APPENDIX A. ATSDR MINIMAL RISK LEVEL WORKSHEETS

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

MRLs are derived for hazardous substances using the NOAEL/uncertainty factor approach. They are below levels that might cause adverse health effects in the people most sensitive to such chemical-induced effects. MRLs are derived for acute (1–14 days), intermediate (15–364 days), and chronic (≥365 days) durations and for the oral and inhalation routes of exposure. Currently, MRLs for the dermal route of exposure are not derived because ATSDR has not yet identified a method suitable for this route of exposure. MRLs are generally based on the most sensitive substance-induced endpoint considered to be of relevance to humans. Serious health effects (such as irreparable damage to the liver or kidneys, or birth defects) are not used as a basis for establishing MRLs. Exposure to a level above the MRL does not mean that adverse health effects will occur.

MRLs are intended only to serve as a screening tool to help public health professionals decide where to look more closely. They may also be viewed as a mechanism to identify those hazardous waste sites that are not expected to cause adverse health effects. Most MRLs contain a degree of uncertainty because of the lack of precise toxicological information on the people who might be most sensitive (e.g., infants, elderly, nutritionally or immunologically compromised) to the effects of hazardous substances. ATSDR uses a conservative (i.e., protective) approach to address this uncertainty consistent with the public health principle of prevention. Although human data are preferred, MRLs often must be based on animal studies because relevant human studies are lacking. In the absence of evidence to the contrary, ATSDR assumes that humans are more sensitive to the effects of hazardous substances than animals and that certain persons may be particularly sensitive. Thus, the resulting MRL may be as much as 100-fold below levels that have been shown to be nontoxic in laboratory animals.

APPENDIX A

Proposed MRLs undergo a rigorous review process: Health Effects/MRL Workgroup reviews within the Office of Innovation and Analytics, Toxicology Section, expert panel peer reviews, and agency-wide MRL Workgroup reviews, with participation from other federal agencies and comments from the public. They are subject to change as new information becomes available concomitant with updating the toxicological profiles. Thus, MRLs in the most recent toxicological profiles supersede previously published MRLs. For additional information regarding MRLs, please contact the Office of Innovation and Analytics, Toxicology Section, Agency for Toxic Substances and Disease Registry, 1600 Clifton Road NE, Mailstop S106-5, Atlanta, Georgia 30329-4027.

APPENDIX A

MINIMAL RISK LEVEL (MRL) WORKSHEET

Chemical name(s):	Nitrobenzene
CAS number(s):	98-95-3
Date:	January 2024
Profile Status:	Final
Route:	Inhalation
Duration:	Acute
MRL	0.1 ppm (0.5 mg/m^3)
Critical Effect:	Increased methemoglobin
Reference:	Medinsky and Irons 1985
Point of Departure:	$BMCL_{1SD}$ = 16.3 ppm ($BMCL_{HEC}$ = 2.91 ppm)
Uncertainty Factor:	30
LSE Graph Key:	2
Species:	Rat

MRL Summary: An acute-duration inhalation MRL of 0.1 ppm was derived for nitrobenzene based on a LOAEL of 9.1 ppm ($BMCL_{1SD}$ of 16.3 ppm) for increased methemoglobin in a 14-day study in female CD rats (Medinsky and Irons 1985). The $BMCL_{1SD}$ of 16.3 ppm was adjusted to continuous-duration exposure and converted to a human equivalent concentration ($BMCL_{HEC}$) of 2.91 ppm. The $BMCL_{HEC}$ was divided by a total uncertainty factor of 30 (10 for human variability and 3 for animal to human extrapolation after dosimetric adjustment).

Selection of the Critical Effect: The database of acute-duration inhalation toxicity studies of nitrobenzene consists of two developmental toxicity studies in rabbits (Biodynamics 1983, 1984), a developmental toxicity study in rats (Tyl et al. 1987), and 14-day studies in CD and F344 rats and B6C3F1 mice that included comprehensive toxicological evaluations (Medinsky and Irons 1985). Table A-1 shows the lowest effect levels from the acute-duration studies. The studies by Tyl et al. (1987) and Biodynamics (1983, 1984) identified developmental effects in rats and rabbits at exposure concentrations of 39.4 and 41 ppm, respectively. Maternal effects seen in the rat dams included decreased maternal weight gain at 39.4 ppm and increased spleen weight at 9.8 ppm. Increased methemoglobin levels were observed in maternal rabbits exposed to 41 ppm in the developmental toxicity study (Biodynamics 1984). In the 14-day studies (Medinsky and Irons 1985), hematological effects occurred at all exposure levels in all species; the LOAEL was 9.1 ppm. The hematologic effects seen in rats and mice at the lowest exposure level consisted of increased spleen weight (F344 and CD rats), decreased erythrocyte count (CD rats), and splenic lesions (all animals). In F344 rats, increased liver and kidney weight were also seen at the LOAEL of 9.1 ppm and mild hepatocyte necrosis was observed in CD rats at 35.8 ppm. The data presented in Table A-1 indicate that hematological effects occur at the lowest exposure concentrations and appear to represent a critical effect of nitrobenzene.

Table A-1. Summary of Acute-Duration Inhalation NOAEL and LOAEL Values for Nitrobenzene

Species	Duration	NOAEL (ppm)	LOAEL (ppm)	$NOAEL_{ADJ}$[a] (ppm)	$LOAEL_{ADJ}$[a] (ppm)	Effect	Reference
Hematological effects							
F344 rat	14 days, 5 days/week, 6 hours/day		9.1		1.6	Splenic lesions (not specified); increased relative spleen weight (33%)	Medinsky and Irons (1985)
CD rat	14 days, 5 days/week, 6 hours/day		9.1		1.6	Decreased RBC count; increased relative spleen weight (44% in females); splenic lesions (not specified); increased methemoglobin in females (6.3 versus 4.8% in controls)	Medinsky and Irons (1985)
B6C3F1 mouse	14 days, 5 days/week, 6 hours/day		9.1		1.6	Splenic lesions (not specified)	Medinsky and Irons (1985)
CD rat	GDs 6–15, 6 hours/day	1.06	9.8	0.3	2.5	Increased absolute and relative maternal spleen weights (13–15%)	Tyl et al. (1987)
New Zealand white rabbit	GDs 7–19, 6 hours/day	9.9	41	2.5	10	Increased maternal methemoglobin (1.4 versus 1.0% in controls)	Biodynamics (1984)
Renal effects							
F344 rat	14 days, 5 days/week, 6 hours/day		9.1 M		1.6 M	Increased relative kidney weight (15%) in males	Medinsky and Irons (1985)
Hepatic effects							
F344 rat	14 days, 5 days/week, 6 hours/day		9.1 M		1.6 M	Increased relative liver weight (13%) in males	Medinsky and Irons (1985)
CD rat	14 days, 5 days/week, 6 hours/day	9.1	35.8	2.3	8.7	Mild hepatocyte necrosis	Medinsky and Irons (1985)

Table A-1. Summary of Acute-Duration Inhalation NOAEL and LOAEL Values for Nitrobenzene

Species	Duration	NOAEL (ppm)	LOAEL (ppm)	$NOAEL_{ADJ}$[a] (ppm)	$LOAEL_{ADJ}$[a] (ppm)	Effect	Reference
Developmental effects							
CD rat	GDs 6–15, 6 hours/day	9.8	39.4	2.5	9.9	Increased incidences of litters with variations (hole in parietal bone and ecchymosis on trunk)	Tyl et al. (1987)
New Zealand white rabbit	GDs 7–19, 6 hours/day	9.9	41	2.5	10	Decreased mean number viable male fetuses	Biodynamics (1984)

[a]Acute-duration inhalation studies were adjusted from intermittent daily exposures to the equivalent of 24-hour continuous exposure. The duration adjusted values were calculated as:

$$Adjusted\ Daily\ Dose = Intermittent\ dose \times \frac{hours\ per\ day\ exposed}{24\ hours} \times \frac{days\ per\ week\ exposed}{7\ days}$$

F = female(s); GD = gestation day; LOAEL = lowest-observed-adverse-effect level; $LOAEL_{ADJ}$ = LOAEL adjusted to continuous exposure; M = male(s); NOAEL = no-observed-adverse-effect level; $NOAEL_{ADJ}$ = NOAEL adjusted to continuous exposure

***Selection of the Principal Study*:** Medinsky and Irons (1985) conducted comprehensive toxicological evaluations, while the gestational exposure studies were limited to developmental endpoints. In addition, the lowest LOAELs were identified for hematological, renal, and liver effects in rats and mice in the study by Medinsky and Irons (1985). Therefore, the study by Medinsky and Irons (1985) was selected as the principal study.

Summary of the Principal Study:

Medinsky MA, Irons RD. 1985. Sex, strain, and species differences in the response. In: Ricker D, ed. Toxicity of nitroaromatic compounds. New York: Hemisphere Publishing Corporation, 35-51.

Medinsky and Irons (1985) compared effects of nitrobenzene in F344 rats, CD rats, and B6C3F1 mice. Groups of 10 male and 10 female animals per species and strain were exposed whole body to nitrobenzene vapor at concentrations of 0 (control), 10, 35, or 125 ppm (nominal) or 0, 9.1 or 35.8 ppm (analytical) for 6 hours/day, 5 days/week for 14 days. Animals were observed for general health status prior to and at the end of each exposure. Half of the animals (5/sex/species/strain/concentration) were sacrificed 3 days after the last exposure and the other half were sacrificed 14 days after the end of exposure. At necropsy, animals were weighed, and blood was collected for hematologic (erythrocyte and leukocyte counts; hematocrit, hemoglobin concentration, and mean cell volume) and serum chemistry analysis. Selected organs were weighed (liver, spleen, kidney, testes, and brain) and the following tissues were examined for histopathology (specifically the adrenal glands, bone marrow, brain, sternum, colon, duodenum, testes with epididymis, heart, ileum, left kidney, liver, mesenteric lymph nodes, lungs, nose and turbinates, ovaries, pancreas, spleen, stomach, thyroid glands, thymus, trachea, urinary bladder, uterus, and gross lesions).

There were no deaths or clinical signs of toxicity in F344 rats. In contrast, CD rats and mice exposed to 124.5 ppm nitrobenzene exhibited premature mortality and morbidity. After the 4^{th} exposure day, five male and three female CD rats were found dead; the rest of the group showed hyperpnea and wheezing and were sacrificed at the end of the first week. Mice at this exposure level were prostrate and exhibited dyspnea and were sacrificed between exposure days 2 and 4. Histopathology evaluation showed perivascular hemorrhage in the cerebellar peduncle in both rats and mice sacrificed early. No deaths occurred at lower exposure levels, and no brain lesions were observed in F344 rats at any exposure level or in CD rats or mice exposed to lower concentrations.

The study authors reported methemoglobin levels and relative organ weights in rats (but not mice) at each of the two sacrifice times (3 and 14 days after end of exposure). Table A-2 shows methemoglobin levels and significant organ weight changes in the groups sacrificed 3 days after exposure ended. As Table A-2 shows, a significant increase in methemoglobin level was reported in female CD rats exposed to 124.5 ppm. These animals were sacrificed early, so it is not clear when the methemoglobin levels were measured. No other statistically significant alterations in methemoglobin level were observed in rats at either sacrifice time, although there were concentration-dependent increases in methemoglobin in F344 rats sacrificed 3 days after treatment ended. For mice, the study authors reported only that the methemoglobin level in the group exposed to 124.5 ppm and sacrificed early was between 13 and 31%.

Table A-2. Methemoglobin Levels (%)[a] in 14-Day Inhalation Study of Rats and Mice

Effect	0	9.1 ppm	35.8 ppm	124.5 ppm
F344 rats, male	0	1.9±0.7	6.6±0.2	11.7±1.2
F344 rats, female	3.6±2.2	4.8±0.8	6.6±0.8	13.4±2.1
CD rats, male	6.9±1.3	6.1±0.5	8.7±1.0	14.0±1.3[b]
CD rats, female	4.8±0.7	6.3±0.6	7.3±1.4	31.3±2.5*[b]

[a]Methemoglobin levels are reported as mean±standard error for five animals/group. Results marked with an asterisk (*) are significantly different from control ($p<0.05$) as reported by the study authors. Methemoglobin levels were not reported for mice.
[b]Five male and three female CD rats died after the 4th day of exposure and the rest were sacrificed at the end of week 1 (5 days of exposure). It is not clear when the methemoglobin levels were measured in these animals.

Source: Medinsky and Irons 1985

Other hematology changes were primarily described qualitatively. F344 rats showed dose-related decreases in erythrocyte counts and decreased hemoglobin and hematocrit at 124.5 ppm. CD rats at 124.5 ppm exhibited decreased erythrocyte counts (~40% lower than controls), and the study authors noted that "less marked suppression was observed in animals exposed to 35 and 10 ppm nitrobenzene sacrificed on day 3 after exposure." Hemoglobin and hematocrit values were decreased, and MCV increased only at the highest concentration in CD rats. In mice, the only hematology change was an increase in MCV at the highest concentration.

No information was reported in the publication on organ weights in mice. At sacrifice 3 days after exposure ended (see Table A-3), relative liver weights were increased at ≥9.1 ppm in male F344 rats (≥13% relative to controls) and at concentrations ≥35.8 ppm in female F344 rats (≥ 27%). Both male and female F344 rats showed increased relative kidney weights at ≥9.1 ppm (≥15% in males and ≥6% in females). In the groups sacrificed 2 weeks after exposure ended, liver and kidney weights did not differ from controls. No effects on liver or kidney weights were observed in CD rats at either sacrifice time.

Rats of both strains showed markedly increased relative spleen weights. At sacrifice 3 days after exposure ended (see Table A-3), relative spleen weights were significantly increased at ≥9.1 ppm in female CD rats (≥44%) and at ≥35.8 ppm in male CD rats (74% relative to controls), male F344 rats (≥89%), and female F344 rats (≥111%). In the groups sacrificed 2 weeks after exposure ended, spleen weights did not differ from controls in CD rats, while spleen weights remained elevated relative to controls at ≥35.8 ppm in female F344 rats and at 124.5 ppm in male F344 rats.

A marked (44% relative to controls) decrease in testes weight was observed in male F344 rats exposed to 124.5 ppm and sacrificed 3 days after the end of treatment (see Table A-3). A similar magnitude of decrease was seen in the group exposed to this concentration and sacrificed after 2 weeks of recovery. No effect on testes weight was reported in male CD rats exposed to concentrations up to 35.8 ppm at either sacrifice time.

Table A-3. Significant Organ Weight Changes[a] in 14-Day Inhalation Study of Rats and Mice (Groups Sacrificed 3 Days after Exposure Ended)

Effect	0	9.1 ppm	35.8 ppm	124.5 ppm
F344 rats, male				
Relative liver weight (% body weight)	3.78±0.21	4.28±0.15* (13%)	5.02±0.08* (33%)	5.52±0.30* (46%)
Relative kidney weight (% body weight)	0.34±0.02	0.39±0.02* (15%)	0.39±0.01* (15%)	0.42±0.02* (24%)
Relative spleen weight (% body weight)	0.18±0.03	0.22±0.02 (22%)	0.34±0.02* (89%)	0.58±0.05* (222%)
Relative testes weight (% body weight)	1.31±0.05	1.33±0.03 (2%)	1.41±0.08 (8%)	0.73±0.08* (-44%)
F344 rats, female				
Relative liver weight (% body weight)	3.32±0.29	3.42±0.22 (3%)	4.22±0.34* (27%)	4.88±0.25* (47%)
Relative kidney weight (% body weight)	0.35±0.05	0.37±0.02* (6%)	0.43±0.08* (23%)	0.43±0.03* (23%)
Relative spleen weight (% body weight)	0.18±0.04	0.24±0.01 (33%)	0.38± 0.05* (111%)	0.68±0.06* (278%)
CD rats, male				
Relative spleen weight (% body weight)	0.19±0.04	0.20±0.03 (5%)	0.33±0.05* (74%)	NR[b]
CD rats, female				
Relative spleen weight (% body weight)	0.18±0.04	0.26±0.02* (44%)	0.35±0.06* (94%)	NR[b]

[a]Organ weights are reported as mean±standard error (percent change from control) for five animals/group. Results marked with an asterisk (*) are significantly different from control ($p<0.05$) as reported by the study authors. Organ weights were not reported for mice.
[b]Five male and three female CD rats died after the 4th day of exposure and the rest were sacrificed at the end of week 1 (5 days of exposure); organ weights were not reported for these animals.

NR = not reported

Source: Medinsky and Irons 1985

Histopathology changes related to treatment were observed in the spleen, kidneys, liver, and testes of rats (both strains) and mice; and in the lungs of CD rats and B6C3F1 mice. The study authors reported the histopathology incidences for the 35.8 and 124.5 ppm groups combined across the two sacrifice times. Histopathology findings were not reported quantitatively for the control or 9.1 ppm groups, and only selected endpoints were reported for the 35.8 ppm group.

The study authors stated that "splenic lesions were evident in all animals and dose groups exposed to nitrobenzene." Table A-4 shows the incidences of splenic lesions in the 35.8 and 124.5 ppm groups; the specific nature and incidences of splenic lesions in the 9.1 ppm group were not reported.

Table A-4. Splenic Noncancer Histopathology Changes[a] in 14-Day Inhalation Study of Rats and Mice (Combined Across Groups Sacrificed 3 and 14 Days after Exposure Ended)

Effect	0	9.1 ppm[b]	35.8 ppm	124.5 ppm
F344 rats, male				
Spleen, hemosiderosis	NR	Lesions present, nature and incidence not specified	10/10	10/10
Spleen, sinusoidal congestion			10/10	10/10
Spleen, EMH			10/10	10/10
Spleen, capsular hyperplasia			5/10	7/10
F344 rats, female				
Spleen, hemosiderosis	NR	Lesions present, nature and incidence not specified	10/10	10/10
Spleen, congestion			10/10	10/10
Spleen, EMH			10/10	10/10
Spleen, capsular hyperplasia			1/10	0/10
CD rats, male				
Spleen, hemosiderosis	NR	Lesions present, nature and incidence not specified	10/10	9/10
Spleen, congestion			5/10	10/10
Spleen, EMH			9/10	10/10
Spleen, lymphoid hypoplasia			9/10	0/10
Spleen, stromal hyperplasia			4/10	0/10
CD rats, female				
Spleen, hemosiderosis	NR	Lesions present, nature and incidence not specified	10/10	9/10
Spleen, congestion			4/10	10/10
Spleen, EMH			10/10	10/10
B6C3F1 mice, male				
Spleen, hemosiderosis	NR	Lesions present, nature and incidence not specified	2/9	0/10
Spleen, congestion			0/9	10/10
Spleen, EMH			9/10	10/10
Spleen, lymphoid hypoplasia			5/9	0/10
Spleen, stromal hyperplasia			3/9	0/10

Table A-4. Splenic Noncancer Histopathology Changes[a] in 14-Day Inhalation Study of Rats and Mice (Combined Across Groups Sacrificed 3 and 14 Days after Exposure Ended)

Effect	0	9.1 ppm[b]	35.8 ppm	124.5 ppm
B6C3F1 mice, female				
Spleen, hemosiderosis	NR	Lesions present, nature and incidence not specified	5/10	0/10
Spleen, congestion			5/10	10/10
Spleen, EMH			10/10	10/10
Spleen, lymphoid hypoplasia			1/10	0/10
Spleen, stromal hyperplasia			5/10	0/10

[a]Histopathology findings are reported as number affected/number examined. Results marked with an asterisk (*) are significantly different from control ($p<0.05$) as reported by the study authors.
[b]The study authors reported that "splenic lesions were evident in all animals and dose groups exposed to nitrobenzene" but did not specify the nature or the incidence(s) of the lesions in the 9.1 ppm group.

EMH = extramedullary hematopoiesis; NR = not reported

Source: Medinsky and Irons 1985

Rats of both sexes and strains showed renal changes. At the highest concentration, hydropic degeneration of cortical tubular cells, focal hyalinosis, and basophilic degeneration of tubular epithelial cells were reported in CD rats, and severe hyaline nephrosis was reported in F344 rats. The study authors noted that "degenerative tubular epithelial changes occurred in a small number of mice," and that minimal to moderate multifocal degenerative changes were noted in renal tubular epithelium of male mice exposed to 35 ppm." Incidences of renal lesions in mice were not given.

Hepatic effects in CD rats and in mice included necrosis and hydropic degeneration, with most rats and most male mice (and a few female mice) affected at 124.5 ppm (both CD rats and mice exposed to this concentration were sacrificed early). The study authors also reported (qualitatively) that all male CD rats sacrificed 3 days after the end of exposure to 35 ppm showed mild individual hepatocyte necrosis, while mice of both sexes exposed to 35 ppm exhibited nonnecrotic hepatocyte degenerative changes.

In the testes, multinucleated giant cells were observed in both species and strains at 124.5 ppm. In addition, F344 and CD rats in this exposure group exhibited dyspermiogenesis, while F344 rats also showed interstitial edema and interstitial Sertoli cell hyperplasia. Most mice at the highest exposure level had testicular degeneration. The study authors noted also that 1 of 10 mice exposed to 35 ppm showed acute testicular degeneration.

