorption, distribution, metabolism, and elimination of xylenes in children. Although the absorption and distribution of xylenes

are driven by their lipophilic properties and diffusion, and the main metabolizing enzyme (CYP2E1) is available at adult levels in children by year 1 (Vieira et al. 1996), not enough information is available for children <1 year old. No data are available relative to the possible contribution of other microsomal enzymes to xylene metabolism in young animals. The elimination of xylene and its metabolites from the body is relatively rapid and expected to be similar in children and adults. However, the balance between factors tending to facilitate xylene clearance in children (lower body fat content, larger liver-to-body weight ratio) and factors tending to slow clearance (immaturity of enzyme systems) is not well understood. Data are especially needed to compare pulmonary metabolism of xylenes to hepatic metabolism, since the contribution of pulmonary metabolism may be significant for inhalation exposures (Adams et al. 2005). These suggestions are consistent with the recommendations of the peer consultation meeting of the Voluntary Children's Chemical Evaluation Program (VCCEP 2006). PBPK modeling studies indicate that xylenes can be excreted into breast milk (Fisher et al. 1997), but no PBPK models have been developed for children, fetuses, or pregnant women exposed to xylenes. Placental transfer of xylenes has been demonstrated in mice, but uptake by fetal tissues was lower than in maternal tissues (Ghantous and Danielsson 1986). Elimination of xylene and its metabolites is so rapid that there does not seem to be a need for pediatric-specific methods to reduce peak absorption.

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

3.12.3 Ongoing Studies

The Federal Research in Progress database (FEDRIP 2006) listed research by Dr. Karla Thrall on the assessment of human exposure to volatile organic chemicals, including xylene, at Superfund sites. The study, which is supported by the National Institute of Environmental Health Sciences, is intended to extend existing PBPK models to describe uptake kinetics and brain dosimetry following exposure. The results of this research program are intended to improve the extrapolation from animal studies to relevant human exposure situations and to develop biomarkers of susceptibility and response in potentially exposed populations. Dr. Steven Dewey is developing methods for quantifying exposure of fetal macaques after administration of positron-labeled inhalants such as ^{C11}toluene to the macaque mother. This research, sponsored by the National Institute on Drug Abuse, could be useful in the future for evaluating fetal and postnatal behavioral deficits resulting from maternal exposure to such inhalants.