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. LOAELs for serious health effects (such as irreparable damage to the liver or kidneys, or serious 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.

HCH is a mixture of eight isomers, four of which are of commercial significance: α-HCH (CAS Registry Number 319-84-6), β-HCH (CAS Registry Number 319-85-7), γ-HCH (CAS Registry Number 58-89-9), and δ-HCH (CAS Registry Number 319-86-8). Technical HCH, which is used as an insecticide, is made up of the various isomers at different concentrations. The wide variations in isomer composition of technical HCH preclude the possibility of MRL derivation. MRL derivation was considered for the isomers included in this toxicological profile: α-, β-, γ-, and δ-HCH.

APPENDIX A

MINIMAL RISK LEVEL (MRL) WORKSHEET

***Chemical Name*:** α-HCH
CAS Number: 319-84-6
Date: March 2024
Profile Status: Final
Route: Inhalation
Duration: Acute

MRL Summary: There are insufficient data for derivation of an acute-duration inhalation MRL for α-HCH.

Rationale for Not Deriving an MRL: No acute-duration inhalation studies of α-HCH in humans or animals were located, precluding derivation of an acute-duration inhalation MRL.

Agency Contacts (Chemical Managers): Malcolm Williams

MINIMAL RISK LEVEL (MRL) WORKSHEET

***Chemical Name*:** α-HCH
CAS Number: 319-84-6
Date: March 2024
Profile Status: Final
Route: Inhalation
Duration: Intermediate

MRL Summary: There are insufficient data for derivation of an intermediate-duration inhalation MRL for α-HCH.

Rationale for Not Deriving an MRL: No intermediate-duration inhalation studies of α-HCH in humans or animals were located, precluding derivation of an intermediate-duration inhalation MRL.

Agency Contacts (Chemical Managers): Malcolm Williams

APPENDIX A

MINIMAL RISK LEVEL (MRL) WORKSHEET

***Chemical Name*:** α-HCH
CAS Number: 319-84-6
Date: March 2024
Profile Status: Final
Route: Inhalation
Duration: Chronic

MRL Summary: There are insufficient data for derivation of a chronic-duration inhalation MRL for α-HCH.

Rationale for Not Deriving an MRL: No chronic-duration inhalation studies of α-HCH in humans or animals were located, precluding derivation of a chronic-duration inhalation MRL.

Agency Contacts (Chemical Managers): Malcolm Williams

APPENDIX A

MINIMAL RISK LEVEL (MRL) WORKSHEET

***Chemical Name*:** α-HCH
CAS Number: 319-84-6
Date: March 2024
Profile Status: Final
Route: Oral
Duration: Acute

MRL Summary: There are insufficient data for derivation of an acute-duration oral MRL for α-HCH.

Rationale for Not Deriving an MRL: No adequate exposure-response data were available for humans. Data on effects in laboratory animals exposed orally to α-HCH for acute durations are limited to a freestanding NOAEL of 20 mg/kg/day for body weight changes during the first 14 days of a 28-day study comparing gene expression profiles for α- and γ-HCH (Sumida et al. 2007). Increased relative liver weight (24%) and decreased serum ALP (19%) were seen at this dose after 14 days, but histopathology was not evaluated at this time, so reliable hepatic effect levels could not be determined for this study. Thus, this study did not provide an adequate basis for MRL derivation.

Agency Contacts (Chemical Managers): Malcolm Williams

APPENDIX A

MINIMAL RISK LEVEL (MRL) WORKSHEET

***Chemical Name*:** α-HCH
CAS Number: 319-84-6
Date: March 2024
Profile Status: Final
Route: Oral
Duration: Intermediate
MRL: 0.002 mg/kg/day (2 µg/kg/day)
Critical Effect: Increased liver weight and liver histopathology changes
Reference: Sumida et al. 2007
Point of Departure: 2 mg/kg/day (NOAEL)
Uncertainty Factor: 100
Modifying Factor: 10
LSE Graph Key: 6
Species: Rat

MRL Summary: An intermediate-duration oral MRL of 0.002 mg/kg/day (2 µg/kg/day) was derived based on a NOAEL of 2 mg/kg/day for liver effects in a 28-day study in rats (Sumida et al. 2007). The NOAEL was divided by a total uncertainty factor of 100 (10 for human variability and 10 for animal to human extrapolation) and a modifying factor of 10 (for lack of studies examining developmental and immunological effects and limitations in available data on neurotoxicity).

Selection of the Critical Effect: No dose-response data are available for humans. In animal studies, hepatic effects (including cancers) occurred at lower doses than effects on body weight or kidneys. A single study found no effects on motor nerve conduction velocity in rats exposed to 106.2 mg/kg/day for 30 days (Muller et al. 1981). The α-hexachlorocyclohexane database only contains studies with body weight, renal, hepatic, neurological, and cancer endpoints; no other effects were evaluated in the available studies. Table A-1 summarizes the hepatic effects from intermediate-duration oral studies in laboratory animals exposed to doses up to 70 mg/kg/day.

Table A-1. Summary of NOAELs and LOAELs from Candidate Intermediate-Duration Studies in Laboratory Animals Orally Exposed to α-Hexachlorocyclohexane

Species	Exposure scenario	NOAEL (mg/kg/day)	LOAEL (mg/kg/day)	Effect	Reference
Hepatic effects					
Mouse (dd) 20–40 M	24 weeks (F)	ND	18	33% relative liver weight increase; hepatocellular hypertrophy	Ito et al. 1973
Rat (Fischer-344) 4 M	28 days (GO)	2	20	Increased relative liver weight (25%); centrilobular hepatocellular hypertrophy	Sumida et al. 2007
Rat (W strain) 18–24 M	48 weeks (F)	ND	35	Hepatocellular hypertrophy	Ito et al. 1975
Rat (Wistar) 8 M	24 weeks (F)	ND	45	Mild liver cell hypertrophy; 2-fold increase in liver weight	Nagasaki et al. 1975

Table A-1. Summary of NOAELs and LOAELs from Candidate Intermediate-Duration Studies in Laboratory Animals Orally Exposed to α-Hexachlorocyclohexane

Species	Exposure scenario	NOAEL (mg/kg/day)	LOAEL (mg/kg/day)	Effect	Reference
Hamster (Golden Syrian) 6–10 M	24 weeks (F)	ND	45	20–38% increase in liver weight; liver cell hypertrophy	Nagasaki et al. 1975
Rat (Wistar) 10 F, 10 M	6–9 months (F)	ND	60 M 70 F (serious LOAEL for decreased survival)	Decreased survival; moderate histopathology changes (focal necrosis, fatty degeneration); >2-fold increase in liver weight	Fitzhugh et al. 1950

Principal study for the MRL.

(F) = feed; F = female(s); (GO) = gavage in oil vehicle; LOAEL = lowest-observed-adverse-effect level; M = male(s); ND = not determined; NOAEL = no-observed-adverse-effect level

Selection of the Principal Study: As Table A-1 shows, the lowest effect level (18 mg/kg/day) was associated with a relative liver weight increase of 33% and hepatocellular hypertrophy in mice exposed via diet for 24 weeks (Ito et al. 1973). A NOAEL was not identified for this study. In a study of rats exposed by gavage for only 28 days (Sumida et al. 2007), a slightly higher LOAEL of 20 mg/kg/day was identified for similar hepatic effects; the NOAEL in that study was 2 mg/kg/day. All of the other studies identified hepatic effects at the lowest doses tested (no other NOAEL was identified). The study by Sumida et al. (2007) was selected as the principal study for MRL derivation because the NOAEL identified in the study was lower than the LOAELs identified in all other studies and because a NOAEL was not identified in the study by Ito et al. (1973). Although the LOAEL (18 mg/kg/day) from Ito et al. (1973) is slightly lower than the LOAEL (20 mg/kg/day) identified by Sumida et al. (2007), use of the LOAEL from Ito et al. (1973) as the point of departure (POD) would necessitate the use of an uncertainty factor of 10, while use of the NOAEL (2 mg/kg/day) from Sumida et al. (2007) does not. In addition, if the LOAEL from Ito et al. (1973) were used as the POD, it would be equivalent to the value derived based on the NOAEL from Sumida et al. (2007) (18 mg/kg/day divided by an uncertainty factor of 10 for use of a LOAEL yields 1.8 mg/kg/day, which rounds to 2 mg/kg/day).

Summary of the Principal Study:

Sumida K, Siato K, Oeda K, et al. 2007. A comparative study of gene expression profiles in rat liver after administration of α-hexachlorocyclohexane and lindane. J Toxicol Sci 32(3):261-288.