Lung histopathology changes in CD rats were limited to the highest exposure group (124.5 ppm, at which animals died or were sacrificed early) and consisted of perivascular edema and vascular congestion. In mice, moderate bronchiolar hyperplasia was observed in most animals of both sexes at the highest exposure level, and mild bronchial hyperplasia was seen in mice sacrificed 3 days after exposure to 35 ppm (incidence not reported).

Selection of the Point of Departure for the MRL: As shown in Table A-1, the lowest LOAELs for acute inhalation exposure to nitrobenzene were identified for hematological, renal, and liver effects in rats and mice in the 14-day study by Medinsky and Irons (1985). The effects of nitrobenzene on the blood, spleen, liver, and kidneys are all believed to originate with the formation of methemoglobin from

nitrobenzene metabolites, as detailed in *Metabolic Mechanisms* in Section 3.1.3. Thus, the point of departure (POD) for the acute-duration inhalation MRL was based on benchmark dose (BMD) modeling of methemoglobin levels. Medinsky and Irons (1985) provided quantitative data on methemoglobin levels in rats, but not in mice (see Table A-2). The highest methemoglobin level was measured in female CD rats exposed to 124.5 ppm, despite the fact that these animals were exposed for only 5 days. Thus, the data for female CD rats were selected for BMD modeling.

BMD modeling was conducted to identify a POD using the data for changes in methemoglobin levels in female CD rats administered nitrobenzene via inhalation (Table A-2). The highest exposure group (124.5 ppm) was omitted from modeling due to the mortality and early sacrifice of these animals. BMD modeling of continuous data was conducted with the EPA's Benchmark Dose Software (BMDS) (version 3.2). For these data, the Exponential, Hill, Linear, Polynomial, and Power continuous models available within the software were fit employing a benchmark response (BMR) of 1 standard deviation (SD). An adequate fit was judged based on the $\chi 2$ goodness-of-fit p value ($p>0.1$), magnitude of the scaled residuals in the vicinity of the BMR, and visual inspection of the model fit. In addition to these three criteria for judging adequacy of model fit, a determination was made as to whether the variance across dose groups was constant. If a constant variance model was deemed appropriate based on the statistical test provided in BMDS (i.e., Test 2; p-value >0.1), the final BMD results were estimated from a constant variance model. If the test for homogeneity of variance was rejected (p-value <0.1), the model was run again while modeling the variance as a power function of the mean to account for this nonconstant variance. If this nonconstant variance model did not adequately fit the data (i.e., Test 3; p-value <0.1), the data set was considered unsuitable for BMD modeling. Among all models providing adequate fit, the lowest BMCL was selected if the BMCLs estimated from different models varied >3-fold; otherwise, the BMCL from the model with the lowest Akaike information criterion (AIC) was selected.

Results of the modeling are shown in Table A-5. For female CD rat data, there was a marginal difference in the response and variance among concentration levels (test 1 p-value=0.088). The constant variance model did provide an adequate fit to the data (Test 2; p-value >0.1). With the constant variance model applied, the Exponential 2, Exponential 3, Polynomial, Power, and Linear models provided adequate fit to the means; the Exponential 4, Exponential 5 and Hill models did not. Of the fit models, the BMCLs were sufficiently close (<3-fold). The 2-degree Polynomial and Power models converged on the Linear model and had the lowest AIC; therefore, the Linear model was selected. Predicted BMC_{1SD} and $BMCL_{1SD}$ values are 31 and 16 ppm, respectively. Figure A-1 shows the fit of the linear model to the data.

Table A-5. Results from Benchmark Dose (BMD) Analysis (Constant Variance) of Increased Methemoglobin Levels in Female CD Rats Following Inhalation Exposure to Nitrobenzene (Medinsky and Irons 1985)

					Scaled residuals[c]	
Model	BMC_{1SD}[a] (ppm)	$BMCL_{1SD}$[a] (ppm)	p-Value[b]	AIC	Dose below BMC	Dose above BMC
Exponential (model 2)[d]	33.17	19.57	0.40	68.89	0.64	-0.13
Exponential (model 3)[d]	33.18	19.57	0.40	68.89	0.64	-0.13
Exponential (model 4)[d]			NA	70.18	-1.2×10^{-6}	-6.1×10^{-7}
Exponential (model 5)[d]			65,535	72.18	-0.0004	0.0006
Hill[d]			<0.0001	72.18	-1.5×10^{-7}	1.0×10^{-7}
Polynomial (2-degree)[d]	31.39	16.27	0.43	68.79	0.61	-0.15

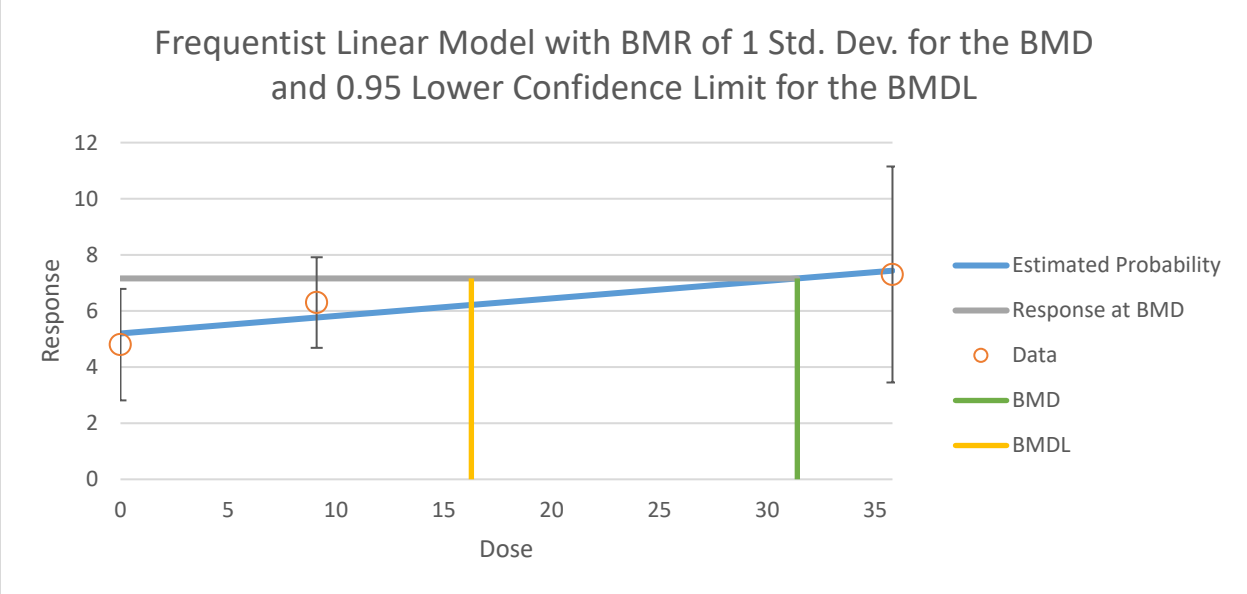
Table A-5. Results from Benchmark Dose (BMD) Analysis (Constant Variance) of Increased Methemoglobin Levels in Female CD Rats Following Inhalation Exposure to Nitrobenzene (Medinsky and Irons 1985)

Model	BMC_{1SD}[a] (ppm)	$BMCL_{1SD}$[a] (ppm)	p-Value[b]	AIC	Scaled residuals[c]	
					Dose below BMC	Dose above BMC
Power[d]	31.39	16.27	0.43	68.79	0.61	-0.15
Linear[e]	**31.39**	**16.27**	**0.43**	**68.79**	**0.61**	**-0.15**

[a]BMC and BMCL values for models that do not provide adequate fit are not included in this table.
[b]Values <0.1 fail to meet conventional χ^2 goodness-of-fit criteria.
[c]Scaled residuals at doses immediately below and above the BMC.
[d]Restricted model.
[e]Recommended model (lowest AIC). The constant variance model provided an adequate fit to the data (Test 2; p-value >0.1). With the constant variance model applied, the Exponential 2, Exponential 3, Polynomial, Power, and Linear models provided adequate fit to the means; the Exponential 4, Exponential 5, and Hill models did not. Of the fit models, the BMCLs were sufficiently close (<3-fold). The 2-degree Polynomial and Power models converged on the Linear model and had the lowest AIC; therefore, the Linear model was selected.

AIC = Akaike Information Criterion; BMC = maximum likelihood estimate of the exposure concentration associated with the selected benchmark response; BMCL = 95% lower confidence limit on the BMC (subscripts denote benchmark response: i.e., $_{1SD}$ = exposure dose associated with a change of 1 standard deviation from the control)

Figure A-1. Fit of the Linear Model (Constant Variance) to Data for Nitrobenzene, Methemoglobin Levels in Female CD Rats (Medinsky and Irons 1985)



Adjustment for Intermittent Exposure: The animals in the study by Medinsky and Irons (1985) were exposed 6 hours/day, 5 days/week. Therefore, the $BMCL_{1SD}$ of 16.3 ppm was adjusted for intermittent exposure as follows:

$$BMCL_{ADJ} = BMCL1SD \times \frac{6\ hours}{24\ hours} \times \frac{5\ days}{7\ days} = 16.3\ ppm \times \frac{6\ hours}{24\ hours} \times \frac{5\ days}{7\ days} = 2.91\ ppm$$

Human Equivalent Concentration: The hematological effects of nitrobenzene are systemic, so the $BMCL_{ADJ}$ was converted to a human equivalent concentration ($BMCL_{HEC}$) using guidance from EPA (1994) on dosimetric adjustments for extrarespiratory (systemic) effects. Blood:gas partition coefficients were not identified in the available literature for nitrobenzene. In the absence of a chemical-specific blood:gas partition coefficient, EPA (1994) recommends using a default value of 1. Therefore, the $BMCL_{HEC}$ was calculated by the following equation:

$$BMCL_{HEC} = BMCL_{ADJ} \times \frac{(HB/g)_A}{(HB/g)_H} = 2.91 * 1 = 2.91\ ppm$$

where:

$\frac{(HB/g)_A}{(HB/g)_H}$ = is the blood: air partition coefficient for animals (a) to humans (h)

Uncertainty Factor: The $BMCL_{HEC}$ is divided by a total uncertainty factor of 30:

- 10 for human variability
- 3 for animal to human extrapolation after dosimetric adjustment

MRL = $BMCL_{HEC}$ ÷ uncertainty factors
MRL = 2.91 ppm ÷ (3x10) = 0.097 ppm ≈ 0.1 ppm (rounded to one significant figure)

Agency Contacts (Chemical Managers): Malcolm Williams

APPENDIX A

MINIMAL RISK LEVEL (MRL) WORKSHEET

Chemical name(s):	Nitrobenzene
CAS number(s):	98-95-3
Date:	January 2024
Profile Status:	Final
Route:	Inhalation
Duration:	Intermediate
MRL	0.003 ppm (0.02 mg/m^3)
Critical Effect:	Hematological, renal, hepatic, and endocrine effects
Reference:	Hamm et al. 1984
Point of Departure:	LOAEL = 5 ppm ($LOAEL_{HEC}$ = 0.89 ppm)
Uncertainty Factor:	300
LSE Graph Key:	8
Species:	Rat

MRL Summary: An intermediate-duration inhalation MRL of 0.003 ppm was derived for nitrobenzene based on a LOAEL of 5 ppm for hematological, renal, hepatic, and endocrine effects (anemia, hemosiderin deposits, and histopathological changes in the kidneys, liver, and adrenal glands) in a 90-day study in rats and mice (Hamm et al. 1984). The LOAEL of 5 ppm was adjusted to continuous-duration exposure and converted to a human equivalent concentration ($LOAEL_{HEC}$) of 0.89 ppm. The $LOAEL_{HEC}$ was divided by a total uncertainty factor of 300 (10 for human variability, 10 for use of a LOAEL, and 3 for animal to human extrapolation after dosimetric adjustment).

Selection of the Critical Effect: There are two studies that evaluated toxicity in animals exposed by inhalation for intermediate durations (Dodd et al. 1987; Hamm et al. 1984). Table A-6 shows the lowest effect levels from these two studies. Dodd et al. (1987) was a 2-generation reproduction study with Sprague-Dawley rats in which a serious LOAEL of 40 ppm was identified for effects that included decreased fertility rates, atrophy of seminiferous tubules, spermatocyte degeneration, and reduced testicular and epididymal weights. The NOAEL was 10 ppm. Hamm et al. (1984) exposed F344 and Sprague-Dawley rats and B6C3F1 mice to nitrobenzene for 90 days and evaluated a comprehensive list of endpoints. Effects seen at the lowest exposure level (5 ppm) in the 90-day studies included hematological, renal, hepatic, and endocrine changes in rats and mice. All of these effects were considered for use in deriving the intermediate-duration inhalation MRL.

Selection of the Principal Study: The LOAEL of 5 ppm in the study by Hamm et al. (1984) was lower than the NOAEL of 10 ppm in the reproductive toxicity study by Dodd et al. (1987). Therefore, Hamm et al. (1984) was selected as the principal study for derivation of the intermediate-duration inhalation MRL.

Table A-6. Summary of Relevant NOAEL and LOAEL Values Considered for Derivation of an Intermediate-Duration Inhalation MRL for Nitrobenzene

Species	Duration	NOAEL (ppm)	LOAEL (ppm)	$NOAEL_{ADJ}$[a] (ppm)	$LOAEL_{ADJ}$[a] (ppm)	Effect	Reference
Hematological effects							
F344 rat	90 days, 5 days/week, 6 hours/day		5		0.89	Increased methemoglobin in males (3.0 versus 1.2% in controls); hematology changes indicative of hemolytic anemia in females; minimal to slight acute sinusoidal congestion and moderate to marked hemosiderin deposition in spleen in both sexes	Hamm et al. 1984
CD rat	90 days, 5 days/week, 6 hours/day		5		0.89	Splenic lesions: moderate hemosiderin pigmentation (males); slight to moderate sinusoidal congestion (both sexes)	Hamm et al. 1984
B6C3F1 mouse	90 days, 5 days/week, 6 hours/day		5		0.89	Splenic lesions: minimal to slight hemosiderin deposition (both sexes); slight to moderate sinusoidal congestion (females)	Hamm et al. 1984
Renal effects							
F344 rat	90 days, 5 days/week, 6 hours/day		5 M		0.89	Minimal nephrosis in males	Hamm et al. 1984
Hepatic effects							
CD rat	90 days, 5 days/week, 6 hours/day		5 M		0.89 M	Hepatic microgranulomas in males	Hamm et al. 1984
Endocrine effects							
B6C3F1 mouse	90 days, 5 days/week, 6 hours/day		5 F		0.89 F	Minimal to slight cortical cell vacuolization in zona reticularis of adrenal glands in females	Hamm et al. 1984

Table A-6. Summary of Relevant NOAEL and LOAEL Values Considered for Derivation of an Intermediate-Duration Inhalation MRL for Nitrobenzene

Species	Duration	NOAEL (ppm)	LOAEL (ppm)	$NOAEL_{ADJ}$[a] (ppm)	$LOAEL_{ADJ}$[a] (ppm)	Effect	Reference
Reproductive and developmental effects							
CD rat	10 weeks (2 generations), 5 days/week, 6 hours/day	10	40 (SLOAEL)	1.8	7.1	Decreased fertility rate, atrophy of seminiferous tubules, spermatocyte degeneration, and reduced testicular and epididymal weights; decreased F1 offspring body weights (12%) also occurred at this exposure level	Dodd et al. 1987

[a]Intermediate-duration inhalation studies were adjusted from intermittent daily exposures to the equivalent of 24-hour continuous exposure. The duration adjusted values were calculated as:

$$Adjusted\ Daily\ Dose = Intermittent\ dose \times \frac{hours\ per\ day\ exposed}{24\ hours} \times \frac{days\ per\ week\ exposed}{7\ days}$$

LOAEL = lowest-observed-adverse-effect level; $LOAEL_{ADJ}$ = LOAEL adjusted to continuous exposure; NOAEL = no-observed-adverse-effect level; $NOAEL_{ADJ}$ = NOAEL adjusted to continuous exposure

Summary of the Principal Study:

Hamm TE, Gibson JE, Irons RD, et al. 1984. Ninety-day inhalation toxicity study of nitrobenzene in F-344 rats, and CD rats and B6C3Fl mice with cover letter. Chemical Industry Institute of Technology. Submitted to the U.S. Environmental Protection Agency under section 8D. OTS0206507. 878214291.

Groups of male and female F-344 rats, CD rats, and B6C3F1 mice were exposed by inhalation (whole body) to nitrobenzene vapor at concentrations of 0 (control), 5, 16, or 50 ppm (nominal) or 0, 5, 15.8, or 48.7 ppm (analytical) for 6 hours/day, 5 days/week for 90 days. Experimental groups consisted of 10 male and 10 female animals of each strain and species. Animals were examined for clinical abnormalities twice daily and weighed once per week. Urine was collected for urinalysis. Animals were sacrificed at the end of the exposure period. Blood was drawn from the heart for hematology and serum chemistry analysis. Animals underwent gross necropsy and select tissues (spleen, brain, ovaries, kidney, testes, and brain) were weighed. Other tissues were prepared for histological analysis (cerebrum, pituitary gland, larynx, jejunum, pancreas, thymus, kidney, prostate, oviducts, lacrimal gland, sternum, nose/turbinates, thalamus, thyroid glands, esophagus, ileum, salivary gland, spleen, adrenal glands, testes, uterus, mammary glands, bone marrow, gross lesion, cerebellum, parathyroid, stomach, cecum, lymph nodes, heart, lungs, epididymis, urinary bladder, skeletal muscle, rib bone, medulla, trachea, duodenum, colon, liver, seminal vesicles, ovaries, eyes and optic nerve, peripheral nerve, and zymbal's gland).

There was no difference in survival between control groups and the nitrobenzene exposure groups. No biologically significant (≥10% less than controls) effects on body weight were observed. The only clinical sign of toxicity that was noted was diarrhea in female F344 rats at all exposure levels. Treatment-related hematology changes were observed in both rats and mice. Increased serum methemoglobin was observed at all exposure levels in male F344 rats; at ≥15.8 ppm in female F344 and male CD rats; and at 48.7 ppm in female CD rats and mice of both sexes. Hematology changes indicative of hemolytic anemia were seen in rats but not mice. Decreased erythrocyte counts, hematocrit, and/or hemoglobin, and increased erythrocyte width were evident at all exposure concentrations in female F344 rats, at ≥15.8 ppm in male and female CD rats, and at 48.7 ppm in male F344 rats. Clinical chemistry and urinalysis changes were not considered to be biologically significant, with the exception of a 2-fold increase in serum ALT in male mice.

Organ weight changes in male and female F344 rats included increased absolute and relative liver weights and increased absolute spleen weights at ≥15.8 ppm. Males also exhibited reduced testes weight at 48.7 ppm. Male and female CD rats had increased absolute and relative liver and spleen weights (≥15.8 ppm in females and at 48.7 ppm in males). Increased absolute and relative kidney weights and decreased absolute and relative testes weights were also noted in male CD rats exposed to 48.7 ppm. Spleen and liver weights were also increased in male and female mice exposed to 48.7 ppm. Male mice at that concentration showed increases in absolute and relative-to-brain-weight kidney weight, but not relative-to-body-weight kidney weight.

Enlarged spleen was observed at gross necropsy in animals of both species, strains, and sexes at the highest exposure level. Histopathology findings related to nitrobenzene exposure were observed in the respiratory tract, spleen, bone marrow, liver, and adrenal glands of both species, strains, and sexes. In addition, treatment-related increased incidences of lesions were observed in the kidneys and male reproductive organs of both strains of rat and in the thyroid glands of male CD rats.

Table A-7 summarizes the statistically significant treatment-related effects that occurred at all exposure levels in rats or mice. These included hematological, hepatic, renal, and endocrine effects.

Table A-7. Significant Noncancer Effects[a] Occurring at All Concentrations in a 90-Day Inhalation Study of Rats and Mice

Effect	0	5 ppm	15.8 ppm	48.7 ppm
F344 rats, male				
Methemoglobin (%)	1.2 ± 0.4	3.0 ±1.0*	4.4±1.3*	10.1±1.2*
Spleen, sinusoidal congestion	0/10	10/10* (2.0)	10/10* (1.8)	10/10* (2.8)
Spleen, hemosiderin deposition	10/10 (2.0)	10/10 (3.4)*	10/10 (3.7)*	10/10 (3.6)*
Kidney, nephrosis	0/10	6/10* (1.0)	9/10* (1.6)	10/10* (2.2)
F344 rats, female				
Erythrocyte count (10x6/mm^3)	9.07±0.18	8.7±0.18*	8.09±0.21*	7.36±0.23*
Hemoglobin (g/dL)	16.24±0.42	15.67±0.41*	14.81±0.28*	14.34±0.33*
Hematocrit (%)	48.58±1.68	47.12±1.04*	44.83±0.74*	42.25±1.06*
Spleen, sinusoidal congestion	0/10	10/10* (1.4)	10/10* (1.6)	10/10* (2.0)
Spleen, hemosiderin deposition	10/10 (2.5)	10/10 (3.8)*	10/10 (3.9)*	10/10 (4.4)*
CD rats, male				
Spleen, sinusoidal congestion	0/10	8/9* (1.6)	10/10* (2.4)	10/10* (2.4)
Spleen, hemosiderin deposition	10/10 (2.1)	9/9 (3.2)*	10/10 (4.9)*	10/10 (3.8)*
Liver, microgranulomas	0/10	7/9*	9/10*	7/10* (1.0)
CD rats, female				
Spleen, sinusoidal congestion	0/10	7/9* (1.6)	10/10* (2.6)	10/10* (3.9)
B6C3F1 mice, male				
Spleen, hemosiderin deposition	0/10	7/9* (1.0)	7/9* (2.3)	9/9* (2.6)
B6C3F1 mice, female				
Spleen, acute sinusoidal congestion	0/10	10/10* (2.9)	7/7* (4.0)	5/6* (3.8)
Spleen, hemosiderin deposition	1/7 (2.0)	10/10* (1.9)	7/7* (3/1)	6/6* (3.7)
Adrenal gland, cortical cell vacuolization	0/10	10/10* (1.8)	7/7* (3.4)	6/6* (4.3)

[a]Hematology changes are reported as mean±standard deviation. Histopathology findings are reported as number affected/number examined (mean severity score). Severity was scored as follows: 1=minimal; 2=slight; 3=moderate; 4=marked; and 5=very severe. Results marked with an asterisk (*) are significantly different from control ($p<0.05$) as reported by the study authors.

Source: Hamm et al. 1984

In F344 rats, additional treatment-related effects seen at the highest concentration (48.7 ppm) included: hyperplasia of the bronchial epithelium in males; proliferation of mesenchymal cells in the spleen, fibroblastic hyperplasia of the splenic capsule, accumulation of lymphocytes and macrophages in the spleen, and extramedullary hematopoiesis in the spleen; bone marrow hyperplasia; disorganized hepatic cords, vascular ectasia, centrilobular hepatocyte degeneration, periportal hepatocyte basophilia, and focal hepatocyte necrosis; nephrosis in females; increased basophilia of medullary cells in adrenal glands; and male reproductive tract changes consisting of moderate to severe degeneration of tubular epithelial cells, absence of mature sperm in epididymis, slight to moderate interstitial edema, and minimal to slight interstitial cell hyperplasia.

In CD rats, additional treatment-related effects seen at ≥15.8 ppm included centrilobular hepatocyte hypertrophy in females and toxic nephrosis in males and a slight reduction in mature sperm in two males. At the highest concentration (48.7 ppm), additional effects were seen in males including epithelial hyperplasia/metaplasia and goblet cell hyperplasia in the nasal turbinates; basophilia of adrenal medullary cells and thyroid follicular cell hypertrophy; and reproductive organ effects (bilateral testicular atrophy, complete loss of seminiferous epithelium, and absence of mature sperm in lumen of epididymis; increased interstitial cell hyperplasia, interstitial testicular atrophy, and multinucleate giant cells).

At the highest exposure level in B6C3F1 mice, treatment-related effects included hyperplasia of the bronchial mucosa; bone marrow hyperplasia; and centrilobular hepatocyte hyperplasia with some cord disorganization (females) and basophilic hepatocytes (males).

Selection of the Point of Departure for the MRL: As shown in Table A-7, effects seen at the lowest exposure level in the study by Hamm et al. (1984) included spleen congestion and/or hemosiderin deposition (significantly increased in severity but not incidence) in all species, strains, and sexes; increased methemoglobin and nephrosis in male F344 rats; hematology changes in female F344 rats; liver effects in male CD rats; and adrenal lesions in female B6C3F1 mice.

BMD modeling was conducted on the methemoglobin data for male F344 rats. In the absence of a biologically based benchmark for the methemoglobin level that is associated with adverse effects in rodents, a BMR of 1 SD was used. The data were fit to all continuous models in EPA's BMDS (version 3.2). Adequate model fit was judged by four criteria: chi-square goodness-of-fit p-value ($p \geq 0.1$), visual inspection of the dose-response curve, lower confidence limit on the benchmark dose (BMDL) <10 times the lowest non-zero dose, and scaled residual (>-2 and <+2) at the data point (except the control) closest to the predefined BMR. No adequate model fits were obtained.

The incidences of spleen congestion in nearly every species, strain, and sex (as well as adrenal cortical cell vacuolization in female mice) increased from 0 to ~100% at the lowest exposure level, precluding BMD modeling. Modeling was not considered for other histopathology changes (nephrosis, liver microgranulomas) that were seen at the LOAEL because the modeling results might not be adequately protective for the splenic and adrenal lesions.

In the absence of a suitable POD from modeling of the data, the LOAEL of 5 ppm was selected as the POD. Although the effects seen at the LOAEL were numerous and many occurred at high incidence, the lesions are likely all related to the effects of nitrobenzene on the blood, and severity scores indicated that the lesions were of generally minimal to moderate severity. Thus, the application of a 10-fold uncertainty factor for use of a LOAEL should provide adequate protection for the observed effects.

Adjustment for Intermittent Exposure: The animals in the study by Hamm et al. (1984) were exposed 6 hours/day, 5 days/week. Therefore, the LOAEL of 5 ppm was adjusted for intermittent exposure as follows:

$$LOAEL_{ADJ} = LOAEL \times \frac{6\ hours}{24\ hours} \times \frac{5\ days}{7\ days} = 5ppm \times \frac{6\ hours}{24\ hours} \times \frac{5\ days}{7\ days} = 0.89\ ppm$$

Human Equivalent Concentration: Given that all of the effects of nitrobenzene seen at the LOAEL were systemic, the $LOAEL_{ADJ}$ was converted to a human equivalent concentration ($LOAEL_{HEC}$) using guidance from EPA (1994) on dosimetric adjustments for extrarespiratory (systemic) effects. Blood:gas partition coefficients were not identified in the available literature for nitrobenzene. In the absence of a

chemical-specific blood:gas partition coefficient, EPA (1994) recommends using a default value of 1. Therefore, the $LOAEL_{HEC}$ was calculated by the following equation:

$$LOAEL_{HEC} = LOAEL_{ADJ} \times \frac{(HB/g)_A}{(HB/g)_H} = 0.89 * 1 = 0.89\ ppm$$

where:

$\frac{(HB/g)_A}{(HB/g)_H}$ = is the blood: air partition coefficient for animals (a) to humans (h)

Uncertainty Factor: The $LOAEL_{HEC}$ is divided by a total uncertainty factor of 300:

- 10 for human variability
- 10 for use of a LOAEL
- 3 for animal to human extrapolation after dosimetric adjustment

MRL = $LOAEL_{HEC}$ ÷ uncertainty factors
MRL = 0.89 ppm ÷ (3x10x10) = 0.003 ppm

Other Additional Studies or Pertinent Information that Lend Support to this MRL: In human case studies of both inhalation and ingestion exposure to nitrobenzene, methemoglobinemia was the most common finding (Agrawal et al. 2011; Gupta et al. 2012; Ikeda and Kita 1964; Lee et al. 2013; Perera et al. 2009; Saxena and Prakash Saxena 2010). Additionally, experimental animal studies have demonstrated increased methemoglobin levels in mice and rats of both sexes exposed through any exposure route with acute, intermediate, and chronic (≥365 days) exposure durations (Biodynamics 1984; Cattley et al. 1994, 1995; CIIT 1993; Hamm et al. 1984; Medinsky and Irons 1985; Mitsumori et al. 1994; NTP 1982, 1983b). A variety of related adverse effects in the hematologic system, such as hemolytic anemia; extramedullary hematopoiesis, hemosiderosis, congestion, and lymphoid depletion of the spleen; and changes in bone marrow in response to anemia have also been observed with inhalation, oral, and dermal exposures in B6C3F1 mice, F344 rats, and Sprague-Dawley rats (Burns et al. 1994; Cattley et al. 1994, 1995; CIIT 193; Hamm et al. 1984; Medinsky and Irons 1985; Mitsumori et al. 1994; NTP 1982, 1983a, 1983b).

Agency Contacts (Chemical Managers): Malcolm Williams

APPENDIX A

MINIMAL RISK LEVEL (MRL) WORKSHEET

Chemical name(s):	Nitrobenzene
CAS number(s):	98-95-3
Date:	January 2024
Profile Status:	Final
Route:	Inhalation
Duration:	Chronic
MRL	0.0002 ppm (0.001 mg/m^3)
Critical Effect:	Hyperplasia of the nasal squamous epithelium
Reference:	Cattley et al. 1994, 1995; CIIT 1993
Point of Departure:	LOAEL= 1 ppm ($LOAEL_{HEC}$= 0.054 ppm)
Uncertainty Factor:	300
LSE Graph Key:	12
Species:	Rat

MRL Summary: A chronic-duration inhalation MRL of 0.0002 ppm was derived for nitrobenzene based on hyperplasia of the squamous epithelium in male CD rats following exposure to nitrobenzene via inhalation for 2 years (Cattley et al. 1994, 1995; CIIT 1993). The MRL is based on a LOAEL of 1 ppm, which was adjusted to continuous-duration exposure and converted to a $LOAEL_{HEC}$ of 0.054 ppm. The $LOAEL_{HEC}$ was divided by a total uncertainty factor of 300 (10 for use of a LOAEL, 10 for human variability, and 3 for animal to human extrapolation after applying dosimetric adjustment).

Selection of the Critical Effect: The database of chronic-duration inhalation toxicity studies for nitrobenzene consists of 2-year experiments in male and female F344 rats, male CD rats, and male and female B6C3F1 mice reported in a single publication (Cattley et al. 1994, 1995). The unpublished report (CIIT 1993) provides additional details not reported in the publication. An overview of the lowest NOAEL and LOAEL values from this study are presented in Table A-8. The effects observed at the lowest exposure level in rats (1 ppm) and the lowest exposure level in mice (5 ppm) included effects on the respiratory tract (nasal lesions in rats and mice; pulmonary changes in mice) as well as systemic effects (hematological and hepatic effects). To provide a consistent basis for comparison across species, the LOAELs were adjusted for intermittent exposure and converted to HECs following EPA (1994) methodology. NOAELs were not identified for the most sensitive effects, so $NOAEL_{HECs}$ were not calculated. For systemic (extrarespiratory) effects, the HEC is calculated by multiplying the duration-adjusted animal NOAEL or LOAEL by the ratio of the blood:gas partition coefficients in animals and humans. Partition coefficients were not located for nitrobenzene, so the default value of 1 was used for the ratio. For effects on the respiratory tract, the regional gas dose ratio (RGDR) corresponding to the part of the respiratory tract that is affected was used. Thus, the RGDR for extrathoracic effects was used for nasal lesions and the RGDR for pulmonary effects was used for lung lesions. The $RGDR_{ET}$ (extrathoracic) values for mice (0.2) and rats (0.3) were calculated as follows:

$$RGDR_{ET} = (Mva/Saa) \div (MVh/Sah)$$

where:

Mva = minute volume for mice = 0.06 m^3/day; for rats = 0.47 m^3/day
MVh = minute volume for humans = 20 m^3/day
Saa = extrathoracic surface area for mice = 3 cm^2; for rats = 15 cm^2
Sah = extrathoracic surface area for humans = 200 cm^2

Table A-8. Summary of Relevant NOAEL and LOAEL Values Considered for Derivation of a Chronic Inhalation MRL for Nitrobenzene

Species	Duration	NOAEL (ppm)	LOAEL (ppm)	$NOAEL_{ADJ}$[a] (ppm)	$LOAEL_{ADJ}$[a] (ppm)	$LOAEL_{HEC}$ (ppm)	Effect	Reference
Respiratory effects								
CD rat (male)	2 years, 5 days/week, 6 hours/day		1		0.18	0.054	Increased nasal squamous epithelial hyperplasia/metaplasia and olfactory epithelial pigment deposition	Cattley et al. 1994, 1995; CIIT 1993
F344 rat	2 years, 5 days/week, 6 hours/day		1		0.18	0.054	Increased nasal olfactory epithelial pigment deposition (both sexes)	Cattley et al. 1994, 1995; CIIT 1993
B6C3F1 mouse	2 years, 5 days/week, 6 hours/day		5		0.89	0.18 (nasal) 2.89 (lung)	Increased nasal olfactory degeneration/necrosis in females; nasal olfactory epithelial pigment deposition (both sexes); bronchiolization of alveolar walls (both sexes)	Cattley et al. 1994, 1995; CIIT 1993
Hematologic effects								
CD rat (male)	2 years, 5 days/week, 6 hours/day		1		0.18	0.18	Increased methemoglobin at 15-month sacrifice (4.08 versus 1.18% in controls); spleen congestion at termination	Cattley et al. 1994, 1995; CIIT 1993
F344 rat	2 years, 5 days/week, 6 hours/day		1		0.18	0.18	Spleen congestion (both sexes); pigment deposition in spleen (males)	Cattley et al. 1994, 1995; CIIT 1993
B6C3F1 mouse	2 years, 5 days/week, 6 hours/day	5	24.8	0.89	4.43	4.43	Increased methemoglobin (in males (3.97 versus 1.97% in controls) and females (2.22 versus 1.39%)	Cattley et al. 1994, 1995; CIIT 1993

Table A-8. Summary of Relevant NOAEL and LOAEL Values Considered for Derivation of a Chronic Inhalation MRL for Nitrobenzene

Species	Duration	NOAEL (ppm)	LOAEL (ppm)	$NOAEL_{ADJ}$[a] (ppm)	$LOAEL_{ADJ}$[a] (ppm)	$LOAEL_{HEC}$ (ppm)	Effect	Reference
Hepatic effects								
CD rat (male)	2 years, 5 days/week, 6 hours/day		1		0.18	0.18	Kupffer cell pigmentation	Cattley et al. 1994, 1995; CIIT 1993
B6C3F1 mouse	2 years, 5 days/week, 6 hours/day		5		0.89	0.89	Increased centrilobular hepatocytomegaly and multinucleated hepatocytes in males	Cattley et al. 1994, 1995; CIIT 1993
Endocrine effects								
B6C3F1 mouse	2 years, 5 days/week, 6 hours/day		24.8		4.43	4.43	Thyroid follicular cell hyperplasia in males	Cattley et al. 1994, 1995; CIIT 1993
Renal effects								
B6C3F1 mouse	2 years, 5 days/week, 6 hours/day		49.1		8.77	8.77	Kidney cysts in males	Cattley et al. 1994, 1995; CIIT 1993

[a]Inhalation studies were adjusted from intermittent daily exposures to the equivalent of 24-hour continuous exposure. The duration adjusted values were calculated as:

$$Adjusted\ Daily\ Dose = Intermittent\ dose \times \frac{hours\ per\ day\ exposed}{24\ hours} \times \frac{days\ per\ week\ exposed}{7\ days}$$

LOAEL = lowest-observed-adverse-effect level; $LOAEL_{ADJ}$ = LOAEL adjusted to continuous exposure; $LOAEL_{ADJ(HEC)}$ = LOAEL adjusted to continuous exposure and converted to human equivalent concentration (HEC; see text); NOAEL = no-observed-adverse-effect level; $NOAEL_{ADJ}$ = NOAEL adjusted to continuous exposure

The $RGDR_{PU}$ (pulmonary) was calculated as follows:

$$RGDR_{PU} = (Mva/Saa) \div (MVh/Sah)$$

where:

Mva = minute volume for mice = 0.06 m^3/day
MVh = minute volume for humans = 20 m^3/day
Saa = pulmonary surface area for mice = 0.05 m^2
Sah = pulmonary surface area for humans = 54 m^2

As Table A-8 shows, the lowest $LOAEL_{HEC}$ values were 0.054 ppm for nasal epithelial hyperplasia and olfactory pigment deposition in rats and 0.18 ppm for nasal olfactory degeneration in mice, and increased methemoglobin (male CD rats), spleen congestion (F344 and CD rats), and Kupffer cell pigmentation in the liver (male CD rats).

***Selection of the Principal Study*:** Only one chronic inhalation study of nitrobenzene was located (Cattley et al. 1994, 1995; CIIT 1993). In this study, male and female B6C3F1 mice, male and female F344 rats and male CD rats were exposed by inhalation for 2 years and comprehensive toxicological endpoints were evaluated. This study was selected as the principal study for derivation of the chronic-duration inhalation MRL.

Summary of the Principal Study:

Cattley RC, Everitt JI, Gross EA, et al. 1994. Carcinogenicity and toxicity of inhaled nitrobenzene in B6C3F1 mice and F344 and CD rats. Fundam Appl Toxicol 22(3):328-340.

Cattley RC, Everitt JI, Gross EA, et al. 1995. Erratum: Carcinogenicity and toxicity of inhaled nitrobenzene in B6C3F1 mice and F344 and CD rats. Fundam Appl Toxicol 25:159. https://doi.org/10.1006/faat.1994.1039.

CIIT. 1993. Initial submission: A chronic inhalation toxicity study of nitrobenzene in B6CF1 mice, Fischer 344 rats and Sprague-Dawley (CD) rats. First Chemical Corporation. Submitted to the U.S. Environmental Protection Agency under TSCA Section 8E. OTS0538399. 88930000170. 8EHQ02938723. https://ntrl.ntis.gov/NTRL/dashboard/searchResults/titleDetail/OTS0538399.xhtml. November 9, 2022.

Cattley et al. (1994, 1995; CIIT 1993) evaluated the chronic toxicity and carcinogenicity of inhaled nitrobenzene in groups of 70 male and female F344 rats, male CD rats, and male and female B6C3F1 mice. Only selected results were included in the published report (Cattley et al. 1994, 1995); additional details were obtained from the unpublished version (CIIT 1993). All animals were exposed to nitrobenzene for 6 hours/day, 5 days/week for 2 years in an inhalation chamber. Rats were exposed to target concentrations of 0, 1, 5, or 25 ppm nitrobenzene, and mice were exposed to 0, 5, 25, or 50 ppm nitrobenzene (analytically measured concentrations for 0, 1, 5, 25, and 50 ppm exposures were 0, 1, 5, 24.8, and 49.1 ppm, respectively). Animals were examined twice daily for mortality and clinical abnormalities. Body weight was measured weekly for 13 weeks and biweekly thereafter. Groups of 10 rats of each strain and sex were sacrificed after 15 months of exposure; all other animals were sacrificed after the 2-year exposure period. At sacrifice, evaluations included hematology, clinical chemistry, organ weights (spleen, liver, kidney, and brain), gross necropsy, and histopathology (including nose, lung, liver, thyroid, parathyroid, spleen, adrenal glands, femur [bone marrow], sternum, and any tissues with gross lesions). There was no difference in survival between control groups and the

nitrobenzene exposure groups. No exposure-related clinical signs or biologically significant (≥10% less than controls) effects on body weight were observed in the animals. Treatment-related hematology changes were observed in both rats and mice. Male and female F344 rats exhibited decreased erythrocyte counts, hematocrit, and hemoglobin as well as increased methemoglobin at 24.8 ppm; these changes were evident at both the interim and final sacrifices in F344 rats. At the interim sacrifice, male CD rats exhibited increased methemoglobin at all exposure levels (≥1 ppm), while at termination the difference from control was significant only at 24.8 ppm. In male mice the changes consisted of decreased erythrocyte counts and hematocrit, and increased methemoglobin at 49.1 ppm. In female mice, methemoglobin was increased at ≥24.8 ppm.

Organ weight changes in both male and female F344 rats consisted of increased absolute and relative liver and kidney weights at 24.8 ppm. Male CD rats showed no significant changes in spleen, liver, kidney, or brain weights at any exposure level. In female mice, absolute and relative liver and kidney weights were increased at the highest exposure level (49.1 ppm); no biologically relevant organ weight changes were observed in male mice.

Treatment-related increases in the incidences of nonneoplastic lesions were observed in the nose, spleen, liver, kidneys, and pancreas of F344 rats; in the nose, liver, testes, and epididymides of CD rats; and in the nose, lung, bone marrow, liver, kidney, thyroid, pancreas, and thymus of B6C3F1 mice. Nasal lesions occurring at all exposure concentrations (≥1 ppm) in rats included olfactory epithelial pigment deposition (F344 and CD) and squamous epithelial hyperplasia (CD rats). At higher exposures, nasal findings in the rats included inflammation, sometimes with submucosal gland hypertrophy, and suppurative exudate. Mice exhibited more severe lesions in the nasal passages, including the following findings that were seen at significantly increased incidences at all exposure levels (≥5 ppm): glandularization of the respiratory epithelium, olfactory epithelial degeneration, increased secretory product in the respiratory epithelium, olfactory epithelial pigment deposition, and dilatation of the submucosal glands.

No lung effects of nitrobenzene were seen in rats. Male and female mice exhibited increased incidences of alveolar bronchiolization at all exposure levels (≥5 ppm), and males showed increased alveolar/ bronchiolar hyperplasia at ≥24.8 ppm.