Groups of four male F344 rats were administered α-HCH (99% purity, in corn oil) by gavage at doses of 0 (corn oil control), 2, or 20 mg/kg/day for 28 consecutive days. The following parameters were evaluated: body weight, serum clinical chemistry (AST, ALT, ALP, and total bilirubin in blood collected at necropsy), liver weight, hepatic histopathology, and gene expression in the liver. There were no effects on body weight at any dose. After 28 days of exposure, small but statistically significant increases in AST and ALT were seen at 2 mg/kg/day, but not at 20 mg/kg/day. At the high dose, ALP was decreased by 19% at the end of 28 days. At 20 mg/kg/day, liver weights were increased by 20% (absolute) and 25% (relative) compared to controls. At 2 mg/kg/day, relative liver weight was increased by a small, but

statistically significant margin of 6%. Hepatocellular hypertrophy was observed in 4/4 animals at 20 mg/kg/day compared to 0/4 in control and 2 mg/kg/day groups. Based on the increase in liver weight and histological changes at 20 mg/kg/day, this dose is a LOAEL. No exposure-related changes occurred at the 2 mg/kg/day, indicating that this dose is a NOAEL.

Selection of the Point of Departure for the MRL: The NOAEL of 2 mg/kg/day was selected as the POD for derivation of the intermediate-duration oral MRL for α-HCH. Although quantitative data on histopathology findings in the liver were reported, the incidence data increased from 0/4 at 2 mg/kg/day to 4/4 at 20 mg/kg/day, so the data were not amenable to benchmark dose (BMD) modeling. BMD modeling of relative liver weight data was undertaken, but that dataset was also not amenable to BMD modeling, as neither the constant nor nonconstant variance models provide an adequate fit to the variance data. Thus, the NOAEL was selected as the POD.

Adjustment for Intermittent Exposure: Not applicable.

Uncertainty and Modifying Factor: The NOAEL of 2 mg/kg/day was divided by a total uncertainty factor (UF) of 100 and a modifying factor (MF) of 10:

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

A modifying factor of 10 was applied to the NOAEL to account for lack of data on developmental toxicity and immunotoxicity and limitations in available data on neurotoxicity. These are sensitive endpoints for other HCH isomers.

MRL = NOAEL ÷ (UF x MF)
2 mg/kg/day ÷ ((10 x 10) x 10) = 0.002 mg/kg/day

Other Additional Studies or Pertinent Information that Lend Support to this MRL: Hepatic effects have been observed in rats, mice, and hamsters after intermediate- and chronic-duration oral exposures to α-HCH. Observed effects include increases in absolute and relative liver weight, hepatocellular hypertrophy and/or hyperplasia, focal necrosis, fatty degeneration, hepatomegaly, bile duct proliferation, oval cells, nodular hyperplasia, and megalocytosis (Fitzhugh et al. 1950; Ito et al. 1973, 1975, 1976; Nagasaki et al. 1975; Sumida et al. 2007; Tryphonas and Iverson 1983). Both rats and mice have also developed liver tumors at higher doses of α-HCH (Hanada et al. 1973; Ito et al. 1973, 1975, 1976; Nagasaki et al. 1975; Tryphonas and Iverson 1983; Tsukada et al. 1979).

There are no studies on the effects of α-HCH on the developing organism or on the immune system. Studies of β- and γ-HCH have shown developmental effects (e.g., Di Consiglio et al. 2009; Hassoun and Stohs 1996a; La Sala et al. 2009; Rivera et al. 1998; Sauviat et al. 2005; Srinivasan et al. 1991), and for γ-HCH, these are the effects occurring at the lowest doses in animal studies. γ-HCH, and to a lesser extent β-HCH, have been demonstrated to induce suppression of the immune system in several species (e.g., Banerjee et al. 1996; Cornacoff et al. 1988; Desi et al. 1978; Dewan et al. 1980; Hong and Boorman 1993; Khurana et al. 1999; Koner et al. 1998; Mediratta et al. 2008; Meera et al. 1992; Van Velsen et al. 1986). Animal studies of γ-HCH indicate that these effects occur at lower doses than hepatic effects. Thus, the lack of developmental and immunotoxicity data for α-HCH is a significant limitation of the existing database for this isomer.

Data on the neurotoxicity of α-HCH are limited to a single study showing no change in motor nerve conduction velocity after 30 days of exposure (Muller et al. 1981). Both β- and γ-HCH have induced neurotoxic effects in laboratory rodents (e.g., Cornacoff et al. 1988; EPA 1999a; Gilbert and Mack 1995;

Parmar et al. 2003; Van Velsen et al. 1986), and γ-HCH has been shown to induce neurotoxicity in humans exposed orally (e.g., Davies et al. 1983; Harris et al. 1969; Munk and Nantel 1977; Nordt and Chew 2000; Powell 1980; Starr and Clifford 1972; Storen 1955).

Agency Contacts (Chemical Managers): Malcolm Williams

APPENDIX A

MINIMAL RISK LEVEL (MRL) WORKSHEET

***Chemical Name*:** α-HCH
CAS Number: 319-84-6
Date: March 2024
Profile Status: Final
Route: Oral
Duration: Chronic
MRL: 0.0009 mg/kg/day (0.9 µg/kg/day)
Critical Effect: Increased liver weight and liver histopathology changes
Reference: Fitzhugh et al. 1950
Point of Departure: 0.9 mg/kg/day (NOAEL)
Uncertainty Factor: 100
Modifying Factor: 10
LSE Graph Key: 14
Species: Rat

MRL Summary: A chronic-duration oral MRL of 0.0009 mg/kg/day (0.9 µg/kg/day) was derived for α-HCH based on a NOAEL of 0.9 mg/kg/day and a LOAEL of 4 mg/kg/day for increased liver weight and liver histopathology changes in rats exposed to α-HCH in the diet for 107 weeks (Fitzhugh et al. 1950). A total uncertainty factor of 100 (10 for extrapolation from animals to humans and 10 for human variability) and a modifying factor of 10 (for lack of immunotoxicity data and limitations in data on potential neurotoxicity) were applied to the NOAEL to derive the MRL.

Selection of the Critical Effect: Two chronic-duration oral studies of α-HCH were located: Fitzhugh et al. (1950) and Ito et al. (1975). While Ito et al. (1975) examined only cancer endpoints, Fitzhugh et al. (1950) evaluated histopathology in a large number of organs and identified the liver as the most sensitive target of chronic-duration oral exposure.

Selection of the Principal Study: Only Fitzhugh et al. (1950) evaluated endpoints other than cancer, so this study was selected for use in derivation of the chronic-duration oral MRL.

Summary of the Principal Study:

Fitzhugh OG, Nelson AA, Frawley JP. 1950. The chronic toxicities of technical benzene hexachloride and its α, β, and γ isomers. J Pharmacol Exp Ther 100:59-66.

Groups of 10 male and 10 female Wistar rats were treated with 0, 10, 50, 100, or 800 ppm α-HCH in food for life. ATSDR calculated doses corresponding to these concentrations using default food intake and body weight values for male and female Wistar rats in chronic studies as reported in EPA (1988b). Estimated doses were 0, 0.7, 4, 7, or 60 mg/kg/day in males and 0, 0.9, 4, 9, or 70 mg/kg/day in females. The lifetime of the animals sacrificed at the end of the experiment was taken as 107 weeks. Endpoints included clinical signs, body weight, food consumption, organ weights (liver, kidney, and spleen), gross pathology, and histopathology (lung, heart, liver, spleen, pancreas, stomach, small intestine, colon, kidney, adrenal, thyroid, leg muscles and bones, bone marrow, and testis or uterus and ovary). The numbers of animals per group evaluated for histopathology were 10, 8, 14, and 10 in the control through 100 ppm groups.

Survival was significantly reduced at the high dose (mean survival 35.9 weeks at 800 ppm [60–70 mg/kg/day] versus 58.3 weeks in controls), so effects in this group are considered to reflect intermediate-duration exposure. The mean age at death in the remaining groups did not differ from

controls (58.3, 54.6, 54.9, and 56.2 weeks for control through 100 ppm groups, respectively). Body weight gain through the first 6 months on study was tabulated for the 100 ppm (7–9 mg/kg/day) and 800 ppm (60–70 mg/kg/day) groups, but not for the 10 or 50 ppm groups. However, the text of the publication indicated that "lower experimental dosage levels had no effect on growth." Additionally, there was no significant difference in body weight gain in the 100 ppm (7–9 mg/kg/day) group, and no effect on food consumption in any group.

Histopathology findings were reported for the kidney, testes, and liver. Kidney pathological effects were not observed in groups receiving 10, 50, or 100 ppm, with the exception of slight brown pigmentation of the convoluted tubular epithelium at 100 ppm (7–9 mg/kg/day). The 800 ppm (60–70 mg/kg/day) group had slight to moderate kidney damage primarily consisting of tubular dilatation and/or atrophy, glomerular fibrosis and/or atrophy, and interstitial cell infiltration. The study authors reported a "questionable" increase in the degree of testicular atrophy in the group exposed to 800 ppm (60–70 mg/kg/day) α-HCH, but no further information was provided.