Increased incidences of spleen congestion were reported in male and female F344 rats and male CD rats at all exposure levels (≥1 ppm), and increased spleen pigmentation was reported at all exposures in male F344 rats. Splenic lesions were not observed in male mice; females showed an increased incidence of lymphoid hyperplasia at 49.1 ppm. An increased incidence of bone marrow hypercellularity was reported for male mice exposed to 49.1 ppm.

The liver was a target organ for nitrobenzene in both rats and mice. In F344 rats, increased incidences of eosinophilic foci and centrilobular hepatocytomegaly were observed in males exposed to ≥5 ppm, and an increased incidence of spongiosis hepatis was seen at 24.8 ppm. Increased incidences of eosinophilic foci and spongiosis hepatis were noted in females exposed to 24.8 ppm. An increase in the incidence of pigmentation in the Kupffer cells occurred in male CD rats at all exposure levels (≥1 ppm); in addition, increased centrilobular hepatocytomegaly and increased spongiosis hepatis occurred at ≥5 and 24.8 ppm, respectively. In mice, increased incidences of centrilobular hepatocytomegaly and multinucleated hepatocytes were reported for males at all exposure levels (≥5); females exhibited an increased incidence of centrilobular hepatocytomegaly at the highest exposure (49.1 ppm).

Renal effects were seen in both rats and mice. In F344 rats, increases in renal tubular hyperplasia, cysts, (in males) and chronic nephropathy (females) were reported at 24.8 ppm, as well as renal tubular suppurative inflammation in both sexes. The only renal effect observed in male CD rats was

mineralization at 24.8 ppm. Male mice, but not female mice, showed a higher incidence of kidney cysts at 49.1 ppm.

Effects on male reproductive organs were noted at higher concentrations. Male CD rats had increased incidences of bilateral atrophy of the testes, bilateral hypospermia in the epididymis, and atrophy of the seminal vesicles at 24.8 ppm. Male mice exposed to nitrobenzene also exhibited hypospermia of the epididymis at 49.1 ppm.

In addition to the effects noted above, male F344 rats exhibited an increased incidence of focal pancreatic acinar cell hyperplasia at 24.8 ppm. Other effects reported to occur at increased incidence in male CD rats at 24.8 ppm included mineralization of the aorta, myocardium, stomach muscle, and kidney; and fibrous osteodystrophy in the nose and bone. In male mice, increased incidences of thyroid follicular cell hyperplasia were seen at ≥24.8 ppm. Other effects observed in female mice consisted of increased incidences of adrenal gland cortical cell vacuolization (≥24.8 ppm), as well as thymic involution and mononuclear cell infiltrate of the pancreas (at 49.1 ppm).

Table A-9 shows the incidences of treatment-related nonneoplastic histopathology changes that occurred at all exposure levels in rats or mice.

Table A-9. Significant Noncancer Effects[a] Occurring at All Concentrations in Chronic Inhalation Study of Rats and Mice

Effect	0	1 ppm	5 ppm	24.8 ppm	49.1 ppm
F344 rats, male					
Spleen, congestion	43/69 (62)	61/69 (88)*	62/78 (79)*	61/78 (78)*	
Spleen, pigmentation	55/69 (80)	63/69 (91)*	64/70 (91)*	70/70 (100)*	
Nose, olfactory epithelium pigment deposition	40/67(60)	53/67 (79)*	67/70 (96)*	68/69 (99)*	
F344 rats, female					
Spleen, congestion	42/69 (61)	55/66 (83)*	58/66 (88)*	65/69 (94)*	
Nose, olfactory epithelium pigment deposition	37/67 (55)	54/65 (83)*	60/65 (92)*	66/66 (100)*	
CD rats, male					
Methemoglobin (%), 15 months[b]	1.18±0.34	4.08±0.80*	6.22±1.6*	5.85±0.83*	
Spleen, congestion	25/63 (40)	43/67 (64)*	44/69 (64)*	38/65 (58)*	
Nose (L1), squamous epithelial hyperplasia	11/63 (17)	42/65 (65)*	34/66 (52)*	42/62 (68)*	
Liver, Kupffer cell pigmentation	0/63	6/67 (9)*	10/70 (14)*	42/65 (65)*	
B6C3F1 mice, male					
Lung, bronchiolization	0/68		58/67 (87)*	58/65 (89)*	62/66 (94)*
Liver, centrilobular hepatocytomegaly	1/68 (1)		15/65 (23)*	44/65 (68)*	57/64 (89)*
Liver, multinucleated hepatocytes	2/68 (3)		14/65 (22)*	45/65 (69)*	56/64 (88)*
Nose (L1), glandularization of respiratory epithelium	4/67 (6)		36/67(54)*	45/65 (69)*	50/66 (76)*
Nose (L3), olfactory epithelium pigment deposition	0/67		7/66 (11)*	46/65 (71)*	49/66 (74)*
Nose (L3), dilatation of submucosal glands	3/67 (4)		13/66 (20)*	39/65 (50)*	32/66 (48)*

Table A-9. Significant Noncancer Effects[a] Occurring at All Concentrations in Chronic Inhalation Study of Rats and Mice

Effect	0	1 ppm	5 ppm	24.8 ppm	49.1 ppm
	B6C3F1 mice, female				
Lung, bronchiolization	0/53		53/60 (92)*	63/64 (98)*	62/62 (100)*
Nose (L1) glandularization of respiratory epithelium	3/52 (6)		17/60 (28)*	28/63 (44)*	46/62 (74)*
Nose (L1), increased secretory product in respiratory epithelium	0/52		34/60 (57)*	50/63 (79)*	35/62 (56)*
Nose (L2), olfactory degeneration	0/52		9/60 (15)*	28/63 (44)*	32/62 (52)*
Nose (L3), olfactory degeneration	0/52		19/60 (32)*	47/63 (75)*	42/61 (69)*
Nose (L4), olfactory degeneration	0/52		30/59 (51)*	37/63 (59)*	48/61 (79)*
Nose (L3), olfactory epithelium pigment deposition	0/52		6/60 (10)*	37/63 (59)*	29/61 (48)*
Nose (L3), dilatation of submucosal glands	9/52 (17)		22/60 (37)*	40/63 (63)*	46/61 (75)*
Nose (L4), dilatation of submucosal glands	2/52 (4)		11/59 (19)*	20/63 (32)*	39/61 (64)*

[a]Histopathology findings are reported as number affected/number examined (percent incidence). Results marked with an asterisk (*) are significantly different from control ($p<0.05$) as reported by the study authors.
[b]Methemoglobin levels are reported as mean±standard error.

L1, L2, L3, L4 = segments of the nasal cavity; shaded = concentration not tested

Sources: Cattley et al. 1994, 1995; CIIT 1993

Exposure to nitrobenzene was associated with increased incidences of several tumor types. Male F344 rats exhibited increased incidences of hepatocellular adenomas or carcinomas and renal tubular adenomas or carcinomas at 24.8 ppm. An increased incidence of hepatocellular adenomas or carcinomas was also observed in male CD rats exposed to 24.8 ppm. In female F344 rats, the only treatment-related neoplastic change was an increased incidence of endometrial stromal polyps at 24.8 ppm. Male B6C3F1 mice exhibited significantly increased incidences of alveolar/bronchiolar adenomas or carcinomas at all exposure levels (≥5 ppm). Female mice showed a significant increase in the incidence of mammary gland adenocarcinomas at 49.1 ppm; mammary glands were not examined for histopathology at lower concentrations.

Selection of the Point of Departure for the MRL: After conversion to HECs, the lowest LOAEL values (see Table A-8) were for nasal lesions in rats (squamous epithelial hyperplasia in male CD rats and olfactory epithelium pigment deposition in F344 rats). The selection of this endpoint is supported by the fact that rats are generally more sensitive to the toxicity of nitrobenzene than mice are (see Section 3.1.6); thus, rats were tested at a lower exposure concentration (1 ppm) than mice (5 ppm) in the chronic study (Cattley et al. 1994, 1995; CIIT 1993). The nasal lesions in male CD rats included squamous epithelial hyperplasia and olfactory epithelium pigment deposition. The toxicological significance of the pigment deposition is uncertain. Pigment deposition observed in the livers and spleens of exposed rats could have resulted from hemolysis, an established effect of nitrobenzene; however, it seems unlikely that products of hemolysis would be distributed to or preferentially deposited in the nasal passages. Given the uncertainty in the relevance of this effect, pigment

deposition was not considered further for use in deriving the MRL. The data on squamous epithelial hyperplasia in male CD rats were subjected to BMD modeling.

The data were fit to all dichotomous models in EPA's BMDS (version 3.2) using a BMR of 10% relative deviation. Adequate model fit was judged by four criteria: chi-square goodness-of-fit p-value ($p \geq 0.1$), visual inspection of the dose-response curve, BMDL <10 times the lowest non-zero dose, and scaled residual (>-2 and <+2) at the data point (except the control) closest to the predefined BMR. Among models providing adequate fit to the data, the lowest $BMDL_{10}$ was selected as the POD when the difference between the BMDLs estimated from these models was >3 fold; otherwise, the $BMDL_{10}$ from the model with the lowest AIC was chosen. No model fit was achieved with the data.

In the absence of a suitable POD from modeling of the data, the LOAEL of 1 ppm for squamous epithelial hyperplasia in the nose of rats was selected as the POD.

Adjustment for Intermittent Exposure: The animals in the study by Cattley et al. (1994, 1995; CIIT 1993) were exposed 6 hours/day, 5 days/week. Therefore, the LOAEL of 1 ppm was adjusted for intermittent exposure as follows:

$$LOAEL_{ADJ} = LOAEL \times \frac{6\ hours}{24\ hours} \times \frac{5\ days}{7\ days} = 1\ ppm \times \frac{6\ hours}{24\ hours} \times \frac{5\ days}{7\ days} = 0.18\ ppm$$

Human Equivalent Concentration: The critical effect at the LOAEL was in the respiratory system (nasal epithelial hyperplasia). Therefore, the $LOAEL_{ADJ}$ was converted to a $LOAEL_{HEC}$ using guidance from EPA (1994) on dosimetric adjustments for respiratory effects. The $LOAEL_{ADJ}$ was converted to a $LOAEL_{HEC}$ using the RGDR for extrathoracic effects (EPA 1994) calculated as follows:

$$RGDR_{ET} = \frac{MV_a}{SA_a} \div \frac{MV_h}{SA_h}$$

where:

MV_a = minute volume for rats = 0.47 m^3 per day
SA_a = ET surface area for rats = 15 cm^2
MV_h = minute volume for humans = 20 m^3 per day
SA_h = ET surface area for humans = 200 cm^2

Applying this equation results in an RGDR of 0.3 for extrathoracic effects in rats, and the HEC is calculated as:

$$LOAEL_{HEC} = LOAEL_{ADJ} \times RGDR = 0.18\ ppm \times 0.3 = 0.054\ ppm$$

Uncertainty Factor: The $LOAEL_{HEC}$ was divided by a composite uncertainty factor of 300:

- 10 for use of a LOAEL
- 10 for human variability
- 3 for animal to human extrapolation after dosimetric adjustment.

This results in the following MRL:

$$MRL = \frac{LOAEL_{HEC}}{UFs} = \frac{0.054\ ppm}{300} = 0.0002\ ppm$$

Other Additional Studies or Pertinent Information that Lend Support to this MRL: Results from inhalation studies indicate effects of nitrobenzene exposure on the nasal passages of rats and mice and the lungs of mice. In chronic-duration studies, mice had degeneration of the nasal olfactory epithelium and glandularization of respiratory epithelium and rats had squamous epithelial hyperplasia, pigment deposition in the olfactory epithelium, and inflammatory changes (Cattley et al. 1994, 1995; CIIT 1993). In the same study, increases in bronchiolization of the alveoli and alveolar/bronchiolar hyperplasia were observed in mice (Cattley et al. 1994, 1995; CIIT 1993). Acute- and intermediate-duration dermal exposure studies have demonstrated lung congestion after nitrobenzene exposure in F344 rats (NTP 1982).

Agency Contacts (Chemical Managers): Malcolm Williams

MINIMAL RISK LEVEL (MRL) WORKSHEET

Chemical name(s): Nitrobenzene
CAS number(s): 98-95-3
Date: January 2024
Profile Status: Final
Route: Oral
Duration: Acute
MRL: 0.05 mg/kg/day
Critical Effect: Proliferative changes in the bone marrow
Reference: Burns et al. 1994
Point of Departure: $BMDL_{1SD}$ of 4.7 mg/kg/day
Uncertainty Factor: 100
LSE Graph Key: 10
Species: Mouse

MRL Summary: An acute-duration oral MRL of 0.05 mg/kg/day was derived for nitrobenzene based on a LOAEL of 30 mg/kg/day ($BMDL_{1SD}$ of 4.7 mg/kg/day) for increased DNA synthesis indicating proliferative changes in the bone marrow in a 14-day study in mice (Burns et al. 1994). The $BMDL_{1SD}$ was divided by a total uncertainty factor of 100 (10 for extrapolation from animals to humans and 10 for human variability).

Selection of the Critical Effect: There are nine studies that evaluated toxicity in animals administered oral doses for acute durations (Burns et al. 1994; Iida et al. 1997; Kawaguchi et al. 2004; Kawashima et al. 1995; Levin et al. 1988; Linder et al. 1992; McLaren et al. 1993a; Morgan et al. 1985). In eight of these studies, a single dose, usually 60 mg/kg, was used to evaluate the effect on the male reproductive systems. The only study testing multiple doses and evaluating other health outcomes was the 14-day gavage study in mice by Burns et al. (1994). Burns et al. (1994) administered nitrobenzene to female B6C3F1 mice via gavage in corn oil for 14 days and evaluated immune system endpoints in addition to several other systemic endpoints for acute toxicity. Table A-10 shows the lowest effect levels from the acute-duration oral studies of nitrobenzene.

Table A-10. Summary of Relevant NOAEL and LOAEL Values Considered for Derivation of an Acute Oral MRL for Nitrobenzene

Species	Duration	NOAEL (mg/kg/day)	LOAEL (mg/kg/day)	Effect	Reference
Hematological effects					
B6C3F1 mouse	Once/day for 14 days (GO)		30	Increased number of cells, DNA synthesis, and number of granulocyte-monocyte progenitor cells in bone marrow	Burns et al. 1994
Reproductive effects					
Sprague-Dawley rat	Once/day for 14 days (GO)		60	Decreased testes weight and epididymal weights, 34% decrease in sperm count, and significant decrease in sperm motility with no effect on copulation or fertility rate	Iida et al. 1997; Kawashima et al. 1995

Table A-10. Summary of Relevant NOAEL and LOAEL Values Considered for Derivation of an Acute Oral MRL for Nitrobenzene

Species	Duration	NOAEL (mg/kg/day)	LOAEL (mg/kg/day)	Effect	Reference
Rat (Wistar)	Once/day for 14 days (GO)		100	Decreased testicular and epididymal weights; decreased serum prolactin, luteinizing hormone, follicle stimulating hormone, and testosterone; atrophic and degenerated seminiferous tubules	Oladele et al. 2020c
Other effects					
B6C3F1 mouse	Once/day for 14 days (GO)	30	100	Hepatic: Increased liver weight, hepatomegaly Immune: Decreased IgM AFC in spleen cells; decreased natural killer cell activity	Burns et al. 1994
Rat (Wistar)	Once/day for 14 days (GO)		100	Renal: Increased serum urea and creatinine; renal lesions including mild fibrosis and hemorrhage and marked glomerular shrinkage Neurological (SLOAEL): Neurobehavioral changes (decreased exploratory behavior and increased defecation); increased acetylcholinesterase activity in brain; degenerative lesions in the cerebellum, cerebrum, and hippocampus Endocrine: Decreased serum TSH Reproductive: Decreased testicular and epididymal weights; decreased serum prolactin, luteinizing hormone, follicle stimulating hormone, and testosterone; atrophic and degenerated seminiferous tubules	Oladele et al. 2020a, 2020b, 2021

AFC = antibody-forming cell; DNA = deoxyribonucleic acid; GD = gestation day; (GO) = gavage in oil vehicle; IgM = immunoglobulin M; LOAEL = lowest-observed-adverse-effect level; NOAEL = no-observed-adverse-effect level; SLOAEL = serious LOAEL; TSH = thyroid-stimulating hormone

The lowest LOAEL was 30 mg/kg/day in the study by Burns et al. (1994). Effects seen at the LOAEL included increased DNA synthesis, cell number, and numbers of granulocyte-monocyte progenitor cells in the bone marrow, indicating proliferative effects on this tissue. A NOAEL was not determined. These effects were considered for use in deriving the acute-duration MRL.

Selection of the Point of Departure: Burns et al. (1994) is the only oral exposure 14-day study that had reliable, dose-related data to inform an MRL derivation for the acute exposure duration. There was a clear and significant impact on bone marrow at 30 mg/kg/day that was linked to hematological effects

observed at ≥100 mg/kg/day (significant increase in reticulocytes) and consistent with effects seen in other studies of nitrobenzene (Burns et al. 1994).

Summary of the Principal Study:

Burns LA, Bradley SG, White KL, et al. 1994. Immunotoxicity of nitrobenzene in female B6C3F1 mice. Drug Chem Toxicol 17(3):271-315.

Groups of female B6C3F1 mice (eight/group) were administered nitrobenzene via gavage in corn oil at doses of 0 (vehicle control), 30, 100, or 300 mg/kg/day for 14 days. Animals were weighed on study days 1, 8, and 15. Animals were sacrificed on day 15 and blood was collected for hematology (erythrocyte and leukocyte number, hemoglobin, hematocrit, MCV, MCH, mean corpuscular hemoglobin concentration [MCHC], and total and differential leukocyte counts) and serum chemistry (aspartate aminotransferase [AST], ALT, urea nitrogen, glucose, albumin, and total protein). Animals underwent necropsy and select organs (liver, thymus, spleen, lungs, lymph nodes, kidneys, and brain) were removed, weighed, and prepared for histologic examination. Bone marrow was harvested for examination of immune cell (macrophage and granulocyte/monocyte) progenitors and DNA synthesis. Immune function assays included spleen IgM and IgG antibody response following stimulation with sheep erythrocytes (sRBC), spleen cell proliferation following stimulation with various mitogens, mixed leukocyte response, delayed hypersensitivity response, serum complement proteins, reticuloendothelial system sRBC clearance, peritoneal cell number, macrophage phagocytic activity, natural killer cell activity, and host resistance assay.

Death occurred in 8.5% of mice across several experiments at 300 mg/kg/day. Animals in the 300 mg/kg/day group showed treatment-related clinical signs, including ataxia, lethargy, circling, and head bobbing. No biologically significant (≥10% less than controls) effects on body weight were observed. Treatment-related hematology changes were observed and include increased MCV, MCH, and percent reticulocytes at ≥100 mg/kg/day. At 300 mg/kg/day, erythrocytes were decreased by 9%. Serum chemistry changes included increased ALT and bilirubin at 300 mg/kg/day. Total protein was increased at all doses.

Terminal body weights at 300 mg/kg/day were increased by 11%. Organ weight changes included increased absolute and relative liver weights and spleen weights at ≥100 mg/kg/day. Absolute lung weights were increased at all doses and relative lung to brain weight was increased at 300 mg/kg/day.

Mice exhibited hepatomegaly and splenomegaly and the spleen was dark and congested at ≥100 mg/kg/day. In mice in the 300 mg/kg group, there was mild hydropic degeneration foci around central veins in the liver. Mice in the 100 mg/kg group had mild congestion in the red pulp areas of the spleen and at 300 mg/kg/day, the spleen was dark red, with enlarged red pulp areas exhibiting severe congestion, hemosiderin, and extramedullary hematopoiesis (indicated by nucleated erythrocytes and immature granulocytes present). Incidence of these lesions was not reported.

Analysis of bone marrow showed increased DNA synthesis, cells per femur, and increased CFUs (granulocyte monocyte) per femur at all doses.

In the sRBC assay on day 4, there was a significant increase in spleen weight and spleen cell number at 300 mg/kg/day and reduced IgM AFC/10^6 spleen cells at ≥100 mg/kg/day. After 5 days, there was a significant increase in spleen weight and spleen cell number at ≥100 mg/kg/day. These effects did not persist after a 20-day recovery period.

In a spleen cell mitogenic response assay, 3H-thymidine incorporation in response to T cell mitogens (phytohemagglutinin and concanavalin A) was suppressed at ≥100 mg/kg/day. No effect was observed on B-cell mitogen (LPS) response. Decreased stimulation index was observed at ≥100 mg/kg/day, indicating reduced ability of splenic T cells to recognize and respond to alloantigens. At 300 mg/kg/day, there was an increase in the number of peritoneal cells and an increase in phagocytic activity. Natural killer cell activity in peritoneal cells was decreased at ≥100 mg/kg/day.

In the host resistance assay, mice treated with 300 mg/kg/day of nitrobenzene were more susceptible to *L. monocytogenes* and had a 338% higher mortality rate following a challenge of $6x10^3$ CFUs. Susceptibility to *Streptococcus pneumoniae* or *Plasmodium berghei* or herpes simplex virus were comparable to controls.

Table A-11 summarizes the statistically significant treatment-related effects that occurred at all dose levels, as well as the related data on reticulocyte count increases at ≥100 mg/kg/day.

Table A-11. Significant Noncancer Effects[a] Female B6C3F1 Mice Orally Exposed to Nitrobenzene for 14 Days

Effect	0	30 mg/kg/day	100 mg/kg/day	300 mg/kg/day
Hematological effects				
Reticulocyte percent[b]	1.03±0.09	1.09±0.17	3.51±0.43*	4.57±0.48*
Cells/femur ($x10^6$)	6.3±0.6	9.5±0.6*	9.4±0.7*	10.2±0.7*
DNA synthesis (cpm $x10^3$)	71.0±6.5	117.3±8.8*	126.6±4.7*	127.6±7.2*
CFU-GM/femur ($x10^4$)	0.58±0.06	0.88±0.06*	0.82±0.05*	1.01±0.06*

[a]Hematology changes are reported as mean ± standard error. Results marked with an asterisk (*) are significantly different from control ($p<0.05$) as reported by the study authors. Number of animals n=7–8 CFU-GM.
[b]Reticulocyte count is reported as the percentage of red blood cells.

CFU = colony-forming unit; cpm = counts per minute; DNA = deoxyribonucleic acid; GM = granulocyte monocyte

Source: Burns et al. 1994

Selection of the Point of Departure for the MRL: BMD modeling was conducted on the bone marrow effects occurring at all doses as reported in Table A-11 after converting the standard errors to SDs. The data were fit to all available continuous models in EPA's BMDS (version 3.2) using a BMR of 1 SD change from control. Adequate model fit was judged by four criteria: goodness-of-fit statistics (p-value >0.1), visual inspection of the dose-response curve, a BMD) that is not 10 times lower than the lowest non-zero dose, and scaled residual within ±2 units at the data point (except the control) closest to the predefined BMR. Among all of the models providing adequate fit to the data, the lowest BMDL was selected as the POD when the difference between the BMDLs estimated from these models was >3-fold; otherwise, the BMDL from the model with the lowest AIC was chosen. No suitable models were identified for CFU-granulocyte monocyte/femur.

In accordance with the selection criteria mentioned above, the exponential 4 model (constant variance) was selected as the best fit for the number of cells/femur data and the exponential model 5 (constant variance) was selected as the best fit for the DNA synthesis data. The BMD_{1SD} and $BMDL_{1SD}$ for increased number of cells/femur were 10.6 and 6.8 mg/kg/day, respectively. The BMD_{1SD} and $BMDL_{1SD}$ for increased DNA synthesis were 6.7 and 4.7 mg/kg/day, respectively. Therefore, the $BMDL_{1SD}$ of 4.7 mg/kg/day for increased DNA synthesis (as a marker for cell proliferation in the bone

marrow) was selected as the POD for the acute-duration oral MRL. The output from the BMDS modeling for increased DNA synthesis is presented in Table A-12 and the fit of the data to the selected model is presented in Figure A-2.

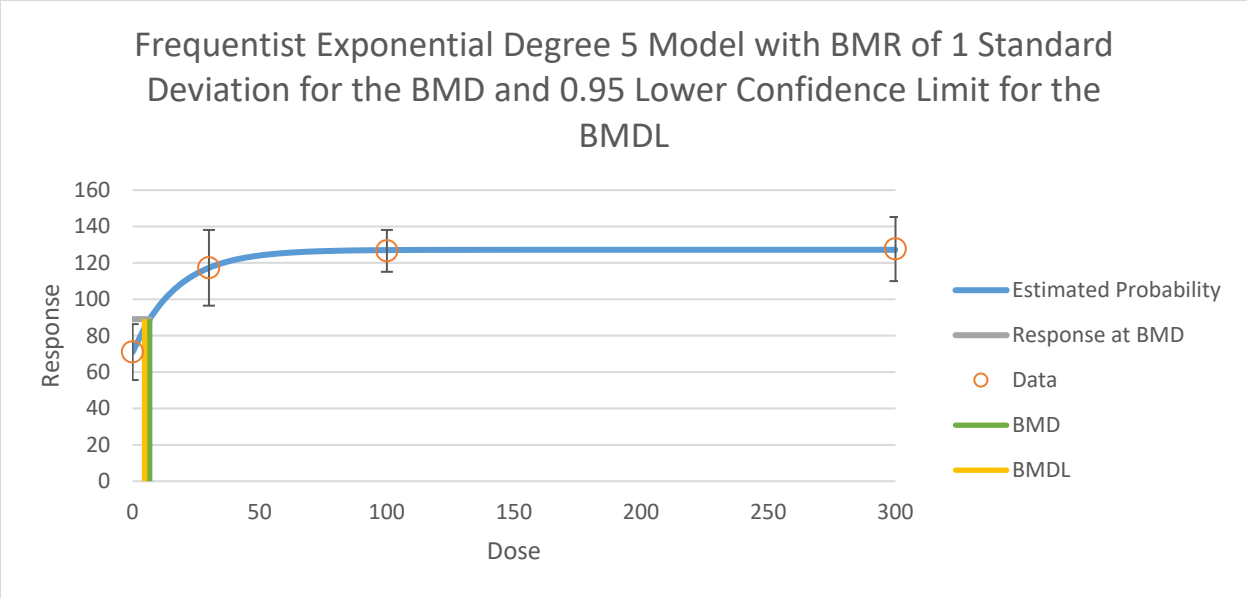
Table A-12. Results from BMD Analysis (Constant Variance) for DNA Synthesis in Femur Bone Marrow of Female B6C3F1 Mice after 14 Days of Oral Exposure to Nitrobenzene (Burns et al. 1994)

					Scaled residuals[c]	
Model	BMD_{1SD}[a] (mg/kg/day)	$BMDL_{1SD}$[a] (mg/kg/day)	p-Value[b]	AIC	Dose below BMD	Dose above BMD
Exponential (model 2)[d]			<0.0001	286.57	1.83	-0.64
Exponential (model 3)[d]			<0.0001	286.57	1.83	-0.64
Exponential (model 4)[d]	6.72	4.69	0.93	266.83	-0.0006	0.005
Exponential (model 5)[d,e]	**6.72**	**4.69**	**0.93**	**266.83**	**-0.0004**	**0.003**
Hill[f]			NA	268.83	-5.7×10^{-8}	3.96×10^{-8}
Polynomial (3-degree)[f]			<0.0001	285.87	1.78	-0.79
Polynomial (2-degree)[f]			<0.0001	285.87	1.78	-0.79
Power			<0.0001	285.87	1.78	-0.79
Linear			<0.0001	285.87	1.78	-0.79

[a]BMD and BMDL values for models that do not provide adequate fit are not included in this table.
[b]Values <0.1 fail to meet conventional χ^2 goodness-of-fit criteria.
[c]Scaled residuals at doses immediately below and above the BMD.
[d]Power restricted to ≥1.
[e]Selected model. The constant variance model provided an adequate fit to the data. Only the Exponential 4 and 5 models provided adequate fits to the means. The BMDLs were similar and sufficiently close (<3-fold); therefore, the model with the (slightly) lowest AIC was selected (Exponential 5 model).
[f]Coefficients restricted to be positive.

AIC = Akaike Information Criterion; BMD = benchmark dose (maximum likelihood estimate of the concentration associated with the selected benchmark response); BMDL = 95% lower confidence limit on the BMD (subscripts denote benchmark response); SD = standard deviation

Figure A-2. Fit of the Exponential 5 Model (Constant Variance) to Data for DNA Synthesis in Female B6C3F1 Mice with 14 Days of Oral Exposure to Nitrobenzene (Burns et al. 1994)



Using this POD an MRL was derived as follows.

Uncertainty Factor: The $BMDL_{1SD}$ of 4.7 mg/kg/day was divided by a total uncertainty factor of 100:

- 10 for extrapolation from animals to humans
- 10 for human variability

This results in the following MRL:

$$MRL = \frac{BMDL}{UF} = \frac{4.7}{100} = 0.05 \text{ mg/kg/day}$$

Other Additional Studies or Pertinent Information that Lend Support to this MRL: In human case studies of both inhalation and ingestion exposure to nitrobenzene, methemoglobinemia was the most common finding (Agrawal et al. 2011; Gupta et al. 2012; Ikeda and Kita 1964; Lee et al. 2013; Perera et al. 2009; Saxena and Prakash Saxena 2010). Additionally, experimental animal studies have demonstrated increased methemoglobin levels in mice and rats of both sexes exposed through any exposure route with acute, intermediate, and chronic (≥365 days) exposure durations (Biodynamics 1984; Cattley et al. 1994, 1995; CIIT 1993; Hamm et al. 1984; Medinsky and Irons 1985; Mitsumori et al. 1994; NTP 1982, 1983b). A variety of related adverse effects in the hematologic system, such as hemolytic anemia; extramedullary hematopoiesis, hemosiderosis, congestion, and lymphoid depletion of the spleen; and changes in bone marrow in response to anemia have also been observed with inhalation, oral, and dermal exposures in B6C3F1 mice, F344 rats, and Sprague-Dawley rats (Burns et al. 1994; Cattley et al. 1994, 1995; CIIT 1993; Hamm et al. 1984; Medinsky and Irons 1985; Mitsumori et al. 1994; NTP 1982, 1983a, 1983b).

Agency Contacts (Chemical Managers): Malcolm Williams

APPENDIX A

MINIMAL RISK LEVEL (MRL) WORKSHEET

Chemical name(s): Nitrobenzene
CAS number(s): 98-95-3
Date: January 2024
Profile Status: Final
Route: Oral
Duration: Intermediate
MRL 0.02 mg/kg/day
Critical Effect: Increased methemoglobin
Reference: NTP 1983a
Point of Departure: $BMDL_{1SD}$ of 1.8 mg/kg/day
Uncertainty Factor: 100
LSE Graph Key: 16
Species: Rat

MRL Summary: An intermediate-duration oral MRL of 0.02 mg/kg/day was derived for nitrobenzene based on evidence of increased methemoglobin levels in male F344 rats administered nitrobenzene via gavage at 9.375 mg/kg/day for 90 days (NTP 1983a). The MRL is based on a $BMDL_{1SD}$ of 1.8 mg/kg/day for increased methemoglobin. This was divided by a total uncertainty factor of 100 (10 for extrapolation from animals to humans and 10 for human variability).

Selection of the Critical Effect: There are three intermediate-duration studies that evaluated the adverse effects of nitrobenzene on the male reproductive system at the same doses (0 or 60 mg/kg/day) (Iida et al. 1997; Kawaguchi et al. 2004; Kawashima et al. 1995). In all of these studies, adverse effects on the testes and epididymis were observed. In addition, Kawaguchi et al. (2004) and Kawashima et al. (1995) reported adverse effects on sperm, including reduced count or concentration, motility, and viability.

There are two multi-dose oral intermediate-duration studies (Mitsumori et al. 1994; NTP 1983a) that also tested lower doses and evaluated comprehensive endpoints. Table A-13 shows the lowest effect levels from these two studies. Mitsumori et al. (1994) was a combined repeated-dose and reproduction/ developmental toxicity study with Sprague-Dawley rats in which a LOAEL of 20 mg/kg/day was identified for hematological, hepatic, renal, and reproductive effects. No NOAEL was identified. NTP (1983a) administered nitrobenzene doses via gavage to B6C3F1 mice and F344 rats for 90 days and evaluated a comprehensive list of endpoints. Effects observed at the lowest dose (9.375 mg/kg/day) included hematological, hepatic, and renal effects. All of these effects were considered for use in deriving the intermediate-duration oral MRL.

Table A-13. Summary of Relevant NOAEL and LOAEL Values Evaluated for Derivation of an Intermediate Oral MRL for Nitrobenzene

Species	Duration/route	NOAEL (mg/kg/day)	LOAEL (mg/kg/day)	Effect	Reference
Hematological effects					
F344 rat	Once/day for 90 days (GO)		9.375	Increased absolute reticulocyte count and decreased hemoglobin; increased methemoglobin in males (2.752 versus 1.131% in controls) and females (2.059 versus 0.941% in controls)	NTP 1983a
B6C3F1 mouse	Once/day for 90 days (GO)		18.75	Increased methemoglobin in males (2.162 versus 1.074% in controls) and females (1.198 versus 0.871% in controls), increased reticulocytes in females	NTP 1983a
F344 rat	Once/day for 54 days (GO)		20	Decreased erythrocytes, hemoglobin, and packed cell volume; increased hematopoiesis in bone marrow; increased spleen weight; extramedullary hematopoiesis and hemosiderin deposition in the spleen in males	Mitsumori et al. 1994
Hepatic effects					
F344 rat	Once/day for 90 days (GO)		9.375	Increased liver weights	NTP 1983a
F344 rat	Once/day for 54 days (GO)		20	Increased liver weights; centrilobular swelling of hepatocytes, hemosiderin deposition in Kupffer cells, extramedullary hematopoiesis in males	Mitsumori et al. 1994
Renal effects					
F344 rat	Once/day for 90 days (GO)		9.375 M	Increased kidney weights in males	NTP 1983a

Table A-13. Summary of Relevant NOAEL and LOAEL Values Evaluated for Derivation of an Intermediate Oral MRL for Nitrobenzene

Species	Duration/route	NOAEL (mg/kg/day)	LOAEL (mg/kg/day)	Effect	Reference
Reproductive effects					
F344 rat	Once/day for 90 days (GO)	18.75 M	37.5 M	Atrophic seminiferous tubules in one male; atrophied uteri in two females	NTP 1983a
Sprague-Dawley rat	Once/day for 54 days (GO)	20 M	60 M (SLOAEL)	~60% decrease in testes weight and ~20% decrease in epididymides weight; atrophy of seminiferous tubules (all males), Leydig cell hyperplasia, loss of intraluminal sperm or cell debris in epididymis	Mitsumori et al. 1994
B6C3F1 mouse	Once/day for 90 days (GO)	75M	150 M	Testicular atrophy	NTP 1983a

(GO) = gavage in oil vehicle; LOAEL = lowest-observed-adverse-effect level; M = male(s); NOAEL = no-observed-adverse-effect level; SLOAEL = serious LOAEL

Selection of the Principal Study and Point of Departure: While both Mitsumori et al. (1994) and NTP (1983a) are well-conducted studies demonstrating an array of toxicological effects of nitrobenzene exposure, the LOAEL of 9.375 mg/kg/day in the study by NTP (1983a) is lower than the LOAEL of 20 mg/kg/day (lowest dose tested) in Mitsumori et al. (1994). Neither study identified a NOAEL. Therefore, NTP (1983a) was selected as the principal study for derivation of the intermediate-duration oral MRL.

Summary of the Principal Study:

NTP. 1983a. Report on subchronic toxicity via gavage of nitrobenzene (C60082) in Fischer 344 rats and B6C3F1 mice. Worcester, MA: National Toxicology Program. MRI-NTP 09-83-19.

Groups of male and female F344 rats and B6C3F1 mice were administered nitrobenzene via gavage in corn oil at doses of 0, 9.345, 18.75, 37.5, 75, and 150 mg/kg/day and 0, 18.75, 37.5, 75, 150, and 300 mg/kg/day, respectively, for 90 days. Experimental groups consisted of 10 male and 10 female animals of each strain and species. Animals were examined for clinical abnormalities twice daily and weighed weekly. Animals were sacrificed at the end of the exposure period. Blood was drawn from the external jugular for hematology and serum chemistry analysis. Animals underwent gross necropsy and select tissues (liver, brain, right kidney, thymus, heart, lung, and right testis) were weighed. Other tissues were prepared for histological analysis (cerebrum, cerebellum, pituitary gland, larynx, tongue, jejunum, pancreas, gall bladder, thymus, kidney, prostate, sternum, nose/turbinates, thyroid glands, esophagus, ileum, salivary gland, spleen, adrenal glands, testes, uterus, mammary glands, bone marrow, gross lesion, parathyroid glands, stomach, cecum, lymph nodes, heart, lungs, urinary bladder, skeletal muscle, rib bone, trachea, duodenum, colon, rectum, liver, seminal vesicles, epididymides, ovaries, eyes, sciatic nerve, skin, and zymbal's gland).

Mortality occurred in nine male and three female rats in the 150 mg/kg/day group. Mortality in mice included three males at 300 mg/kg/day and one male at 75 mg/kg/day. One female in the control and one in the 18.75 mg/kg/day died accidentally. Clinical signs of ataxia, head tilt, lethargy, and trembling were observed at 150 and 300 mg/kg/day in rats and mice, respectively. Cyanosis was observed in all rats at doses ≥75 mg/kg/day. Due to high mortality in male rats at the high dose, study data from these animals were not included.

Regarding hematologic effects, mice and rats of both sexes had increased methemoglobin levels at all doses. Rats of both sexes had increased reticulocytes at ≥9.375 mg/kg/day as well as decreased hemoglobin. In addition, male rats had decreased MCV and MCH, while female rats had decreased hematocrit at all doses. Increased reticulocytes were observed in female mice at ≥18.75 mg/kg/day and in males at ≥37.5 mg/kg/day; decreases in red blood cells, hematocrit, and hemoglobin were seen in both sexes at ≥150 mg/kg/day. Male mice exhibited anisocytosis and polychromasia at 300 mg/kg/day.

Liver weights and liver to body and brain weight ratios were increased in rats of both sexes at all doses. Right kidney and kidney to body weights were increased in a dose related manner in rats of both sexes at ≥9.375 in males and ≥18.75 in females and right kidney to brain weight ratios were increased ≥18.75 mg/kg/day in rats of both sexes. In male rats, weights of the right testis and right testis to body and brain weight ratios were reduced at 75 mg/kg/day. In female rats, heart weights and heart to brain weight ratios were increased at all doses and heart to body weight ratios were increased at ≥75 mg/kg/day and lung weights and lung to brain and body weight ratios were increased at ≥18.75 mg/kg/day. In male mice, liver weights and liver to brain and body weight ratios were increased ≥150 mg/kg/day. Right testis weight and testis to body and brain weight ratios were decreased in male mice at 300 mg/kg/day. In female mice, liver weights were increased at all doses and liver to body and brain weight ratios were increased at ≥37.5 mg/kg/day. Thymus weights and thymus to body and brain weight ratios were increased at all doses in female mice.

Histological effects include congestion of the spleen in most of the treated rats of both sexes. Hemosiderin pigment was observed in the red pulp of the spleen, while lymphoid depletion was noted in the white pulp. In mice lymphoid depletion of the spleen was noted at ≥150 mg/kg/day. Hepatocellular hypertrophy in the centrilobular zone were observed in mice of both sexes. Female rats had increased incidence of renal tubular cell pigmentation. There was increased incidence of brainstem hemorrhage in 5/10 male rats at 75 mg/kg/day and 7/10 female rats at 150 mg/g/day. In male rats, seminiferous tubules were atrophied at ≥37.5 mg/kg/day; atrophied testes and hypospermatogenesis were noted in male rats (10/10) at 75 mg/kg/day and in male mice (4/10) at 300 mg/kg/day. In mice lymphoid depletion of the spleen was noted at ≥150 mg/kg/day. Hepatocellular hypertrophy in the centrilobular zone was observed in mice of both sexes.

In summary, effects of nitrobenzene were seen at all doses in both rats and mice. Because rats were tested at a lower dose (9.375 versus 18.75 mg/kg/day in mice) and are more sensitive to the effects of nitrobenzene, data for rats were considered for MRL derivation. The main effects (hematologic, hepatic, and renal) observed at ≥9.375 mg/kg/day in rats in this study are shown in Table A-14.

Table A-14. Significant Noncancer Effects[a] F344 Rats Orally Exposed to Nitrobenzene for 90 Days

	Dose (mg/kg/day)					
Effect	0	9.375	18.75	37.5	75	150
Male						
Methemoglobin percent	1.131±0.579	2.752±0.583*	4.22±1.145*	5.624±0.85*	7.307±1.438*	–
Relative (to body weight) liver weight	3.51847± 0.21612	4.03612± 0.20049*	4.37246± 0.14415*	4.76839± 0.21788*	5.1545± 0.14804*	–
Relative (to body weight) kidney weight	3.09542± 0.20207	3.29664± 0.20499*	3.36071± 0.11055*	3.29808± 0.37913	3.44410± 0.23201*	–
Female						
Methemoglobin percent	0.941±0.031	2.059±0.449*	3.623±1.088*	5.269±0.757*	6.851±2.250*	12.771± 1.825*
Relative (to body weight) liver weight	3.42905± 0.15529	3.76085± 0.08417*	3.95375± 0.17757*	4.22849± 0.17951*	4.87532± 0.21802*	5.20999± 0.28071*
Absolute reticulocyte count	0.218±0.034	0.297±0.029*	0.38±0.053*	0.491±0.071*	0.637±0.108*	1.871±0.173*
Relative reticulocyte count[b]	2.6±0.37	3.69±0.32*	4.75±0.68*	6.28±0.9*	8.72±1.49*	32.07±3.56*

[a]Hematology changes are reported as mean±standard deviation. Results marked with an asterisk (*) are significantly different from control ($p<0.05$) as reported by the study authors. Number of animals = 10 except in the 150 mg/kg/day group (n=7).
Greyed cells: data not included for males at the high dose due to high mortality (only one male survived at this dose)
[b]Reticulocyte count is reported as the percentage of red blood cells that are reticulocytes (number of reticulocytes divided by the total number of red blood cells, multiplied by 100). For adults, a normal range is considered to be 0.5–1.5%.