Significant increases in relative liver weight (both sexes grouped for analysis) were seen at 50 ppm (32%) and 100 ppm (44%). Gross and microscopic pathology findings were limited to the liver in the groups exposed for the full duration; there were no microscopic changes in the controls. Liver histopathology findings in treated animals were described qualitatively as very slight histological changes at 50 ppm (4 mg/kg/day) and slight histological changes at 100 ppm (7–9 mg/kg/day). The lesions were described as "characteristic of certain chlorinated cyclic compounds" with citation to earlier studies of dichloro-diphenyltrichloroethane (DDT). The earlier studies (e.g., Fitzhugh and Nelson 1947; Laug et al. 1950) characterized the histological changes as primarily centrilobular hepatocellular hypertrophy with increased cytoplasmic "oxyphilia" of these cells along with basophilia and margination of cytoplasmic granules and hyalinization of cytoplasm. There was evidence of increased severity in the group exposed to 800 ppm and surviving less than a year; these animals exhibited moderate histological damage including hepatic cell enlargement or atrophy, fatty degeneration, and focal necrosis.

Based on the increase in liver weight and histological changes at 50 ppm, this dose (4 mg/kg/day) is a LOAEL. No exposure-related changes occurred at the low dose in either sex, indicating that the NOAEL is 10 ppm (0.7–0.9 mg/kg/day).

Selection of the Point of Departure for the MRL: The NOAEL of 0.9 mg/kg/day (for females) was selected as the POD for MRL derivation (ATSDR policy is to select the highest NOAEL associated with the lowest LOAEL for the POD). BMD modeling of the liver weight data was not possible because the study did not report the numbers of animals per group evaluated for liver weights. Liver histology findings were reported qualitatively and without incidences, so BMD modeling was not feasible for these effects.

Adjustment for Intermittent Exposure: Not applicable.

Uncertainty and Modifying Factor: The NOAEL of 0.9 mg/kg/day was divided by a total uncertainty factor (UF) of 100:

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

A modifying factor of 10 was applied to the NOAEL to account for the limitations (no immune studies, one neurotoxicity study) in the toxicological database for α-HCH. Immune and nervous system effects are sensitive endpoints for other HCH isomers.

MRL = NOAEL ÷ (UF x MF)
0.9 mg/kg/day ÷ ((10 x 10) x 10) = 0.0009 mg/kg/day

Other Additional Studies or Pertinent Information that Lend Support to this MRL: Hepatic effects have been observed in rats, mice, and hamsters after intermediate- and chronic-duration oral exposures to α-HCH. Observed effects include increases in absolute and relative liver weight, hepatocellular hypertrophy and/or hyperplasia, focal necrosis, fatty degeneration, hepatomegaly, bile duct proliferation, oval cells, nodular hyperplasia, and megalocytosis (Fitzhugh et al. 1950; Ito et al. 1973, 1975, 1976; Nagasaki et al. 1975; Sumida et al. 2007; Tryphonas and Iverson 1983). Both rats and mice have also developed liver tumors at higher doses of α-HCH (Hanada et al. 1973; Ito et al. 1973, 1975, 1976; Nagasaki et al. 1975; Tryphonas and Iverson 1983; Tsukada et al. 1979).

There are no studies on the effects of α-HCH on the immune system. Studies of γ-HCH, and a few studies of β-HCH, have shown suppression of the immune system (e.g., Banerjee et al. 1996; Cornacoff et al. 1988; Desi et al. 1978; Dewan et al. 1980; Hong and Boorman 1993; Khurana et al. 1999; Koner et al. 1998; Mediratta et al. 2008; Meera et al. 1992; Van Velsen et al. 1986) and for γ-HCH, these effects occur at lower doses than hepatic effects in animal studies. Thus, the lack of immunotoxicity data for α-HCH is a significant limitation of the existing database for this isomer.

Data on the neurotoxicity of α-HCH are limited to a single study showing no change in motor nerve conduction velocity after 30 days of exposure (Muller et al. 1981). Both β- and γ-HCH have induced neurotoxic effects in laboratory rodents (e.g., Cornacoff et al. 1988; EPA 1999a; Gilbert and Mack 1995; Parmar et al. 2003; Van Velsen et al. 1986), and γ-HCH has induced neurotoxicity in humans exposed orally (e.g., Davies et al. 1983; Harris et al. 1969; Munk and Nantel 1977; Nordt and Chew 2000; Powell 1980; Starr and Clifford 1972; Storen 1955).

Agency Contacts (Chemical Managers): Malcolm Williams

MINIMAL RISK LEVEL (MRL) WORKSHEET

***Chemical Name*:** β-HCH
CAS Number: 319-85-7
Date: March 2024
Profile Status: Final
Route: Inhalation
Duration: Acute

MRL Summary: There are insufficient data for derivation of an acute-duration inhalation MRL for β-HCH.

Rationale for Not Deriving an MRL: No acute-duration inhalation studies of β-HCH in humans or animals were located, precluding derivation of an acute-duration inhalation MRL.

Agency Contacts (Chemical Managers): Malcolm Williams

APPENDIX A

MINIMAL RISK LEVEL (MRL) WORKSHEET

***Chemical Name*:** β-HCH
CAS Number: 319-85-7
Date: March 2024
Profile Status: Final
Route: Inhalation
Duration: Intermediate

MRL Summary: There are insufficient data for derivation of an intermediate-duration inhalation MRL for β-HCH.

Rationale for Not Deriving an MRL: No intermediate-duration inhalation studies of β-HCH in humans or animals were located, precluding derivation of an intermediate-duration inhalation MRL.

Agency Contacts (Chemical Managers): Malcolm Williams

APPENDIX A

MINIMAL RISK LEVEL (MRL) WORKSHEET

***Chemical Name*:** β-HCH
CAS Number: 319-85-7
Date: March 2024
Profile Status: Final
Route: Inhalation
Duration: Chronic

MRL Summary: There are insufficient data for derivation of a chronic-duration inhalation MRL for β-HCH.

Rationale for Not Deriving an MRL: No chronic-duration inhalation studies of β-HCH in humans or animals were located, precluding derivation of a chronic-duration inhalation MRL.

Agency Contacts (Chemical Managers): Malcolm Williams

APPENDIX A

MINIMAL RISK LEVEL (MRL) WORKSHEET

***Chemical Name*:** β-HCH
CAS Number: 319-85-7
Date: March 2024
Profile Status: Final
Route: Oral
Duration: Acute
MRL: 0.08 mg/kg/day
Critical Effect: Ataxia and hypoactivity
Reference: Van Velsen et al. 1986
Point of Departure: 8 mg/kg/day (NOAEL)
Uncertainty Factor: 100
LSE Graph Key: 2
Species: Rat

MRL Summary: An acute-duration oral MRL of 0.08 mg/kg/day was derived for β-HCH based on a NOAEL of 8 mg/kg/day in the first few weeks of a 13-week study (Van Velsen et al. 1986); the higher dose in this study (38 mg/kg/day) was a serious LOAEL for a neurological endpoint of ataxia and hypoactivity progressing in some animals to coma. A total uncertainty factor of 100 (10 each for extrapolation from animals to humans and for human variability) was applied to the NOAEL to derive the MRL.

Selection of the Critical Effect: Of the three studies reporting acute-duration oral exposure to β-HCH, two studies (Cornacoff et al. 1988; Van Velsen et al. 1986) identified clinical signs of neurotoxicity as the critical effect at doses of ≥38 mg/kg/day for 1–2 weeks. Renal effects were observed in the third study (Srinivasan et al. 1984) at a higher dose (72 mg/kg/day, the only dose tested).

Selection of the Principal Study: The lowest LOAEL for acute-duration oral exposure to β-HCH was 38 mg/kg/day for ataxia and hypoactivity signs observed in rats during for the first 2 weeks of a 13-week study (Van Velsen et al. 1986). At a dose of 8 mg/kg/day, no clinical signs of neurotoxicity were observed throughout the 13 weeks of exposure (NOAEL).

Summary of the Principal Study:

Van Velsen FL, Danse LHJC, Van Leeuwen FXR, et al. 1986. The subchronic oral toxicity of the β-isomer of hexachlorocyclohexane in rats. Fundam Appl Toxicol 6:697-712.

Groups of 10 male and 10 female Wistar rats were exposed to β-HCH in diets containing 0, 2, 10, 50, or 250 mg/kg feed in a 13-week study. The animals were weanlings at study initiation. For the acute (first 2 weeks) portion of the study, ATSDR calculated dose values using food intake and body weight for male and female weanling Wistar rats from EPA (1988b) to arrive at 0, 0.3, 1.5, 8, and 38 mg/kg/day doses. Clinical signs of toxicity were noted in the first few weeks of the study. At the end of week 2, two male and two female rats receiving 38 mg/kg/day in the diet exhibited ataxia and hypoactivity, progressing to coma within 3 days. The animals were humanely sacrificed, as were five additional males and six additional females that showed similar signs later in the study. No clinical signs were seen at lower doses of β-HCH at any time during the 13-week exposure period, nor were there histopathology changes in the brain, spinal cord, or sciatic nerve at any dose after 13 weeks of exposure.