Source: NTP 1983a

Selection of the Point of Departure for the MRL: As shown in Table A-14, the endpoint with the largest change at the LOAEL was methemoglobin (which was more than doubled at the LOAEL, compared with smaller changes in other endpoints), suggesting that methemoglobin is the most sensitive endpoint at the LOAEL. In addition, many of the hematological, hepatic, and renal effects of nitrobenzene are related to the induction of methemoglobin (see *Metabolic Mechanisms in* Section 3.1.3). Therefore, the data on methemoglobin in blood of rats were selected for use in deriving the MRL. Table A-14 shows the methemoglobin data in male and female F344 rats in the study by NTP (1983a).

BMD modeling was conducted on the methemoglobin data for both sexes of rat. The highest dose group was dropped for males as a result of a high mortality rate. The data were fit to all available continuous models in EPA's BMDS (version 3.2) using a BMR of 1 SD. Adequate model fit was judged by four criteria: goodness-of-fit statistics (p-value >0.1), visual inspection of the dose-response curve, a BMDL that is not 10 times lower than the lowest non-zero dose and scaled residual within ±2 units at the data point (except the control) closest to the predefined BMR. Among the models providing adequate fit to the data, the lowest BMDL was selected as the POD when the difference between the BMDLs estimated from these models was >3-fold; otherwise, the BMDL from the model with the lowest AIC was chosen. No suitable models were identified for the female rats. The constant variance model did not provide an adequate fit to the male rat variance data. With a nonconstant variance (NCV) model applied, an adequate fit to the variance was provided. The Exponential 4 and 5 models and the Hill model provided adequate fit to the means.

The results of the modeling are presented in Table A-15. In accordance with the above guidance, the model with the lowest AIC among the adequately fitting models was selected; this was the Exponential 5-NCV model. The exposure-response curve for this model is displayed in Figure A-3. The $BMDL_{1SD}$ from this model is 1.8 mg/kg/day.

Table A-15. Results from BMD Analysis (Non-Constant Variance) for Increased Methemoglobin in Male F344 Rats Exposed to Nitrobenzene via Gavage for 90 Days (NTP 1983a)

					Scaled residuals[c]	
Model	BMD_{1SD}[a] (mg/kg/day)	$BMDL_{1SD}$[a] (mg/kg/day)	p-Value[b]	AIC	Dose below BMD	Dose above BMD
Exponential (model 2)[d]			<0.0001	177.02	2.04	2.72
Exponential (model 3)[d]			<0.0001	177.02	2.04	2.72
Exponential (model 4)[d]	2.62	1.81	0.65	136.42	-0.07	-0.20
Exponential (model 5)[d,e]	**2.62**	**1.81**	**0.65**	**136.42**	**-0.06**	**-0.20**
Hill[f]	3.03	1.65	0.42	138.21	-0.08	-0.09
Polynomial (4-degree)[f]			<0.0001	157.95	-2.33	0.84
Polynomial (3-degree)[f]			<0.0001	157.95	-2.33	0.84
Polynomial (2-degree)[f]			<0.0001	157.95	-2.33	0.84

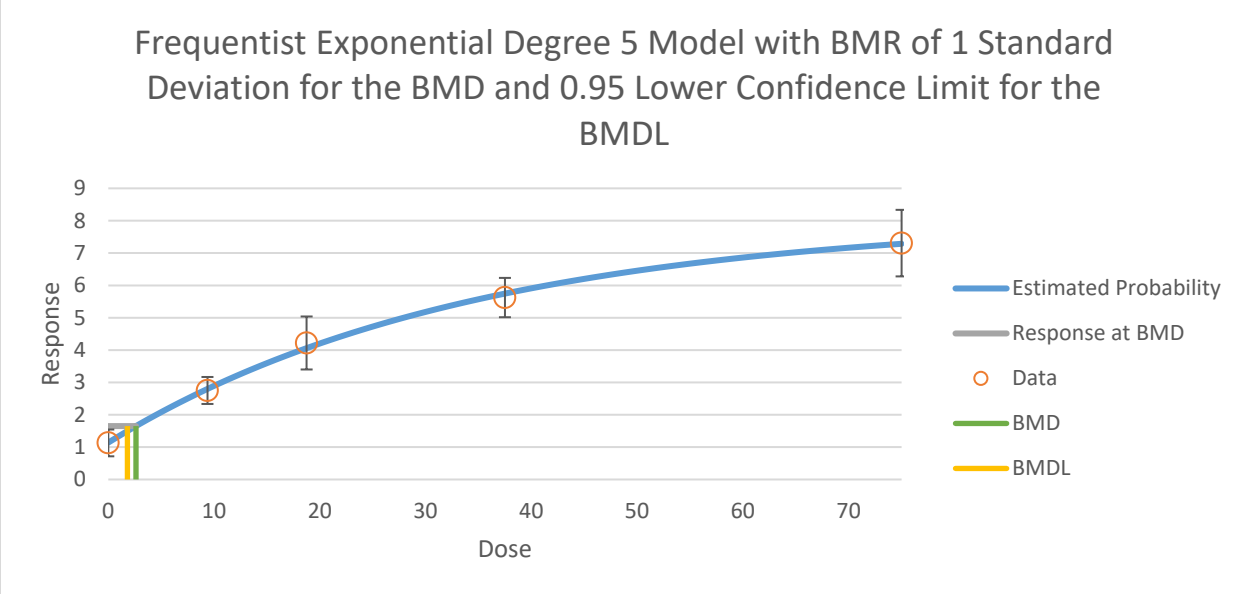
Table A-15. Results from BMD Analysis (Non-Constant Variance) for Increased Methemoglobin in Male F344 Rats Exposed to Nitrobenzene via Gavage for 90 Days (NTP 1983a)

Model	BMD_{1SD}[a] (mg/kg/day)	$BMDL_{1SD}$[a] (mg/kg/day)	p-Value[b]	AIC	Scaled residuals[c]	
					Dose below BMD	Dose above BMD
Power			<0.0001	157.95	-2.33	0.84
Linear			<0.0001	157.95	-2.33	0.84

[a]BMD and BMDL values for models that do not provide adequate fit are not included in this table.
[b]Values <0.1 fail to meet conventional χ^2 goodness-of-fit criteria.
[c]Scaled residuals at doses immediately below and above the BMD.
[d]Power restricted to ≥1.
[e]Selected model. The constant variance model did not provide an adequate fit to the data. With nonconstant variance applied, only the Exponential 4 and 5 models and the Hill model provided adequate fits to the means. The BMDLs of the fit models were sufficiently close (<3-fold); therefore, the model with the lowest AIC was selected (Exponential 5 model).
[f]Coefficients restricted to be positive.

AIC = Akaike Information Criterion; BMD = benchmark dose (maximum likelihood estimate of the concentration associated with the selected benchmark response); BMDL = 95% lower confidence limit on the BMD (subscripts denote benchmark response); SD = standard deviation

Figure A-3. Fit of Exponential 5 Model (Nonconstant Variance) to Data on Increased Methemoglobin in Male F344 Rats Exposed to Nitrobenzene via Gavage for 90 Days (NTP 1983a)



Uncertainty Factor: The $BMDL_{1SD}$ of 1.8 mg/kg/day was divided by a total uncertainty factor of 100:

- 10 for extrapolation from animals to humans
- 10 for human variability

This results in the following MRL:

$$MRL = \frac{BMDL}{UF} = \frac{1.8}{100} = 0.02\frac{mg}{kg}/day$$

Other Additional Studies or Pertinent Information that Lend Support to this MRL: In human case studies of both inhalation and ingestion exposure to nitrobenzene, methemoglobinemia was the most common finding (Agrawal et al. 2011; Gupta et al. 2012; Ikeda and Kita 1964; Lee et al. 2013; Perera et al. 2009; Saxena and Prakash Saxena 2010). Additionally, experimental animal studies have demonstrated increased methemoglobin levels in mice and rats of both sexes exposed through any exposure route with acute, intermediate, and chronic (≥365 days) exposure durations (Biodynamics 1984; Cattley et al. 1994, 1995; CIIT 1993; Hamm et al. 1984; Medinsky and Irons 1985; Mitsumori et al. 1994; NTP 1982, 1983b). A variety of related adverse effects in the hematologic system, such as hemolytic anemia; extramedullary hematopoiesis, hemosiderosis, congestion, and lymphoid depletion of the spleen; and changes in bone marrow in response to anemia have also been observed with inhalation, oral, and dermal exposures in B6C3F1 mice, F344 rats, and Sprague-Dawley rats (Burns et al. 1994; Cattley et al. 1994, 1995; CIIT 1993; Hamm et al. 1984; Medinsky and Irons 1985; Mitsumori et al. 1994; NTP 1982, 1983a, 1983b).

Agency Contacts (Chemical Managers): Malcolm Williams

MINIMAL RISK LEVEL (MRL) WORKSHEET

Chemical name(s): Nitrobenzene
CAS number(s): 98-95-3
Date: January 2024
Profile Status: Final
Route: Oral
Duration: Chronic

MRL Summary: There are insufficient data for derivation of a chronic-duration oral MRL.

Rationale for Not Deriving an MRL: No studies that evaluated chronic-duration oral exposure to nitrobenzene were located; therefore, no MRL can be derived.

Agency Contacts (Chemical Managers): Malcolm Williams

APPENDIX B. LITERATURE SEARCH FRAMEWORK FOR NITROBENZENE

The objective of the toxicological profile is to evaluate the potential for human exposure and the potential health hazards associated with inhalation, oral, or dermal/ocular exposure to nitrobenzene.

B.1 LITERATURE SEARCH AND SCREEN

A literature search and screen were conducted to identify studies examining health effects, toxicokinetics, mechanisms of action, susceptible populations, biomarkers, chemical interactions, physical and chemical properties, production, use, environmental fate, environmental releases, and environmental and biological monitoring data for nitrobenzene. ATSDR primarily focused on peer-reviewed articles without publication date or language restrictions. Non-peer-reviewed studies that were considered relevant to the assessment of the health effects of nitrobenzene have undergone peer review by at least three ATSDR-selected experts who have been screened for conflict of interest. The inclusion criteria used to identify relevant studies examining the health effects of nitrobenzene are presented in Table B-1.

Table B-1. Inclusion Criteria for the Literature Search and Screen

Health Effects
Species
Human
Laboratory mammals
Route of exposure
Inhalation
Oral
Dermal (or ocular)
Parenteral (these studies will be considered supporting data)
Health outcome
Death
Systemic effects
Body weight effects
Respiratory effects
Cardiovascular effects
Gastrointestinal effects
Hematological effects
Musculoskeletal effects
Hepatic effects
Renal effects
Dermal effects
Ocular effects
Endocrine effects
Immunological effects
Neurological effects
Reproductive effects
Developmental effects
Other noncancer effects

Table B-1. Inclusion Criteria for the Literature Search and Screen

Cancer
Toxicokinetics
Absorption
Distribution
Metabolism
Excretion
PBPK models
Biomarkers
Biomarkers of exposure
Biomarkers of effect
Interactions with other chemicals
Potential for human exposure
Releases to the environment
Air
Water
Soil
Environmental fate
Transport and partitioning
Transformation and degradation
Environmental monitoring
Air
Water
Sediment and soil
Other media
Biomonitoring
General populations
Occupation populations

B.1.1 Literature Search

The current literature search was intended to update the Draft Toxicological Profile for Nitrobenzene released for public comment in 2022; thus, the literature search was restricted to studies published between January 2019 and July 2022. The following main databases were searched in July 2022:

- PubMed
- National Technical Reports Library (NTRL)
- Scientific and Technical Information Network's TOXCENTER

The search strategy used the chemical names, Chemical Abstracts Service (CAS) numbers, synonyms, Medical Subject Headings (MeSH) headings, and keywords for nitrobenzene. The query strings used for the literature search are presented in Table B-2.

The search was augmented by searching the Toxic Substances Control Act Test Submissions (TSCATS), NTP website, and National Institute of Health Research Portfolio Online Reporting Tools Expenditures

and Results (NIH RePORTER) databases using the queries presented in Table B-3. Additional databases were searched in the creation of various tables and figures, such as the TRI Explorer, the Substance Priority List (SPL) resource page, and other items as needed. Regulations applicable to nitrobenzene were identified by searching international and U.S. agency websites and documents.

Review articles were identified and used for the purpose of providing background information and identifying additional references. ATSDR also identified reports from the grey literature, which included unpublished research reports, technical reports from government agencies, conference proceedings and abstracts, and theses and dissertations.

Table B-2. Database Query Strings

Database search date	Query string
PubMed	
07/2022	(98-95-3[rn] OR "1-Nitrobenzene"[tw] OR "mononitrobenzene"[tw] OR "Nitrobenzene"[tw] OR "Nitrobenzol"[tw] OR "Oil of Mirbane"[tw] OR "p-Nitrobenzene"[tw] OR "benzene, nitro"[tw] OR "Benzene, nitro-"[tw] OR "Essence of mirbane"[tw] OR "Essence of Myrbane"[tw] OR "Mirbane oil"[tw] OR "Oil of Myrbane"[tw]) AND (2019:2022[mhda] OR 2019:2022[edat] OR 2019:2022[crdat] OR 2019:2022[dp])
NTRL	
07/2022	2019-present "Nitrobenzene" OR "Oil of Mirbane" OR "1-Nitrobenzene" OR "Mononitrobenzene" OR "p-Nitrobenzene" OR "Nitrobenzol" OR "Essence of mirbane" OR "Essence of Myrbane" OR "Mirbane oil" OR "Oil of Myrbane" OR "Benzene, nitro-" OR "benzene, nitro"
Toxcenter	
07/2022	FILE 'TOXCENTER' ENTERED AT 13:29:49 ON 26 JUL 2022 CHARGED TO COST=EH038.13.07.LB.04 L1 7736 SEA FILE=TOXCENTER 98-95-3 L2 7673 SEA FILE=TOXCENTER L1 NOT TSCATS/FS L3 6570 SEA FILE=TOXCENTER L2 NOT PATENT/DT L4 4498 SEA FILE=TOXCENTER L3 AND PY>=1997 ACTIVATE TOXQUERY/Q --------- L5 QUE (CHRONIC OR IMMUNOTOX? OR NEUROTOX? OR TOXICOKIN? OR BIOMARKER? OR NEUROLOG?) L6 QUE (PHARMACOKIN? OR SUBCHRONIC OR PBPK OR EPIDEMIOLOGY/ST,CT, IT) L7 QUE (ACUTE OR SUBACUTE OR LD50# OR LD(W)50 OR LC50# OR LC(W)50) L8 QUE (TOXICITY OR ADVERSE OR POISONING)/ST,CT,IT L9 QUE (INHAL? OR PULMON? OR NASAL? OR LUNG? OR RESPIR?) L10 QUE ((OCCUPATION? OR WORKPLACE? OR WORKER?) AND EXPOS?) L11 QUE (ORAL OR ORALLY OR INGEST? OR GAVAGE? OR DIET OR DIETS OR DIETARY OR DRINKING(W)WATER?) L12 QUE (MAXIMUM AND CONCENTRATION? AND (ALLOWABLE OR PERMISSIBLE)) L13 QUE (ABORT? OR ABNORMALIT? OR EMBRYO? OR CLEFT? OR FETUS?)

Table B-2. Database Query Strings

Database search date	Query string
	L14 QUE (FOETUS? OR FETAL? OR FOETAL? OR FERTIL? OR MALFORM? OR OVUM?)
	L15 QUE (OVA OR OVARY OR PLACENTA? OR PREGNAN? OR PRENATAL?)
	L16 QUE (PERINATAL? OR POSTNATAL? OR REPRODUC? OR STERIL? OR TERATOGEN?)
	L17 QUE (SPERM OR SPERMAC? OR SPERMAG? OR SPERMATI? OR SPERMAS? OR SPERMATOB? OR SPERMATOC? OR SPERMATOG?)
	L18 QUE (SPERMATOI? OR SPERMATOL? OR SPERMATOR? OR SPERMATOX? OR SPERMATOZ? OR SPERMATU? OR SPERMI? OR SPERMO?)
	L19 QUE (NEONAT? OR NEWBORN? OR DEVELOPMENT OR DEVELOPMENTAL?)
	L20 QUE (ENDOCRIN? AND DISRUPT?)
	L21 QUE (ZYGOTE? OR CHILD OR CHILDREN OR ADOLESCEN? OR INFANT?)
	L22 QUE (WEAN? OR OFFSPRING OR AGE(W)FACTOR?)
	L23 QUE (DERMAL? OR DERMIS OR SKIN OR EPIDERM? OR CUTANEOUS?)
	L24 QUE (CARCINOG? OR COCARCINOG? OR CANCER? OR PRECANCER? OR NEOPLAS?)
	L25 QUE (TUMOR? OR TUMOUR? OR ONCOGEN? OR LYMPHOMA? OR CARCINOM?)
	L26 QUE (GENETOX? OR GENOTOX? OR MUTAGEN? OR GENETIC(W)TOXIC?)
	L27 QUE (NEPHROTOX? OR HEPATOTOX?)
	L28 QUE (ENDOCRIN? OR ESTROGEN? OR ANDROGEN? OR HORMON?)
	L29 QUE (OCCUPATION? OR WORKER? OR WORKPLACE? OR EPIDEM?)
	L30 QUE L5 OR L6 OR L7 OR L8 OR L9 OR L10 OR L11 OR L12 OR L13 OR L14 OR L15 OR L16 OR L17 OR L18 OR L19 OR L20 OR L21 OR L22 OR L23 OR L24 OR L25 OR L26 OR L27 OR L28 OR L29
	L31 QUE (RAT OR RATS OR MOUSE OR MICE OR GUINEA(W)PIG? OR MURIDAE OR DOG OR DOGS OR RABBIT? OR HAMSTER? OR PIG OR PIGS OR SWINE OR PORCINE OR MONKEY? OR MACAQUE?)
	L32 QUE (MARMOSET? OR FERRET? OR GERBIL? OR RODENT? OR LAGOMORPHA OR BABOON? OR CANINE OR CAT OR CATS OR FELINE OR MURINE)
	L33 QUE L30 OR L31 OR L32
	L34 QUE (NONHUMAN MAMMALS)/ORGN
	L35 QUE L33 OR L34
	L36 QUE (HUMAN OR HUMANS OR HOMINIDAE OR MAMMALS OR MAMMAL? OR PRIMATES OR PRIMATE?)
	L37 QUE L35 OR L36

	L38 177 SEA FILE=TOXCENTER L4 AND L31
	D SCAN L38

Table B-3. Strategies to Augment the Literature Search

Source	Query and number screened when available
TSCATS via ChemView	
07/2022	98-95-3
NTP	
07/2022	"Nitrobenzene" "Oil of Mirbane" "1-Nitrobenzene" "Mononitrobenzene" "p-Nitrobenzene" "Nitrobenzol" "Essence of mirbane" "Essence of Myrbane" "Mirbane oil" "Oil of Myrbane" "Benzene, nitro-" "benzene, nitro"
Regulations.gov	
07/2022	Nitrobenzene 1-Nitrobenzene p-Nitrobenzene mononitrobenzene Nitrobenzol Oil of Mirbane benzene, nitro Benzene, nitro- Essence of mirbane Essence of Myrbane Mirbane oil Oil of Myrbane
NPIRS	
07/2022	98-95-3
NIH RePORTER	
10/2022	Search Criteria-- Fiscal Year: Active Projects; Text Search: "1-Nitrobenzene" OR "mononitrobenzene" OR "Nitrobenzene" OR "Nitrobenzol" OR "Oil of Mirbane" OR "p-Nitrobenzene" OR "benzene, nitro" OR "Benzene, nitro-" OR "Essence of mirbane" OR "Essence of Myrbane" OR "Mirbane oil" OR "Oil of Myrbane" (advanced) Limit to: Project Title, Project Terms, Project Abstracts
Other	Identified throughout the assessment process

The 2022 results were:

- Number of records identified from PubMed, NTRL, and TOXCENTER (after duplicate removal): 677
- Number of records identified from other strategies: 55
- Total number of records to undergo literature screening: 732

B.1.2 Literature Screening

A two-step process was used to screen the literature search to identify relevant studies on nitrobenzene:

- Title and abstract screen
- Full text screen

Title and Abstract Screen. Within the reference library, titles and abstracts were screened manually for relevance. Studies that were considered relevant (see Table B-1 for inclusion criteria) were moved to the

second step of the literature screening process. Studies were excluded when the title and abstract clearly indicated that the study was not relevant to the toxicological profile.