Selection of the Point of Departure for the MRL: The NOAEL of 8 mg/kg/day was selected as the basis for the MRL. BMD modeling was not considered because the effects at the next highest dose (clinical signs of toxicity) reflected a serious LOAEL.

Adjustment for Intermittent Exposure: Not applicable.

Uncertainty Factor: The NOAEL of 8 mg/kg/day was divided by a total uncertainty factor of 100:

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

MRL = NOAEL ÷ (UF)
8 mg/kg/day ÷ (10 x10) = 0.08 mg/kg/day

Other Additional Studies or Pertinent Information that Lend Support to this MRL: Mice treated with 60 or 200 mg/kg/day β-HCH in the diet in a 30-day study developed ataxia within the first week of treatment (Cornacoff et al. 1988). The animals receiving 60 mg/kg/day recovered within a few days, while those receiving 200 mg/kg/day became markedly worse, leading to humane sacrifice of 80% of the animals in this group (Cornacoff et al. 1988). Effects on peripheral nerves were reported by Muller et al. (1981), who observed a significant delay in tail nerve conduction velocity in rats fed 66.3 mg β-HCH/kg/day for 30 days.

Agency Contacts (Chemical Managers): Malcolm Williams

APPENDIX A

MINIMAL RISK LEVEL (MRL) WORKSHEET

***Chemical Name*:** β-HCH
CAS Number: 319-85-7
Date: March 2024
Profile Status: Final
Route: Oral
Duration: Intermediate
MRL: 0.0006 mg/kg/day (0.6 µg/kg/day)
Critical Effect: Hyalinization of centrilobular cells in the liver
Reference: Van Velsen et al. 1986
Point of Departure: 0.18 mg/kg/day (minimal LOAEL)
Uncertainty Factor: 300
LSE Graph Key: 10
Species: Rat

MRL Summary: An intermediate-duration oral MRL of 0.0006 mg/kg/day (0.6 µg/kg/day) was derived for β-HCH based on a minimal LOAEL of 0.18 mg/kg/day for liver histopathology changes (hyalinization of centrilobular cells) in a 13-week study of rats exposed via the diet (Van Velsen et al. 1986). A total uncertainty factor of 300 (10 each for extrapolation from animals to humans and for human variability, and 3 for use of a minimal LOAEL) was applied to the LOAEL to derive the MRL.

Selection of the Critical Effect: Table A-2 provides a summary of the lowest effect levels in intermediate-duration animal studies of oral exposure to β-HCH. The lowest LOAEL was 0.18 mg/kg/day for hyalinization of centrilobular cells in the liver in male rats exposed for 13 weeks (Van Velsen et al. 1986); these effects increased with dose and are supported by the observation of β-HCH-induced liver effects at higher doses in other intermediate-duration oral studies in rats and mice (Hanada et al. 1973; Ito et al. 1973, 1975).

Table A-2. Summary of NOAELs and LOAELs from Candidate Intermediate-Duration Studies in Laboratory Animals Orally Exposed to β-Hexachlorocyclohexane

Species	Exposure scenario	NOAEL (mg/kg/day)	LOAEL (mg/kg/day)	Effect	Reference
Hepatic effects					
Rat (Wistar) 10 M	13 weeks (F)	ND	0.18 M (minimal LOAEL)	Hyalinization of centrilobular cells	Van Velsen et al. 1986
Mouse (dd) 20–40 M	24 weeks (F)	ND	18	18% increase in relative liver weight	Ito et al. 1973
Rat (W strain) 18–24 M	24–48 weeks (F)		35	Hepatocellular hypertrophy after 48 weeks	Ito et al. 1975
Mouse (dd) 10–11 M, 10–11 F	32 weeks (F)	20	60 F 50 M	Nuclear irregularities in foci of enlarged hepatocytes	Hanada et al. 1973

Table A-2. Summary of NOAELs and LOAELs from Candidate Intermediate-Duration Studies in Laboratory Animals Orally Exposed to β-Hexachlorocyclohexane

Species	Exposure scenario	NOAEL (mg/kg/day)	LOAEL (mg/kg/day)	Effect	Reference
Developmental effects					
Rat (Wistar) 6 F	GDs 0–21 LDs 1–28 (F)	ND	5 20 (serious LOAEL)	Increased liver weight in pups exposed during gestation and lactation 48% pup mortality by PND 5	Srinivasan et al. 1991

Principal study for the MRL.

(F) = feed; F = female(s); GD = gestation day; LD = lactation day; LOAEL = lowest-observed-adverse-effect level; M = male(s); ND = not determined; NOAEL = no-observed-adverse-effect level; PND = postnatal day

Selection of the Principal Study: Van Velsen et al. (1986) was selected as the principal study because it identified the lowest LOAEL among intermediate-duration oral studies.

Summary of the Principal Study:

Van Velsen FL, Danse LHJC, Van Leeuwen FXR, et al. 1986. The subchronic oral toxicity of the β-isomer of hexachlorocyclohexane in rats. Fundam Appl Toxicol 6:697-712.

Groups of 10 male and 10 female Wistar rats were exposed to β-HCH in diets containing 0, 2, 10, 50, or 250 mg/kg feed (>98% pure) for 13 weeks. ATSDR calculated doses corresponding to these concentrations using food factor values for male and female Wistar rats in subchronic studies as reported in EPA (1988b). Estimated dietary doses were 0, 0.18, 0.9, 4.5, or 22.5 mg/kg/day in males, and 0, 0.2, 1.0, 5, or 25 mg/kg/day in females. Clinical signs were monitored daily, and body weights and food intake were measured weekly. At sacrifice at the end of exposure, blood was collected for hematology and clinical chemistry. Necropsy evaluations included organ weights (liver, kidneys, spleen, thymus, adrenals, pituitary, testes, uterus, and ovaries), gross pathology, and comprehensive histopathology.

At the end of week 2, two male and two female rats receiving the highest dose exhibited ataxia and hypoactivity, progressing to coma within 3 days. The animals were humanely sacrificed, as were five additional males and six additional females that showed similar signs later in the study. Terminal body weight was significantly reduced (15.5% relative to controls in both males and females) in the animals from this dose group that survived. Other effects seen at the highest dose (either in the early decedents or in survivors or both), but not at lower doses, included: centrilobular hepatocytic hypertrophy, proliferation of smooth endoplasmic reticulum, increased microsomal activity, and/or increased glycogen content in the livers; hematologic and splenic changes indicative of anemia (decreased red blood cells and hemoglobin, increased extramedullary hematopoiesis); depletion of splenic lymphoid tissue; thymic cortical atrophy; adrenal cortical hypertrophy in both sexes; testicular and ovarian atrophy; and epithelial hyperplasia, metaplasia, and dilation of endometrial glands in the uterus.

No clinical signs, and no reductions in body weight gain or food intake were seen at lower doses of β-HCH. The lower dose groups had increased food intake and increases in body weight, but terminal body weights did not differ significantly from controls. At doses of 0.2–5 mg/kg/day, reduced neutrophil

counts were seen in females, but there were no other significant hematology changes. Clinical chemistry did not show any effects of treatment on serum AST, ALT, ALP, urea, IgM, or IgG levels. Dose-related trends in lower serum lactate and pyruvate were seen, but the only significant difference from controls was for serum lactate in 4.5 mg/kg/day males. Relative, but not absolute, testes weights were reduced (~10%) at 4.5 mg/kg/day, but the difference may have resulted from increased body weight (10% higher than controls) at this dose. At the highest dose, both absolute and relative testes weights were markedly reduced and accompanied by testicular atrophy. Kidney weights were significantly increased at all doses in females, but the increase did not show dose-dependence. In males, significant increases were seen only at ≥4.5 mg/kg/day and were accompanied by renal medullary calcinosis at 22.5 mg/kg/day; this lesion was seen in females at ≥5 mg/kg/day. Increased absolute and/or relative liver weights occurred at ≥0.9 mg/kg/day in males and ≥1.0 mg/kg/day in females. Increased incidences of hyalinization of centrilobular cells were observed in the livers of males at all doses, but not in females except in survivors at the highest dose (25 mg/kg/day). At the lowest dose (0.18 mg/kg/day), the hyalinization in males was characterized as slight. Females exhibited a low incidence of increased mitoses at 5 mg/kg/day. One male each in the 4.5 and 22.5 mg/kg/day groups exhibited focal liver cell necrosis. Periportal fat accumulation and/or focal liver cell necrosis occurred in males and females at ≥4.5 mg/kg/day. Based on the slight liver histopathology changes (hyalinization of centrilobular cells) seen in males at the lowest dose level, 0.18 mg/kg/day is considered to be a minimal LOAEL. A NOAEL could not be determined.

Selection of the Point of Departure for the MRL: The minimal LOAEL of 0.18 mg/kg/day was selected as the POD for MRL derivation because these effects occurred at the lowest dose (increased liver weights were seen at the next higher dose of 0.9–1.0 mg/kg/day). The histopathology findings in the liver did not exhibit a monotonic dose-response relationship and were thus not amenable to BMD modeling.

Adjustment for Intermittent Exposure: Not applicable.

Uncertainty Factor: The LOAEL of 0.18 mg/kg/day was divided by a total uncertainty factor of 300:

- 3 for use of a minimal LOAEL
- 10 for extrapolation from animals to humans
- 10 for human variability

MRL = NOAEL ÷ (UF)
0.18 mg/kg/day ÷ (3 x 10 x 10) = 0.0006 mg/kg/day

Other Additional Studies or Pertinent Information that Lend Support to this MRL: The liver is an established target of β-HCH in other intermediate- and chronic-duration oral studies in rats and mice (Fitzhugh et al. 1950; Ito et al. 1973).

Agency Contacts (Chemical Managers): Malcolm Williams

APPENDIX A

MINIMAL RISK LEVEL (MRL) WORKSHEET

***Chemical Name*:** β-HCH
CAS Number: 319-85-7
Date: March 2024
Profile Status: Final
Route: Oral
Duration: Chronic

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

Rationale for not deriving an MRL: Two chronic-duration oral studies of β-HCH were located: Fitzhugh et al. (1950) and Thorpe and Walker (1973). While Thorpe and Walker (1973) examined only cancer endpoints, Fitzhugh et al. (1950) evaluated histopathology in a large number of organs and identified the liver as the most sensitive target of chronic-duration oral exposure. In this study, the lowest dose (0.7–0.9 mg/kg/day) was identified as a LOAEL based on increased in liver weight and histological changes. A NOAEL was not identified. Thus, the only available chronic LOAEL (0.7–0.9 mg/kg/day) is higher than the LOAEL (0.18 mg/kg/day based on liver effects in a study by Van Velsen et al. 1986) used as the POD for intermediate MRL derivation. Thus, a chronic-duration MRL could not be derived based on the study by Fitzhugh et al. (1950).

Agency Contacts (Chemical Managers): Malcolm Williams

APPENDIX A

MINIMAL RISK LEVEL (MRL) WORKSHEET

***Chemical Name*:** γ-HCH
CAS Number: 58-89-9
Date: March 2024
Profile Status: Final
Route: Inhalation
Duration: Acute

MRL Summary: There are insufficient data for derivation of an acute-duration inhalation MRL for γ-HCH.

Rationale for Not Deriving an MRL: No acute-duration inhalation studies of γ-HCH in humans were located. Acute-duration inhalation studies in animals are shown in Table A-3, and include two studies of rats exposed for 4 hours (Oldiges et al. 1980; Ullmann 1986b) and data on clinical signs in mice in the first 2 weeks of an intermediate-duration study (Klonne and Kintigh 1988). Klonne and Kintigh (1988) observed 16% mortality in the first week of exposure to 10 mg/m3 γ-HCH (6 hours/day, 5 days/week). Other endpoints were not evaluated in the first few weeks of these studies (Klonne and Kintigh 1988; Oldiges et al. 1983). The rat studies of 4-hour exposures (Oldiges et al. 1980; Ullmann 1986b) identified freestanding LOAELs (101 or 237 mg/m^3). The available data are not adequate to identify sensitive targets of inhaled γ-HCH, precluding derivation of an acute-duration inhalation MRL for γ-HCH.

Table A-3. Summary of NOAELs and LOAELs from Acute-Duration Studies in Laboratory Animals Exposed to γ-Hexachlorocyclohexane by inhalation

Species	Exposure scenario	NOAEL (mg/m^3)	LOAEL (mg/m^3)	Effect	Reference
Mouse (CD-1) 45 M, 45 F	1 week 5 days/week 6 hours/day	ND	10 (serious LOAEL)	16% mortality during the first week	Klonne and Kintigh 1988
Rat (Wistar) 5 M, 5 F	4 hours	ND	101	Sedation	Ullmann 1986b
Rat (Wistar) 5 M, 5 F	4 hours	ND	237	Clinical signs of restlessness and hyperactivity	Oldiges et al. 1980

F = female(s); LOAEL = lowest-observed-adverse-effect level; M = male(s); ND = not determined; NOAEL = no-observed-adverse-effect level

Agency Contacts (Chemical Managers): Malcolm Williams

APPENDIX A

MINIMAL RISK LEVEL (MRL) WORKSHEET

***Chemical Name*:** γ-HCH
CAS Number: 58-89-9
Date: March 2024
Profile Status: Final
Route: Inhalation
Duration: Intermediate

MRL Summary: There are insufficient data for derivation of an intermediate-duration inhalation MRL for γ-HCH.

Rationale for Not Deriving an MRL: No intermediate-duration inhalation studies of γ-HCH in humans were located. Intermediate-duration inhalation studies in animals are shown in Table A-4, and include one study of rats exposed for 90 days (Oldiges et al. 1983) and a 14-week study of mice (Klonne and Kintigh 1988). The lowest LOAEL (0.5 mg/m^3) was identified for renal effects in male rats; a NOAEL for this endpoint was not identified (Oldiges et al. 1980). For the 14-week mouse study by Klonne and Kintigh (1988), a serious LOAEL of 1 mg/m^3 was identified for concentration-related increases in mortality. No deaths were observed at 0.3 mg/m^3, and there were no treatment-related effects on body weight, food and water intake, clinical chemistry, organ weight, bone marrow evaluations, ophthalmic evaluations, or gross or microscopic pathology (Klonne and Kintigh 1988). No studies evaluating developmental, neurological, immune system, or reproductive effects of γ-HCH in animals exposed by inhalation were located; these have been demonstrated to be sensitive endpoints of γ-HCH toxicity after oral exposure. The available data were not considered adequate for derivation of an intermediate-duration inhalation MRL due to the lack of studies on sensitive endpoints and because the lowest LOAEL (0.5 mg/m^3 in rats) is only one-half the serious LOAEL of 1 mg/mg^3 for mortality in mice.

Table A-4. Summary of NOAELs and LOAELs from Intermediate-Duration Studies in Laboratory Animals Exposed to γ-Hexachlorocyclohexane by inhalation

Species	Exposure scenario	NOAEL (mg/m^3)	LOAEL (mg/m^3)	Effect	Reference
Rat (Wistar) 5 M, 5 F	90 days day/week NS 6 hours/day	ND	0.5	Dilated renal tubules with protein-containing contents; proliferated tubules in males	Oldiges et al. 1983
Mouse (CD-1) 45 M, 45 F	14 week 5 days/week 6 hours/day	0.3	1 (serious LOAEL)	1/45 males and 1/45 females died at 1 mg/m^3; 5/45 males and 15/45 females died at 5 mg/m^3	Klonne and Kintigh 1988
Rat (Wistar) 5 M, 5 F	90 days days/week NS 6 hours/day	0.5	5	Diarrhea; bone marrow myelogram changes	Oldiges et al. 1983

F = female(s); LOAEL = lowest-observed-adverse-effect level; M = male(s); ND = not determined; NOAEL = no-observed-adverse-effect level; NS = not specified

Agency Contacts (Chemical Managers): Malcolm Williams

MINIMAL RISK LEVEL (MRL) WORKSHEET

***Chemical Name*:** γ-HCH
CAS Number: 58-89-9
Date: March 2024
Profile Status: Final
Route: Inhalation
Duration: Chronic

MRL Summary: There are insufficient data for derivation of a chronic-duration inhalation MRL for γ-HCH.

Rationale for Not Deriving an MRL: No chronic-duration inhalation studies of γ-HCH in humans or animals were located, precluding derivation of a chronic-duration inhalation MRL.

Agency Contacts (Chemical Managers): Malcolm Williams

MINIMAL RISK LEVEL (MRL) WORKSHEET

***Chemical Name*:** γ-HCH
CAS Number: 58-89-9
Date: March 2024
Profile Status: Final
Route: Oral
Duration: Acute
MRL: 0.003 mg/kg/day (3 µg/kg/day)
Critical Effect: Development of male reproductive tract
Reference: Dalsenter et al. 1997b
Point of Departure: 1 mg/kg/day (LOAEL)
Uncertainty Factor: 300
LSE Graph Key: 6
Species: Rat

MRL Summary: An acute-duration oral MRL of 0.003 mg/kg/day (3 µg/kg/day) was derived for γ-HCH based on a minimal LOAEL of 1 mg/kg/day for developmental effects (effects on developing reproductive system in male rat pups exposed during lactation) (Dalsenter et al. 1997b). A total uncertainty factor of 300 (10 for extrapolation from animals to humans, 10 for human variability, and 3 for use of a minimal LOAEL) was applied to the LOAEL to obtain the MRL.