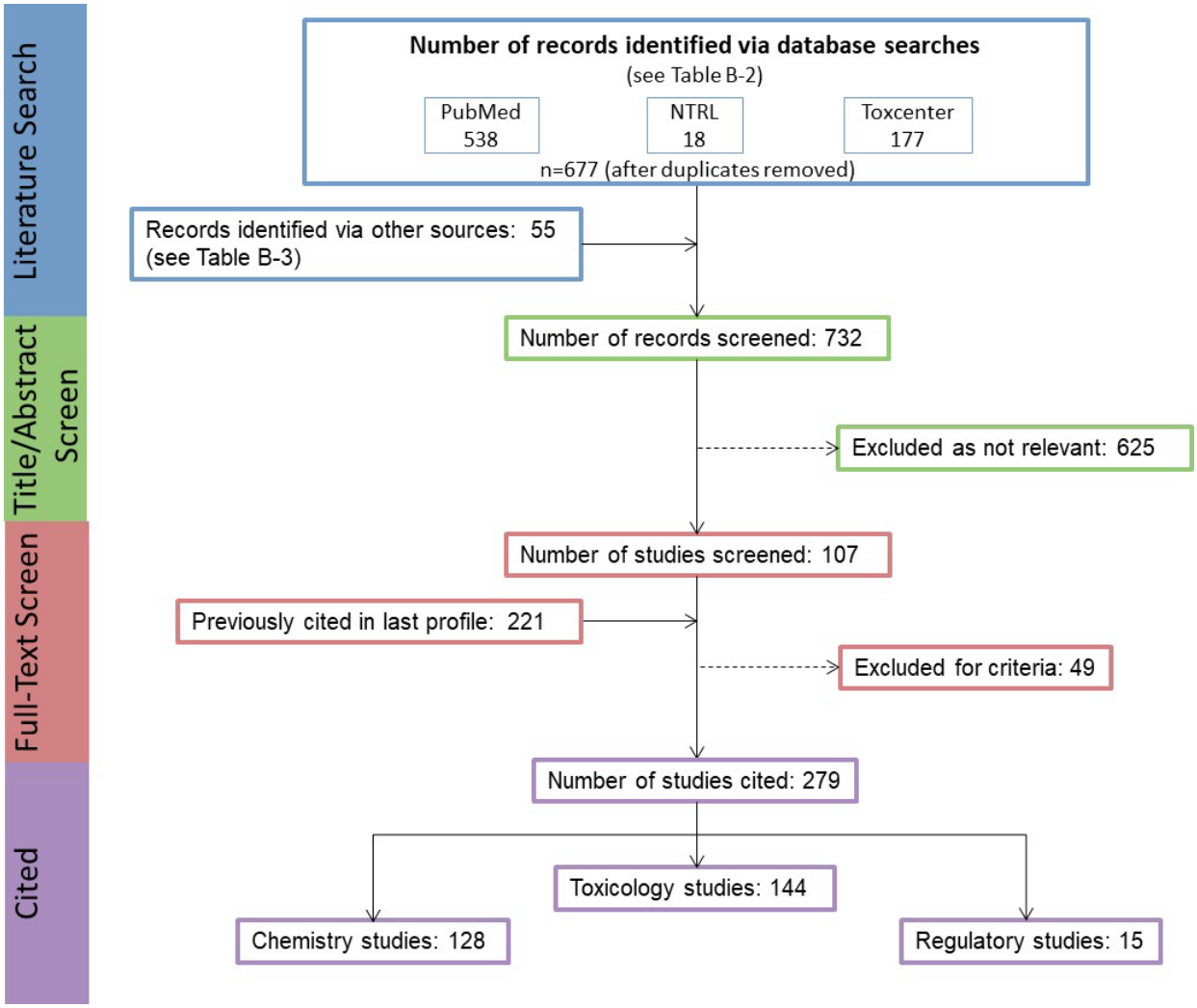
- Number of titles and abstracts screened: 732
- Number of studies considered relevant and moved to the next step: 107

Full Text Screen. The second step in the literature screening process was a full text review of individual studies considered relevant in the title and abstract screen step. Each study was reviewed to determine whether it was relevant for inclusion in the toxicological profile.

- Number of studies undergoing full text review: 107
- Number of studies cited in the pre-public draft of the toxicological profile: 221
- Total number of studies cited in the profile: 279

A summary of the results of the literature search and screening is presented in Figure B-1.

Figure B-1. July 2022 Literature Search Results and Screen for Nitrobenzene



APPENDIX C. USER'S GUIDE

Chapter 1. Relevance to Public Health

This chapter provides an overview of U.S. exposures, a summary of health effects based on evaluations of existing toxicologic, epidemiologic, and toxicokinetic information, and an overview of the minimal risk levels. This is designed to present interpretive, weight-of-evidence discussions for human health endpoints by addressing the following questions:

1. What effects are known to occur in humans?

2. What effects observed in animals are likely to be of concern to humans?

3. What exposure conditions are likely to be of concern to humans, especially around hazardous waste sites?

Minimal Risk Levels (MRLs)

Where sufficient toxicologic information is available, ATSDR derives MRLs for inhalation and oral routes of entry at each duration of exposure (acute, intermediate, and chronic). These MRLs are not meant to support regulatory action, but to acquaint health professionals with exposure levels at which adverse health effects are not expected to occur in humans.

MRLs should help physicians and public health officials determine the safety of a community living near a hazardous substance emission, given the concentration of a contaminant in air or the estimated daily dose in water. MRLs are based largely on toxicological studies in animals and on reports of human occupational exposure.

MRL users should be familiar with the toxicologic information on which the number is based. Section 1.2, Summary of Health Effects, contains basic information known about the substance. Other sections, such as Section 3.2 Children and Other Populations that are Unusually Susceptible and Section 3.4 Interactions with Other Substances, provide important supplemental information.

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

To derive an MRL, ATSDR generally selects the most sensitive endpoint which, in its best judgement, represents the most sensitive human health effect for a given exposure route and duration. ATSDR cannot make this judgement or derive an MRL unless information (quantitative or qualitative) is available for all potential systemic, neurological, and developmental effects. If this information and reliable quantitative data on the chosen endpoint are available, ATSDR derives an MRL using the most sensitive species (when information from multiple species is available) with the highest no-observed-adverse-effect level (NOAEL) that does not exceed any adverse effect levels. When a NOAEL is not available, a lowest-observed-adverse-effect level (LOAEL) can be used to derive an MRL, and an uncertainty factor of 10 must be employed. Additional uncertainty factors of 10 must be used both for human variability to protect sensitive subpopulations (people who are most susceptible to the health effects caused by the substance) and for interspecies variability (extrapolation from animals to humans). In deriving an MRL, these individual uncertainty factors are multiplied together. The product is then divided into the inhalation concentration or oral dosage selected from the study. Uncertainty factors used in developing a

substance-specific MRL are provided in the footnotes of the levels of significant exposure (LSE) tables that are provided in Chapter 2. Detailed discussions of the MRLs are presented in Appendix A.

Chapter 2. Health Effects

Tables and Figures for Levels of Significant Exposure (LSE)

Tables and figures are used to summarize health effects and illustrate graphically levels of exposure associated with those effects. These levels cover health effects observed at increasing dose concentrations and durations, differences in response by species and MRLs to humans for noncancer endpoints. The LSE tables and figures can be used for a quick review of the health effects and to locate data for a specific exposure scenario. The LSE tables and figures should always be used in conjunction with the text. All entries in these tables and figures represent studies that provide reliable, quantitative estimates of NOAELs, LOAELs, or Cancer Effect Levels (CELs).

The legends presented below demonstrate the application of these tables and figures. Representative examples of LSE tables and figures follow. The numbers in the left column of the legends correspond to the numbers in the example table and figure.

TABLE LEGEND

See Sample LSE Table (page C-5)

(1) Route of exposure. One of the first considerations when reviewing the toxicity of a substance using these tables and figures should be the relevant and appropriate route of exposure. Typically, when sufficient data exist, three LSE tables and two LSE figures are presented in the document. The three LSE tables present data on the three principal routes of exposure (i.e., inhalation, oral, and dermal). LSE figures are limited to the inhalation and oral routes. Not all substances will have data on each route of exposure and will not, therefore, have all five of the tables and figures. Profiles with more than one chemical may have more LSE tables and figures.

(2) Exposure period. Three exposure periods—acute (<15 days), intermediate (15–364 days), and chronic (≥365 days)—are presented within each relevant route of exposure. In this example, two oral studies of chronic-duration exposure are reported. For quick reference to health effects occurring from a known length of exposure, locate the applicable exposure period within the LSE table and figure.

(3) Figure key. Each key number in the LSE table links study information to one or more data points using the same key number in the corresponding LSE figure. In this example, the study represented by key number 51 identified NOAELs and less serious LOAELs (also see the three "51R" data points in sample LSE Figure 2-X).

(4) Species (strain) No./group. The test species (and strain), whether animal or human, are identified in this column. The column also contains information on the number of subjects and sex per group. Chapter 1, Relevance to Public Health, covers the relevance of animal data to human toxicity and Section 3.1, Toxicokinetics, contains any available information on comparative toxicokinetics. Although NOAELs and LOAELs are species specific, the levels are extrapolated to equivalent human doses to derive an MRL.

(5) Exposure parameters/doses. The duration of the study and exposure regimens are provided in these columns. This permits comparison of NOAELs and LOAELs from different studies. In this case (key number 51), rats were orally exposed to “Chemical X” via feed for 2 years. For a

more complete review of the dosing regimen, refer to the appropriate sections of the text or the original reference paper (i.e., Aida et al. 1992).

(6) Parameters monitored. This column lists the parameters used to assess health effects. Parameters monitored could include serum (blood) chemistry (BC), biochemical changes (BI), body weight (BW), clinical signs (CS), developmental toxicity (DX), food intake (FI), gross necropsy (GN), hematology (HE), histopathology (HP), immune function (IX), lethality (LE), neurological function (NX), organ function (OF), ophthalmology (OP), organ weight (OW), reproductive function (RX), urinalysis (UR), and water intake (WI).

(7) Endpoint. This column lists the endpoint examined. The major categories of health endpoints included in LSE tables and figures are death, body weight, respiratory, cardiovascular, gastrointestinal, hematological, musculoskeletal, hepatic, renal, dermal, ocular, endocrine, immunological, neurological, reproductive, developmental, other noncancer, and cancer. "Other noncancer" refers to any effect (e.g., alterations in blood glucose levels) not covered in these systems. In the example of key number 51, three endpoints (body weight, hematological, and hepatic) were investigated.

(8) NOAEL. A NOAEL is the highest exposure level at which no adverse effects were seen in the organ system studied. The body weight effect reported in key number 51 is a NOAEL at 25.5 mg/kg/day. NOAELs are not reported for cancer and death; with the exception of these two endpoints, this field is left blank if no NOAEL was identified in the study.

(9) LOAEL. A LOAEL is the lowest dose used in the study that caused an adverse health effect. LOAELs have been classified into "Less Serious" and "Serious" effects. These distinctions help readers identify the levels of exposure at which adverse health effects first appear and the gradation of effects with increasing dose. A brief description of the specific endpoint used to quantify the adverse effect accompanies the LOAEL. Key number 51 reports a less serious LOAEL of 6.1 mg/kg/day for the hepatic system, which was used to derive a chronic exposure, oral MRL of 0.008 mg/kg/day (see footnote "c"). MRLs are not derived from serious LOAELs. A cancer effect level (CEL) is the lowest exposure level associated with the onset of carcinogenesis in experimental or epidemiologic studies. CELs are always considered serious effects. The LSE tables and figures do not contain NOAELs for cancer, but the text may report doses not causing measurable cancer increases. If no LOAEL/CEL values were identified in the study, this field is left blank.

(10) Reference. The complete reference citation is provided in Chapter 8 of the profile.

(11) Footnotes. Explanations of abbreviations or reference notes for data in the LSE tables are found in the footnotes. For example, footnote "c" indicates that the LOAEL of 6.1 mg/kg/day in key number 51 was used to derive an oral MRL of 0.008 mg/kg/day.

FIGURE LEGEND

See Sample LSE Figure (page C-6)

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

(12) Exposure period. The same exposure periods appear as in the LSE table. In this example, health effects observed within the chronic exposure period are illustrated.

(13) Endpoint. These are the categories of health effects for which reliable quantitative data exist. The same health effect endpoints appear in the LSE table.

(14) Levels of exposure. Concentrations or doses for each health effect in the LSE tables are graphically displayed in the LSE figures. Exposure concentration or dose is measured on the log scale "y" axis. Inhalation exposure is reported in mg/m^3 or ppm and oral exposure is reported in mg/kg/day.

(15) LOAEL. In this example, the half-shaded circle that is designated 51R identifies a LOAEL critical endpoint in the rat upon which a chronic oral exposure MRL is based. The key number 51 corresponds to the entry in the LSE table. The dashed descending arrow indicates the extrapolation from the exposure level of 6.1 mg/kg/day (see entry 51 in the sample LSE table) to the MRL of 0.008 mg/kg/day (see footnote "c" in the sample LSE table).

(16) CEL. Key number 59R is one of studies for which CELs were derived. The diamond symbol refers to a CEL for the test species (rat). The number 59 corresponds to the entry in the LSE table.

(17) Key to LSE figure. The key provides the abbreviations and symbols used in the figure.

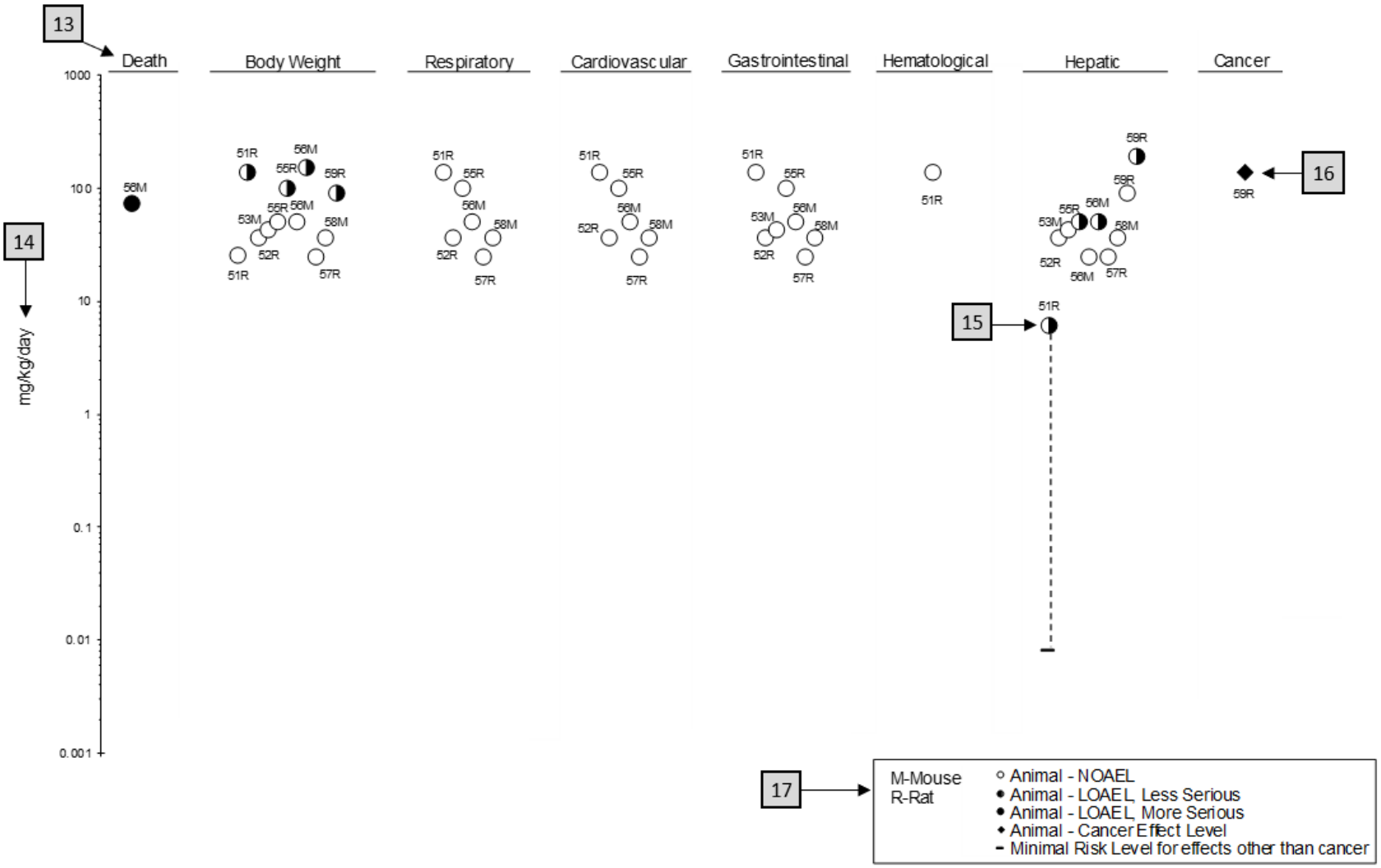
Table 2-X. Levels of Significant Exposure to [Chemical X] – Oral

Figure key[a]	Species (strain) No./group	Exposure parameters	Doses (mg/kg/day)	Parameters monitored	Endpoint	NOAEL (mg/kg/day)	Less serious LOAEL (mg/kg/day)	Serious LOAEL (mg/kg/day)	Effect
CHRONIC EXPOSURE									
51	Rat (Wistar) 40 M, 40 F	2 years (F)	M: 0, 6.1, 25.5, 138.0 F: 0, 8.0, 31.7, 168.4	CS, WI, BW, OW, HE, BC, HP	Bd wt	25.5	138.0		Decreased body weight gain in males (23–25%) and females (31–39%)
					Hemato	138.0			
					Hepatic		6.1[c]		Increases in absolute and relative weights at ≥6.1/8.0 mg/kg/day after 12 months of exposure; fatty generation at ≥6.1 mg/kg/day in males and at ≥31.7 mg/kg/day in females, and granulomas in females at 31.7 and 168.4 mg/kg/day after 12, 18, or 24 months of exposure and in males at ≥6.1 mg/kg/day only after 24 months of exposure
Aida et al. 1992									
52	Rat (F344) 78 M	104 weeks (W)	0, 3.9, 20.6, 36.3	CS, BW, FI, BC, OW, HP	Hepatic	36.3			
					Renal	20.6	36.3		Increased incidence of renal tubular cell hyperplasia
					Endocr	36.3			
George et al. 2002									
59	Rat (Wistar) 58M, 58F	Lifetime (W)	M: 0, 90 F: 0, 190	BW, HP	Cancer		190 F		Increased incidence of hepatic neoplastic nodules in females only; no additional description of the tumors was provided
Tumasonis et al. 1985									

[a]The number corresponds to entries in Figure 2-x.
[b]Used to derive an acute-duration oral minimal risk level (MRL) of 0.1 mg/kg/day based on the $BMDL_{05}$ of 10 mg/kg/day and an uncertainty factor of 100 (10 for extrapolation from animals to humans and 10 for human variability).
[c]Used to derive a chronic-duration oral MRL of 0.008 mg/kg/day based on the $BMDL_{10}$ of 0.78 mg/kg/day and an uncertainty factor of 100 (10 for extrapolation from animals to humans and 10 for human variability).

APPENDIX C

Figure 2-X. Levels of Significant Exposure to [Chemical X] - Oral



APPENDIX D. QUICK REFERENCE FOR HEALTH CARE PROVIDERS

Toxicological Profiles are a unique compilation of toxicological information on a given hazardous substance. Each profile reflects a comprehensive and extensive evaluation, summary, and interpretation of available toxicologic and epidemiologic information on a substance. Health care providers treating patients potentially exposed to hazardous substances may find the following information helpful for fast answers to often-asked questions.

Primary Chapters/Sections of Interest

Chapter 1: Relevance to Public Health: The Relevance to Public Health Section provides an overview of exposure and health effects and evaluates, interprets, and assesses the significance of toxicity data to human health. A table listing minimal risk levels (MRLs) is also included in this chapter.

Chapter 2: Health Effects: Specific health effects identified in both human and animal studies are reported by type of health effect (e.g., death, hepatic, renal, immune, reproductive), route of exposure (e.g., inhalation, oral, dermal), and length of exposure (e.g., acute, intermediate, and chronic).
NOTE: Not all health effects reported in this section are necessarily observed in the clinical setting.

Pediatrics:

Section 3.2 Children and Other Populations that are Unusually Susceptible
Section 3.3 Biomarkers of Exposure and Effect

ATSDR Information Center

Phone: 1-800-CDC-INFO (800-232-4636) or 1-888-232-6348 (TTY)
Internet: http://www.atsdr.cdc.gov

ATSDR develops educational and informational materials for health care providers categorized by hazardous substance, clinical condition, and/or by susceptible population. The following additional materials are available online:

Clinician Briefs discuss health effects and approaches to patient management in a brief/factsheet style. *Clinician Overviews* are narrated PowerPoint presentations with Continuing Education credit available (see https://www.atsdr.cdc.gov/emes/health_professionals/clinician-briefs-overviews.html).

Managing Hazardous Materials Incidents is a set of recommendations for on-scene (prehospital) and hospital medical management of patients exposed during a hazardous materials incident (see https://www.atsdr.cdc.gov/MHMI/index.html).

Fact Sheets (ToxFAQs™) provide answers to frequently asked questions about toxic substances (see https://www.atsdr.cdc.gov/toxfaqs/Index.asp).