Selection of the Critical Effect: Table A-5 provides a summary of the lowest effect levels in acute-duration oral studies of γ-HCH exposure in animals. The lowest effect level was a LOAEL of 1 mg/kg/day for effects on the development of the male reproductive tract in rat pups exposed during LDs 9–14 (Dalsenter et al. 1997b). These effects were seen during assessments on PNDs 65 and 140, as follows. At PND 65, there was no significant effect on relative testicular weight; 7% reduction relative epididymis weight; 29% lower spermatid count; 12% lower sperm count; and 30% lower testosterone levels. At PND 140, the differences from control had declined: testicular weight was 6% lower than controls; spermatid and sperm counts were 13% lower than controls; and there were no significant differences in relative epididymal weights or serum testosterone concentration. There were no effects on mating or fertility. There was no NOAEL associated with the study. The findings in this study are consistent with adverse effects on developing male reproductive organs reported in other animal studies (Agrahari et al. 2019; Dalsenter et al. 1997a, 1997b; Di Consiglio et al. 2009; La Sala et al. 2009; Traina et al. 2003).

Table A-5. Summary of NOAELs and LOAELs from Candidate Acute-Duration Studies in Laboratory Animals Orally Exposed to γ-Hexachlorocyclohexane (Doses ≤10 mg/kg/day)

Species	Exposure scenario	NOAEL (mg/kg/day)	LOAEL (mg/kg/day)	Effect	Reference
Developmental effects					
Rat (BOR: spf) 9 F	LDs 9–14 (GO)	ND	1 (minimal LOAEL)	In male pups, reduced relative testicular and epididymis weight (6–7%), spermatid and sperm counts (12–29%), and testosterone levels (8–30%) at maturity with no effect on fertility	Dalsenter et al. 1997b

Table A-5. Summary of NOAELs and LOAELs from Candidate Acute-Duration Studies in Laboratory Animals Orally Exposed to γ-Hexachlorocyclohexane (Doses ≤10 mg/kg/day)

Species	Exposure scenario	NOAEL (mg/kg/day)	LOAEL (mg/kg/day)	Effect	Reference
Rat (BOR: spf) 9 F	LD 9 or 14 once (GO)	ND	6	In male pups, reduced relative testicular and epididymis weight (~10%), spermatid and sperm counts (~8–10%), testosterone levels (~30-50%), Leydig cell numbers, and spermatogenesis at maturity with no effect on fertility	Dalsenter et al. 1997b
Neurological effects					
Rat (Sprague-Dawley) 9 M	6 days (GO)	ND	3	Increased pineal N-acetyl-transferase, decreased serotonin levels	Attia et al. 1991
Rat (Sprague-Dawley) 7–14 M	4 days (GO)	1	3	Increased kindling acquisition	Joy et al. 1982
Rat (Long-Evans) 14 M	Once (GO)	ND	5	Myoclonic jerks and single clonic seizure in kindled animals	Gilbert and Mack 1995
Rat (Wistar) 5 M, 5 F	3 days (GO)	ND	5 (serious LOAEL)	Decreased myelin in developing brain	Serrano et al. 1990
Hepatic effects					
Rat (Wistar) 6 M	3 days (GO)	ND	5	Fatty degeneration, vacuolation, and necrosis of the liver	Hfaiedh et al. 2012
Hematological effects					
Mouse (B6C3F1) 7 M	10 days (GO)	ND	10	Transient decrease in marrow progenitor cell numbers	Hong and Boorman 1993
Immunological effects					
Rat (Wistar) 8 M	14 days (NS)	ND	10	Reduced delayed-type hypersensitivity (43% decrease in foot pad thickness)	Mediratta et al. 2008
Mouse (B6C3F1) 7 M	10 days (GO)	ND	10	Dose-related decrease in relative thymus and spleen weights	Hong and Boorman 1993

Principal study for the MRL.

F = female(s); (GO) = gavage in oil vehicle; LD = lactation day; LOAEL = lowest-observed-adverse-effect level; M = male(s); ND = not determined; NOAEL = no-observed-adverse-effect level; NS = not specified

Selection of the Principal Study: The study by Dalsenter et al. (1997b) was selected for use in deriving the MRL because the lowest LOAEL (1 mg/kg/day) was identified for developmental effects in this study.

Summary of the Principal Study:

Dalsenter PR, Faqi AS, Webb J, et al. 1997b. Reproductive toxicity and toxicokinetics of lindane in the male offspring of rats exposed during lactation. Hum Exp Toxicol 16:146-153.

Reproductive toxicity was evaluated in male offspring of groups of nine Bor:spf female rats that were administered γ-HCH in peanut oil by gavage as a single 6 mg/kg dose on day 9 or day 14 of lactation, or as daily 1 mg/kg/day doses on days 9–14 of lactation (Dalsenter et al. 1997b). A group of nine controls was administered the vehicle alone on days 9–14 of lactation. Male offspring (10 or 20/group) were terminated on PND 65 (puberty) or 140 (adulthood) and evaluated for the following endpoints: testis and epididymis weights, spermatid and sperm numbers, serum testosterone level, sexual behavior at 130 days of age during 1:1 mating with unexposed females (mount latency, intromission and ejaculatory latency, number and frequency of intromissions), mating index (number sperm positive females/number males mated x100), pregnancy index (number of males that made females pregnant/number of males that made females sperm-positive x100), fertility index (number of days elapsed until males fertilized their female partner), pregnancy endpoints (numbers of litters, implantations/litters, fetuses/litter, resorptions), and testicular histology (6 mg/kg offspring only).

Effects occurred in all treated groups. Findings in the 1 mg/kg/day offspring included statistically significant ($p<0.05$) reductions in relative testicular weight at PND 140 (6% less than controls), relative epididymis weight at PND 65 (7%), spermatid number at PNDs 65 and 140 (29 and 13%, respectively), sperm number at PND 140 (13%), serum testosterone at PND 65 (30%), and increased number of intromissions per minute up to ejaculation at PND 130 (45%). Effects were generally similar in type and magnitude in the 6 mg/kg offspring following exposure on GDs 9 or 14, including significantly reduced relative testicular weight at PNDs 65 and 140 (~10%), spermatid and sperm numbers at PND 140 (~8–10%), and serum testosterone at PND 140 (~50%). There were no significant effects on sexual behavior or fertility in the 1 or 6 mg/kg/day offspring as shown by the mating, pregnancy, and fertility indices or other pregnancy endpoints. Thus, the statistically significant changes observed for relative organ weights, sperm number, hormone levels, and intromission incidence are considered minimally effective for reproduction; their associated dose levels are considered minimal LOAELs. The testicular histological examinations of the 6 mg/kg/day offspring showed large areas of normal tissue, although some areas had distinct changes ranging from small alterations to a pronounced effect. The most affected areas were the tubules in which the effects included necrotic changes and reductions in Leydig cell numbers and spermatogenesis.

Selection of the Point of Departure for the MRL: The minimal LOAEL of 1 mg/kg/day for effects on the developing male reproductive tract (Dalsenter et al. 1997b) was selected as the POD for derivation of the acute-duration oral MRL for γ-HCH. BMD modeling was not possible as only a single exposed group was included in the experiment.

Adjustment for Intermittent Exposure: Not applicable.

Uncertainty Factor: The changes in relative organ weights, sperm number, hormone levels, and intromission incidence at the LOAEL were not associated with effects on sexual behavior or fertility; thus, the dose is considered a minimal LOAEL. Therefore, the LOAEL of 1 mg/kg/day was divided by a total uncertainty factor of 300:

- 10 for extrapolation from animals to humans
- 10 for human variability
- 3 for use of a minimal LOAEL

MRL = LOAEL ÷ (UF)
1 mg/kg/day ÷ (3 x 10 x 10) ≈ 0.003 mg/kg/day

Other Additional Studies or Pertinent Information that Lend Support to this MRL: In male offspring of rats and mice exposed to γ-HCH via oral administration during gestation and/or postnatal development, effects on preputial separation, serum hormone levels, spermatogenesis, reproductive organ weights, and testicular histopathology have been reported (Agrahari et al. 2019; Dalsenter et al. 1997b; Di Consiglio et al. 2009; La Sala et al. 2009; Traina et al. 2003).

Agency Contacts (Chemical Managers): Malcolm Williams

MINIMAL RISK LEVEL (MRL) WORKSHEET

***Chemical Name*:** γ-HCH
CAS Number: 58-89-9
Date: March 2024
Profile Status: Final
Route: Oral
Duration: Intermediate
MRL: $8x10^{-7}$ mg/kg/day (0.8 ng/kg/day)
Critical Effect: Cardiac effects in offspring
Reference: Sauviat et al. 2005
Point of Departure: 0.000076 mg/kg/day (NOAEL)
Uncertainty Factor: 100
LSE Graph Key: 75
Species: Rat

MRL Summary: An intermediate-duration oral MRL of 0.0000008 ($8x10^{-7}$) mg/kg/day (0.8 ng/kg/day) was derived for γ-HCH based on a NOAEL of 0.000076 mg/kg/day for a developmental endpoint of cardiac effects in rat pups (Sauviat et al. 2005). A total uncertainty factor of 100 (10 for extrapolation from animals to humans and 10 for human variability) was applied to the NOAEL to obtain the MRL.

Selection of the Critical Effect: Table A-6 provides a summary of the lowest effect levels in intermediate-duration studies of animals exposed to γ-HCH by oral administration. The lowest effect level (0.00015 mg/kg/day, minimal LOAEL) was identified for effects on cardiac electrophysiology in rat pups exposed to γ-HCH in drinking water (Sauviat et al. 2005). A NOAEL of 0.000076 mg/kg/day was identified for this study. The effects at the LOAEL in this study were minimal, but additional evidence that the effects were adverse is provided by the serious findings of altered heart morphometry and cardiac histopathology changes at the higher dose of 0.0003 mg/kg/day. Histopathology evaluations were not conducted in rat pups receiving the lower doses.

Table A-6. Summary of NOAELs and LOAELs from Candidate Intermediate-Duration Studies in Laboratory Animals Orally Exposed to γ-Hexachlorocyclohexane (Doses ≤1 mg/kg/day)

Species	Exposure scenario	NOAEL (mg/kg/day)	LOAEL (mg/kg/day)	Effect	Reference
Developmental effects					
Rat (Sprague-Dawley) NS F	~13 weeks (premating, mating, gestation, lactation, and 3 weeks postweaning) (W)	0.000076	0.00015 (minimal LOAEL) 0.0003 (serious LOAEL)	LOAEL: altered ventricular electrophysiology. Serious LOAEL: 21% decrease in pup body weight; altered heart morphometry and electrophysiology; cardiac histopathology (hypertrophy in left ventricular area, unorganized collagen bundles and layers, fibroblast destruction)	Sauviat et al. 2005

Table A-6. Summary of NOAELs and LOAELs from Candidate Intermediate-Duration Studies in Laboratory Animals Orally Exposed to γ-Hexachlorocyclohexane (Doses ≤1 mg/kg/day)

Species	Exposure scenario	NOAEL (mg/kg/day)	LOAEL (mg/kg/day)	Effect	Reference
Rat (Wistar) 13–14 F	GDs 5–21 (GO)	0.125	0.25	Persistent hyperactivity	Johri et al. 2007
Rat (Wistar) 25 F	GDs 5–21 (GO)	ND	0.25 (serious LOAEL)	Ultrastructural changes in the brain (moderately distorted mitochondria and demyelinated neurons)	Srivastava et al. 2019
Immunological effects					
Mouse (Swiss albino) 6 F	24 weeks (F)	ND	0.012	Changes in cell- and humoral-mediated immune system	Meera et al. 1992

Principal study for the MRL.

(F) = feed; F = female(s); GD =gestation day; (GO) = gavage in oil vehicle; (W) = water; LOAEL = lowest-observed-adverse-effect level; M = male(s); ND = not determined; NOAEL = no-observed-adverse-effect level; PND = postnatal day

Selection of the Principal Study: The lowest LOAEL (0.00015 mg/kg/day) and NOAEL (0.000076 mg/kg/day) for any effect of γ-HCH was identified in the study by Sauviat et al. (2005). These doses are much lower than those associated with other effects of γ-HCH.

Summary of the Principal Study:

Sauviat MP, Bouvet S, Godeau G, et al. 2005. Electrical activity alterations induced by chronic absorption of lindane (gamma-hexachlorocyclohexane) trace concentrations in adult rat heart. Can J Physiol Pharmacol 83:243-251.

Groups of female Sprague-Dawley rats (number not reported) were administered γ-HCH via "beverage" at doses of 0.5, 1, or 2 ppb. ATSDR estimated corresponding maternal doses of 0, 0.000076, 0.00015, and 0.00030 mg/kg/day using water intake and body weight for female Sprague-Dawley rats in subchronic studies as reported in EPA (1988b). Doses were administered prior to mating for four estrous cycles (~2 weeks); throughout mating (~2 weeks), gestation (3 weeks), lactation (3 weeks), and growth (3 weeks) until pups were 6 weeks of age for a total of ~13 weeks. Exposure of the pups after weaning was not described but assumed to occur via water at the same dose as the dams. Offspring were sacrificed at 6 weeks of age. The left ventricular papillary muscles (LVPM) were dissected from 18 control rats from 7 litters; 5 rats from 2 litters in the 0.000076 mg/kg/day group; 7 rats from 2 litters in the 0.00015 mg/kg/day group; and 18 rats from 7 litters in the 0.0003 mg/kg/day group. Dissected LVPMs were evaluated for the following electrophysiologic measurements: resting potential, action potential, plateau, action potential duration, overshoot, end of early repolarization, and end of terminal repolarization. Cardiac weight, lipid content, and morphometry, as well as left ventricular papillary muscle histopathology were evaluated in pups from the 0.0003 mg/kg/day and control groups only.

The study authors indicated that the high-dose offspring were less sensitive to anesthesia and more sensitive to noise than other groups, but details of these assessments and findings were not provided. Body weights of pups were significantly decreased by 21% in the 0.0003 mg/kg/day group, compared to controls; no significant body weight changes were observed in other groups.

Morphometry analysis showed that hearts from pups in the 0.0003 mg/kg/day group had a 9% increase in heart width (relative to controls), but no significant change in length, with a corresponding 9% decrease in length-to-width ratio. Heart weights and total lipid content were not significantly different in the 0.0003 mg/kg/day group compared to control. At 0.0003 mg/kg/day, offspring heart morphology was described as more round and "cherry like." The study authors reported that hearts of treated offspring showed hypertrophied areas with thinning of the left ventricular wall and few developed papillary muscles. Histopathological examination in 0.0003 mg/kg/day offspring showed that the heart tissue muscle bundles and layers were unorganized and dissociated, with large hemorrhagic interspaces and dispersion of cell nuclei, destruction of fibroblasts, and dispersion and disorganization of collagen bundles, compared to control heart muscle. Incidences of changes were not reported, and these parameters were not assessed in pups from the 0.5 and 0.00015 mg/kg/day groups.

Electrophysiology changes were evident in LVPMs from animals exposed to 0.00015 mg/kg/day and 0.0003 mg/kg/day γ-HCH. Action potential durations were unchanged at 0.000076 mg/kg/day, but the plateau was shortened moderately at 0.00015 mg/kg/day, and significantly shortened at 0.0003 mg/kg/day. At 0.0003 mg/kg/day, the slow repolarizing phase was also significantly shortened.

The effects at the high dose (0.0003 mg/kg/day) represent a serious LOAEL for cardiac effects (histopathology and electrophysiology changes) and significant body weight decrements (21% decrease) in the developing rat. The only effect at the middle dose (0.00015 mg/kg/day) was shortened action potential duration at the initial plateau phase (measured at 0 millivolts); similar results were not observed in the early repolarization or terminal repolarization phases (measured at 40 and 10 millivolts, respectively). However, at the high dose (0.0003 mg/kg/day), there were effects in all three phases, suggesting a dose-response relationship. There was no assessment of cardiac morphometry or histopathology in offspring from the middle dose group. The electrophysiology changes observed at 0.00015 mg/kg/day are considered to represent a minimal LOAEL. The lowest dose (0.000076 mg/kg/day) was not associated with electrophysiology changes and is considered to be a NOAEL.

Selection of the Point of Departure for the MRL: The data on electrophysiology changes in the study by Sauviat et al. (2005) are not amenable to BMD modeling, as the authors did not report variability measures. Thus, the NOAEL of 0.000076 mg/kg/day was selected as the POD for MRL derivation.

Adjustment for Intermittent Exposure: Not applicable.

Uncertainty Factor: The NOAEL of 0.000076 mg/kg/day was divided by a total uncertainty factor (UF) of 100:

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

MRL = NOAEL ÷ (UF)
0.000076 mg/kg/day ÷ (10 x 10) ≈ 0.0000008 mg/kg/day ($8x10^{-7}$ mg/kg/day)

Other Additional Studies or Pertinent Information that Lend Support to this MRL: Sauviat et al. (2007) conducted a related study examining whether the cardiac effects seen after maternal exposure

could be induced by paternal exposure to γ-HCH in drinking water. In this study, male rats were exposed to a concentration of 2 μg/L for an unspecified "chronic" duration prior to mating with untreated females. The lack of information on exposure duration in the males precluded estimation of doses. In offspring sacrificed at 6 weeks of age, there were no effects on pup heart weight or shape or electrophysiology, but there were histopathology changes in the hearts similar to those reported by Sauviat et al. (2005) at the same drinking water concentration.