Other Agencies and Organizations

The National Center for Environmental Health (NCEH) focuses on preventing or controlling disease, injury, and disability related to the interactions between people and their environment outside the workplace. Contact: NCEH, Mailstop F-29, 4770 Buford Highway, NE, Atlanta, GA 30341-3724 • Phone: 770-488-7000 • FAX: 770-488-7015 • Web Page: https://www.cdc.gov/nceh/.

The National Institute for Occupational Safety and Health (NIOSH) conducts research on occupational diseases and injuries, responds to requests for assistance by investigating problems of health and safety in the workplace, recommends standards to the Occupational Safety and Health Administration (OSHA) and the Mine Safety and Health Administration (MSHA), and trains professionals in occupational safety and health. Contact: NIOSH, 395 E Street, S.W., Suite 9200, Patriots Plaza Building, Washington, DC 20201 • Phone: 202-245-0625 or 1-800-CDC-INFO (800-232-4636) • Web Page: https://www.cdc.gov/niosh/.

The National Institute of Environmental Health Sciences (NIEHS) is the principal federal agency for biomedical research on the effects of chemical, physical, and biologic environmental agents on human health and well-being. Contact: NIEHS, PO Box 12233, 104 T.W. Alexander Drive, Research Triangle Park, NC 27709 • Phone: 919-541-3212 • Web Page: https://www.niehs.nih.gov/.

Clinical Resources (Publicly Available Information)

The Association of Occupational and Environmental Clinics (AOEC) has developed a network of clinics in the United States to provide expertise in occupational and environmental issues. Contact: AOEC, 1010 Vermont Avenue, NW, #513, Washington, DC 20005 • Phone: 202-347-4976 • FAX: 202-347-4950 • e-mail: AOEC@AOEC.ORG • Web Page: http://www.aoec.org/.

The American College of Occupational and Environmental Medicine (ACOEM) is an association of physicians and other health care providers specializing in the field of occupational and environmental medicine. Contact: ACOEM, 25 Northwest Point Boulevard, Suite 700, Elk Grove Village, IL 60007-1030 • Phone: 847-818-1800 • FAX: 847-818-9266 • Web Page: http://www.acoem.org/.

The American College of Medical Toxicology (ACMT) is a nonprofit association of physicians with recognized expertise in medical toxicology. Contact: ACMT, 10645 North Tatum Boulevard, Suite 200-111, Phoenix AZ 85028 • Phone: 844-226-8333 • FAX: 844-226-8333 • Web Page: http://www.acmt.net.

The Pediatric Environmental Health Specialty Units (PEHSUs) is an interconnected system of specialists who respond to questions from public health professionals, clinicians, policy makers, and the public about the impact of environmental factors on the health of children and reproductive-aged adults. Contact information for regional centers can be found at http://pehsu.net/findhelp.html.

The American Association of Poison Control Centers (AAPCC) provide support on the prevention and treatment of poison exposures. Contact: AAPCC, 515 King Street, Suite 510, Alexandria VA 22314 • Phone: 701-894-1858 • Poison Help Line: 1-800-222-1222 • Web Page: http://www.aapcc.org/.

APPENDIX E. GLOSSARY

Absorption—The process by which a substance crosses biological membranes and enters systemic circulation. Absorption can also refer to the taking up of liquids by solids, or of gases by solids or liquids.

Acute Exposure—Exposure to a chemical for a duration of ≤14 days, as specified in the Toxicological Profiles.

Adsorption—The adhesion in an extremely thin layer of molecules (as of gases, solutes, or liquids) to the surfaces of solid bodies or liquids with which they are in contact.

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

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

Benchmark Dose (BMD) or Benchmark Concentration (BMC)—is the dose/concentration corresponding to a specific response level estimate using a statistical dose-response model applied to either experimental toxicology or epidemiology data. For example, a BMD_{10} would be the dose corresponding to a 10% benchmark response (BMR). The BMD is determined by modeling the dose-response curve in the region of the dose-response relationship where biologically observable data are feasible. The BMDL or BMCL is the 95% lower confidence limit on the BMD or BMC.

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

Biomarkers—Indicators signaling events in biologic systems or samples, typically classified as markers of exposure, effect, and susceptibility.

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

Carcinogen—A chemical capable of inducing cancer.

Case-Control Study—A type of epidemiological study that examines the relationship between a particular outcome (disease or condition) and a variety of potential causative agents (such as toxic chemicals). In a case-control study, a group of people with a specified and well-defined outcome is identified and compared to a similar group of people without the outcome.

Case Report—A report that describes a single individual with a particular disease or exposure. These reports may suggest some potential topics for scientific research, but are not actual research studies.

Case Series—Reports that describe the experience of a small number of individuals with the same disease or exposure. These reports may suggest potential topics for scientific research, but are not actual research studies.

Ceiling Value—A concentration that must not be exceeded.

Chronic Exposure—Exposure to a chemical for ≥365 days, as specified in the Toxicological Profiles.

Clastogen—A substance that causes breaks in chromosomes resulting in addition, deletion, or rearrangement of parts of the chromosome.

Cohort Study—A type of epidemiological study of a specific group or groups of people who have had a common insult (e.g., exposure to an agent suspected of causing disease or a common disease) and are followed forward from exposure to outcome, and who are disease-free at start of follow-up. Often, at least one exposed group is compared to one unexposed group, while in other cohorts, exposure is a continuous variable and analyses are directed towards analyzing an exposure-response coefficient.

Cross-sectional Study—A type of epidemiological study of a group or groups of people that examines the relationship between exposure and outcome to a chemical or to chemicals at a specific point in time.

Data Needs—Substance-specific informational needs that, if met, would reduce the uncertainties of human health risk assessment.

Developmental Toxicity—The occurrence of adverse effects on the developing organism that may result from exposure to a chemical prior to conception (either parent), during prenatal development, or postnatally to the time of sexual maturation. Adverse developmental effects may be detected at any point in the life span of the organism.

Dose-Response Relationship—The quantitative relationship between the amount of exposure to a toxicant and the incidence of the response or amount of the response.

Embryotoxicity and Fetotoxicity—Any toxic effect on the conceptus as a result of prenatal exposure to a chemical; the distinguishing feature between the two terms is the stage of development during which the effect occurs. Effects include malformations and variations, altered growth, and *in utero* death.

Epidemiology—The investigation of factors that determine the frequency and distribution of disease or other health-related conditions within a defined human population during a specified period.

Excretion—The process by which metabolic waste products are removed from the body.

Genotoxicity—A specific adverse effect on the genome of living cells that, upon the duplication of affected cells, can be expressed as a mutagenic, clastogenic, or carcinogenic event because of specific alteration of the molecular structure of the genome.

Half-life—A measure of rate for the time required to eliminate one-half of a quantity of a chemical from the body or environmental media.

Health Advisory—An estimate of acceptable drinking water levels for a chemical substance derived by EPA and based on health effects information. A health advisory is not a legally enforceable federal standard, but serves as technical guidance to assist federal, state, and local officials.

Immediately Dangerous to Life or Health (IDLH)—A condition that poses a threat of life or health, or conditions that pose an immediate threat of severe exposure to contaminants that are likely to have adverse cumulative or delayed effects on health.

Immunotoxicity—Adverse effect on the functioning of the immune system that may result from exposure to chemical substances.

Incidence—The ratio of new cases of individuals in a population who develop a specified condition to the total number of individuals in that population who could have developed that condition in a specified time period.

Intermediate Exposure—Exposure to a chemical for a duration of 15–364 days, as specified in the Toxicological Profiles.

***In Vitro*—**Isolated from the living organism and artificially maintained, as in a test tube.

***In Vivo*—**Occurring within the living organism.

Lethal Concentration$_{(LO)}$ (LC_{LO})—The lowest concentration of a chemical in air that has been reported to have caused death in humans or animals.

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

Lethal Dose$_{(LO)}$ (LD_{Lo})—The lowest dose of a chemical introduced by a route other than inhalation that has been reported to have caused death in humans or animals.

Lethal Dose$_{(50)}$ (LD_{50})—The dose of a chemical that has been calculated to cause death in 50% of a defined experimental animal population.

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

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

Lymphoreticular Effects—Represent morphological effects involving lymphatic tissues such as the lymph nodes, spleen, and thymus.

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

Metabolism—Process in which chemical substances are biotransformed in the body that could result in less toxic and/or readily excreted compounds or produce a biologically active intermediate.

Minimal Risk Level (MRL)—An estimate of daily human exposure to a hazardous substance that is likely to be without an appreciable risk of adverse noncancer health effects over a specified route and duration of exposure.

Modifying Factor (MF)—A value (greater than zero) that is applied to the derivation of a Minimal Risk Level (MRL) to reflect additional concerns about the database that are not covered by the uncertainty factors. The default value for a MF is 1.

Morbidity—The state of being diseased; the morbidity rate is the incidence or prevalence of a disease in a specific population.

Mortality—Death; the mortality rate is a measure of the number of deaths in a population during a specified interval of time.

Mutagen—A substance that causes mutations, which are changes in the DNA sequence of a cell's DNA. Mutations can lead to birth defects, miscarriages, or cancer.

Necropsy—The gross examination of the organs and tissues of a dead body to determine the cause of death or pathological conditions.

Neurotoxicity—The occurrence of adverse effects on the nervous system following exposure to a hazardous substance.

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

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

Odds Ratio (OR)—A means of measuring the association between an exposure (such as toxic substances and a disease or condition) that represents the best estimate of relative risk (risk as a ratio of the incidence among subjects exposed to a particular risk factor divided by the incidence among subjects who were not exposed to the risk factor). An odds ratio that is greater than 1 is considered to indicate greater risk of disease in the exposed group compared to the unexposed group.

Permissible Exposure Limit (PEL)—An Occupational Safety and Health Administration (OSHA) regulatory limit on the amount or concentration of a substance not to be exceeded in workplace air averaged over any 8-hour work shift of a 40-hour workweek.

Pesticide—General classification of chemicals specifically developed and produced for use in the control of agricultural and public health pests (insects or other organisms harmful to cultivated plants or animals).

Pharmacokinetics—The dynamic behavior of a material in the body, used to predict the fate (disposition) of an exogenous substance in an organism. Utilizing computational techniques, it provides the means of studying the absorption, distribution, metabolism, and excretion of chemicals by the body.

Pharmacokinetic Model—A set of equations that can be used to describe the time course of a parent chemical or metabolite in an animal system. There are two types of pharmacokinetic models: data-based and physiologically-based. A data-based model divides the animal system into a series of compartments, which, in general, do not represent real, identifiable anatomic regions of the body, whereas the physiologically-based model compartments represent real anatomic regions of the body.

Physiologically Based Pharmacodynamic (PBPD) Model—A type of physiologically based dose-response model that quantitatively describes the relationship between target tissue dose and toxic endpoints. These models advance the importance of physiologically based models in that they clearly describe the biological effect (response) produced by the system following exposure to an exogenous substance.

Physiologically Based Pharmacokinetic (PBPK) Model—A type of physiologically based dose-response model that is comprised of a series of compartments representing organs or tissue groups with realistic weights and blood flows. These models require a variety of physiological information, including tissue volumes, blood flow rates to tissues, cardiac output, alveolar ventilation rates, and possibly membrane permeabilities. The models also utilize biochemical information, such as blood:air partition coefficients, and metabolic parameters. PBPK models are also called biologically based tissue dosimetry models.

Prevalence—The number of cases of a disease or condition in a population at one point in time.

Prospective Study—A type of cohort study in which a group is followed over time and the pertinent observations are made on events occurring after the start of the study.

Recommended Exposure Limit (REL)—A National Institute for Occupational Safety and Health (NIOSH) time-weighted average (TWA) concentration for up to a 10-hour workday during a 40-hour workweek.

Reference Concentration (RfC)—An estimate (with uncertainty spanning perhaps an order of magnitude) of a continuous inhalation exposure to the human population (including sensitive subgroups) that is likely to be without an appreciable risk of deleterious noncancer health effects during a lifetime. The inhalation RfC is expressed in units of mg/m^3 or ppm.

Reference Dose (RfD)—An estimate (with uncertainty spanning perhaps an order of magnitude) of the daily oral exposure of the human population to a potential hazard that is likely to be without risk of deleterious noncancer health effects during a lifetime. The oral RfD is expressed in units of mg/kg/day.

Reportable Quantity (RQ)—The quantity of a hazardous substance that is considered reportable under the Comprehensive Environmental Response, Compensation, and Liability Act (CERCLA). RQs are (1) ≥1 pound or (2) for selected substances, an amount established by regulation either under CERCLA or under Section 311 of the Clean Water Act. Quantities are measured over a 24-hour period.

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

Retrospective Study—A type of cohort study based on a group of persons known to have been exposed at some time in the past. Data are collected from routinely recorded events, up to the time the study is undertaken. Retrospective studies are limited to causal factors that can be ascertained from existing records and/or examining survivors of the cohort.

Risk—The possibility or chance that some adverse effect will result from a given exposure to a hazardous substance.

Risk Factor—An aspect of personal behavior or lifestyle, an environmental exposure, existing health condition, or an inborn or inherited characteristic that is associated with an increased occurrence of disease or other health-related event or condition.

Risk Ratio/Relative Risk—The ratio of the risk among persons with specific risk factors compared to the risk among persons without risk factors. A risk ratio that is greater than 1 indicates greater risk of disease in the exposed group compared to the unexposed group.

Short-Term Exposure Limit (STEL)—A STEL is a 15-minute TWA exposure that should not be exceeded at any time during a workday.

Standardized Mortality Ratio (SMR)—A ratio of the observed number of deaths and the expected number of deaths in a specific standard population.

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

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

Threshold Limit Value (TLV)—An American Conference of Governmental Industrial Hygienists (ACGIH) concentration of a substance to which it is believed that nearly all workers may be repeatedly exposed, day after day, for a working lifetime without adverse effect. The TLV may be expressed as a Time-Weighted Average (TLV-TWA), as a Short-Term Exposure Limit (TLV-STEL), or as a ceiling limit (TLV-C).

Time-Weighted Average (TWA)—An average exposure within a given time period.

Toxicokinetic—The absorption, distribution, metabolism, and elimination of toxic compounds in the living organism.

Toxics Release Inventory (TRI)—The TRI is an EPA program that tracks toxic chemical releases and pollution prevention activities reported by industrial and federal facilities.

Uncertainty Factor (UF)—A factor used in operationally deriving the Minimal Risk Level (MRL), Reference Dose (RfD), or Reference Concentration (RfC) from experimental data. UFs are intended to account for (1) the variation in sensitivity among the members of the human population, (2) the uncertainty in extrapolating animal data to the case of human, (3) the uncertainty in extrapolating from data obtained in a study that is of less than lifetime exposure, and (4) the uncertainty in using lowest-observed-adverse-effect level (LOAEL) data rather than no-observed-adverse-effect level (NOAEL) data. A default for each individual UF is 10; if complete certainty in data exists, a value of 1 can be used; however, a reduced UF of 3 may be used on a case-by-case basis (3 being the approximate logarithmic average of 10 and 1).

Xenobiotic—Any substance that is foreign to the biological system.

APPENDIX F. ACRONYMS, ABBREVIATIONS, AND SYMBOLS

AAPCC	American Association of Poison Control Centers
ACGIH	American Conference of Governmental Industrial Hygienists
ACOEM	American College of Occupational and Environmental Medicine
ACMT	American College of Medical Toxicology
ADI	acceptable daily intake
ADME	absorption, distribution, metabolism, and excretion
AEGL	Acute Exposure Guideline Level
AIC	Akaike's information criterion
AIHA	American Industrial Hygiene Association
ALT	alanine aminotransferase
AOEC	Association of Occupational and Environmental Clinics
AP	alkaline phosphatase
AST	aspartate aminotransferase
atm	atmosphere
ATSDR	Agency for Toxic Substances and Disease Registry
AWQC	Ambient Water Quality Criteria
BCF	bioconcentration factor
BMD/C	benchmark dose or benchmark concentration
BMD_X	dose that produces a X% change in response rate of an adverse effect
$BMDL_X$	95% lower confidence limit on the BMD_X
BMDS	Benchmark Dose Software
BMR	benchmark response
BUN	blood urea nitrogen
C	centigrade
CAA	Clean Air Act
CAS	Chemical Abstract Services
CDC	Centers for Disease Control and Prevention
CEL	cancer effect level
CERCLA	Comprehensive Environmental Response, Compensation, and Liability Act
CFR	Code of Federal Regulations
Ci	curie
CI	confidence interval
cm	centimeter
CPSC	Consumer Products Safety Commission
CWA	Clean Water Act
DNA	deoxyribonucleic acid
DOD	Department of Defense
DOE	Department of Energy
DWEL	drinking water exposure level
EAFUS	Everything Added to Food in the United States
ECG/EKG	electrocardiogram
EEG	electroencephalogram
EPA	Environmental Protection Agency
ERPG	emergency response planning guidelines
F	Fahrenheit
F1	first-filial generation
FDA	Food and Drug Administration
FIFRA	Federal Insecticide, Fungicide, and Rodenticide Act
FR	Federal Register

FSH	follicle stimulating hormone
g	gram
GC	gas chromatography
gd	gestational day
GGT	γ-glutamyl transferase
GRAS	generally recognized as safe
HEC	human equivalent concentration
HED	human equivalent dose
HHS	Department of Health and Human Services
HPLC	high-performance liquid chromatography
HSDB	Hazardous Substance Data Bank
IARC	International Agency for Research on Cancer
IDLH	immediately dangerous to life and health
IRIS	Integrated Risk Information System
Kd	adsorption ratio
kg	kilogram
kkg	kilokilogram; 1 kilokilogram is equivalent to 1,000 kilograms and 1 metric ton
K_{oc}	organic carbon partition coefficient
K_{ow}	octanol-water partition coefficient
L	liter
LC	liquid chromatography
LC_{50}	lethal concentration, 50% kill
LC_{Lo}	lethal concentration, low
LD_{50}	lethal dose, 50% kill
LD_{Lo}	lethal dose, low
LDH	lactate dehydrogenase
LH	luteinizing hormone
LOAEL	lowest-observed-adverse-effect level
LSE	Level of Significant Exposure
LT_{50}	lethal time, 50% kill
m	meter
mCi	millicurie
MCL	maximum contaminant level
MCLG	maximum contaminant level goal
MF	modifying factor
mg	milligram
mL	milliliter
mm	millimeter
mmHg	millimeters of mercury
mmol	millimole
MRL	Minimal Risk Level
MS	mass spectrometry
MSHA	Mine Safety and Health Administration
Mt	metric ton
NAAQS	National Ambient Air Quality Standard
NAS	National Academy of Science
NCEH	National Center for Environmental Health
ND	not detected
ng	nanogram
NHANES	National Health and Nutrition Examination Survey
NIEHS	National Institute of Environmental Health Sciences

NIOSH	National Institute for Occupational Safety and Health
NLM	National Library of Medicine
nm	nanometer
nmol	nanomole
NOAEL	no-observed-adverse-effect level
NPL	National Priorities List
NR	not reported
NRC	National Research Council
NS	not specified
NTP	National Toxicology Program
OR	odds ratio
OSHA	Occupational Safety and Health Administration
PAC	Protective Action Criteria
PAH	polycyclic aromatic hydrocarbon
PBPD	physiologically based pharmacodynamic
PBPK	physiologically based pharmacokinetic
PEHSU	Pediatric Environmental Health Specialty Unit
PEL	permissible exposure limit
PEL-C	permissible exposure limit-ceiling value
pg	picogram
PND	postnatal day
POD	point of departure
ppb	parts per billion
ppbv	parts per billion by volume
ppm	parts per million
ppt	parts per trillion
REL	recommended exposure limit
REL-C	recommended exposure limit-ceiling value
RfC	reference concentration
RfD	reference dose
RNA	ribonucleic acid
SARA	Superfund Amendments and Reauthorization Act
SCE	sister chromatid exchange
SD	standard deviation
SE	standard error
SGOT	serum glutamic oxaloacetic transaminase (same as aspartate aminotransferase or AST)
SGPT	serum glutamic pyruvic transaminase (same as alanine aminotransferase or ALT)
SIC	standard industrial classification
SLOAEL	serious lowest-observed-adverse-effect level
SMR	standardized mortality ratio
sRBC	sheep red blood cell
STEL	short term exposure limit
TLV	threshold limit value
TLV-C	threshold limit value-ceiling value
TRI	Toxics Release Inventory
TSCA	Toxic Substances Control Act
TWA	time-weighted average
UF	uncertainty factor
U.S.	United States
USDA	United States Department of Agriculture
USGS	United States Geological Survey

USNRC	U.S. Nuclear Regulatory Commission
VOC	volatile organic compound
WBC	white blood cell
WHO	World Health Organization

>	greater than
≥	greater than or equal to
=	equal to
<	less than
≤	less than or equal to
%	percent
α	alpha
β	beta
γ	gamma
δ	delta
μm	micrometer
μg	microgram
q_1^*	cancer slope factor
–	negative
+	positive
(+)	weakly positive result
(–)	weakly negative result