Agency Contacts (Chemical Managers): Malcolm Williams

MINIMAL RISK LEVEL (MRL) WORKSHEET

***Chemical Name*:** γ-HCH
CAS Number: 58-89-9
Date: March 2024
Profile Status: Final
Route: Oral
Duration: Chronic

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

Rationale for Not Deriving an MRL: No adequate exposure-response data were available for humans. Chronic-duration oral studies of γ-HCH that evaluated noncancer endpoints include a 2-year dog study (Rivett et al. 1978), 18-month and 2-year studies in rats (Ali and Shakoori 1998; Fitzhugh et al. 1950); and 78–80-week studies in mice (EPA 2000a; Herbst et al. 1975; Weisse and Herbst 1977). Table A-7 summarizes effect levels from these chronic studies. As the table shows, all of the effect levels are much higher than the POD (0.000076 mg/kg/day) used for derivation of the intermediate oral MRL for γ-HCH. Thus, the available oral chronic data were not considered adequate for MRL derivation.

Table A-7. Summary of NOAELs and LOAELs from Candidate Chronic-Duration Studies in Laboratory Animals Orally Exposed to γ-Hexachlorocyclohexane

Species	Exposure scenario	NOAEL (mg/kg/day)	LOAEL (mg/kg/day)	Effect	Reference
Dog (Beagle) 4 M, 4 F	104 weeks (F)	2.92	ND	No body weight, hepatic, hematological, or ocular effects	Rivett et al. 1978
Rat (Wistar) 10 M, 10 F	107 weeks (F)	4	7 M	Increased liver weight (35%); very slight microscopic liver damage; very slight microscopic kidney damage	Fitzhugh et al. 1950
Rat (Sprague-Dawley) 3–5 NS	18 month (F)	ND	9	Increased cell, nucleus, and nucleolus size; extensive cytoplasmolysis; slight cytoplasmic degeneration; increasing nuclear distortion	Ali and Shakoori 1998
Mouse (CD-1) 50 M, 50 F	78 weeks (F)	5.2 M	20.5 M	Centrilobular hepatocyte hypertrophy	EPA 2000a
Mouse (NMRI) 50 M, 50 F	80 weeks (F)	8.2 M	ND	No body weight or liver effects	Herbst et al. 1975; Weisse and Herbst 1977

(F) = feed; F = female(s); LOAEL = lowest-observed-adverse-effect level; M = male(s); ND = not determined; NOAEL = no-observed-adverse-effect level; NS = not specified

Agency Contacts (Chemical Managers): Malcolm Williams

MINIMAL RISK LEVEL (MRL) WORKSHEET

***Chemical Name*:** δ-HCH
CAS Number: 319-86-8
Date: March 2024
Profile Status: Final
Route: Inhalation
Duration: Acute

MRL Summary: There are insufficient data for derivation of an acute-duration inhalation MRL for δ-HCH.

Rationale for Not Deriving an MRL: No acute-duration inhalation studies of δ-HCH in humans or animals were located, precluding derivation of an acute-duration inhalation MRL.

Agency Contacts (Chemical Managers): Malcolm Williams

MINIMAL RISK LEVEL (MRL) WORKSHEET

***Chemical Name*:** δ-HCH
CAS Number: 319-86-8
Date: March 2024
Profile Status: Final
Route: Inhalation
Duration: Intermediate

MRL Summary: There are insufficient data for derivation of an intermediate-duration inhalation MRL for δ-HCH.

Rationale for Not Deriving an MRL: No intermediate-duration inhalation studies of δ-HCH in humans or animals were located, precluding derivation of an intermediate-duration inhalation MRL.

Agency Contacts (Chemical Managers): Malcolm Williams

MINIMAL RISK LEVEL (MRL) WORKSHEET

***Chemical Name*:** δ-HCH
CAS Number: 319-86-8
Date: March 2024
Profile Status: Final
Route: Inhalation
Duration: Chronic

MRL Summary: There are insufficient data for derivation of a chronic-duration inhalation MRL for δ-HCH.

Rationale for Not Deriving an MRL: No chronic-duration inhalation studies of δ-HCH in humans or animals were located, precluding derivation of a chronic-duration inhalation MRL.

Agency Contacts (Chemical Managers): Malcolm Williams

MINIMAL RISK LEVEL (MRL) WORKSHEET

***Chemical Name*:** δ-HCH
CAS Number: 319-86-8
Date: March 2024
Profile Status: Final
Route: Oral
Duration: Acute

MRL Summary: There are insufficient data for derivation of an acute-duration oral MRL for δ-HCH.

Rationale for Not Deriving an MRL: No acute-duration oral studies of δ-HCH in humans or animals were located, precluding derivation of an acute-duration oral MRL.

Agency Contacts (Chemical Managers): Malcolm Williams

APPENDIX A

MINIMAL RISK LEVEL (MRL) WORKSHEET

***Chemical Name*:** δ-HCH
CAS Numbers: 319-86-8
Date: March 2024
Profile Status: Final
Route: Oral
Duration: Intermediate

MRL Summary: There are insufficient data for derivation of an intermediate-duration oral MRL for δ-HCH.

Rationale for Not Deriving an MRL: No intermediate-duration oral studies of δ-HCH in humans were located. Two intermediate-duration oral studies of δ-HCH administered in feed were identified: a 48-week study in rats (Ito et al. 1975) and a 24-week study in mice (Ito et al. 1973). Both studies were focused on the evaluation of liver cancer, and only the liver was evaluated (organ weight and histopathology). Due to the lack of data pertaining to other potential target organs, these data are not considered adequate for derivation of an intermediate-duration oral MRL.

Agency Contacts (Chemical Managers): Malcolm Williams

MINIMAL RISK LEVEL (MRL) WORKSHEET

***Chemical Name*:** δ-HCH
CAS Number: 319-86-8
Date: March 2024
Profile Status: Final
Route: Oral
Duration: Chronic

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

Rationale for Not Deriving an MRL: No chronic-duration oral studies of δ-HCH in humans or animals were located, precluding derivation of a chronic-duration oral MRL.

Agency Contacts (Chemical Managers): Malcolm Williams

APPENDIX B. LITERATURE SEARCH FRAMEWORK FOR HCH

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 HCH.

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 HCH. ATSDR primarily focused on peer-reviewed articles without publication date or language restrictions. Foreign language studies are reviewed based on available English-language abstracts and/or tables (or summaries in regulatory assessments, such as International Agency for Research on Cancer [IARC] documents). If the study appears critical for hazard identification or MRL derivation, translation into English is requested. Non-peer-reviewed studies that were considered relevant to the assessment of the health effects of HCH 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 HCH 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

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

Developmental effects
Other noncancer effects
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 2005 draft toxicological profile for HCH released for public comment in January 2023; thus, the literature search was restricted to studies published between January 2020 and June 2023. The following main databases were searched in June 2023:

- 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 HCH. 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 HCH 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	
06/2023	((319-84-6[rn] OR 319-85-7[rn] OR YM80ODM9PD[rn] OR 319-86-8[rn] OR 58-89-9[rn] OR 59NEE7PCAB[rn] OR 608-73-1[rn] OR "Hexachlorocyclohexane"[mh]) AND (("Hexachlorocyclohexane/toxicity"[mh] OR "Hexachlorocyclohexane/adverse effects"[mh] OR "Hexachlorocyclohexane/poisoning"[mh] OR "Hexachlorocyclohexane/pharmacokinetics"[mh] OR "Hexachlorocyclohexane/blood"[mh] OR "Hexachlorocyclohexane/cerebrospinal fluid"[mh] OR "Hexachlorocyclohexane/urine"[mh] OR "Hexachlorocyclohexane/antagonists and inhibitors"[mh] OR ("Hexachlorocyclohexane/metabolism"[mh] AND ("humans"[mh] OR "animals"[mh]))) OR ("Hexachlorocyclohexane"[mh] AND ("environmental exposure"[mh] OR ci[sh])) OR ("Hexachlorocyclohexane"[mh] AND toxicokinetics[mh:noexp]) OR ("Hexachlorocyclohexane"[mh] AND ("endocrine system"[mh] OR "hormones, hormone substitutes, and hormone antagonists"[mh] OR "endocrine disruptors"[mh])) OR ("Hexachlorocyclohexane"[mh] AND ("computational biology"[mh] OR "medical informatics"[mh] OR genomics[mh] OR genome[mh] OR proteomics[mh] OR proteome[mh] OR metabolomics[mh] OR metabolome[mh] OR genes[mh] OR "gene expression"[mh] OR phenotype[mh] OR genetics[mh] OR genotype[mh] OR transcriptome[mh] OR ("systems biology"[mh] AND ("environmental exposure"[mh] OR "epidemiological monitoring"[mh] OR analysis[sh])) OR "transcription, genetic "[mh] OR "reverse transcription"[mh] OR "transcriptional activation"[mh] OR "transcription factors"[mh] OR ("biosynthesis"[sh] AND (RNA[mh] OR DNA[mh])) OR "RNA, messenger"[mh] OR "RNA, transfer"[mh] OR "peptide biosynthesis"[mh] OR "protein biosynthesis"[mh] OR "reverse transcriptase polymerase chain reaction"[mh] OR "base sequence"[mh] OR "trans-activators"[mh] OR "gene expression profiling"[mh])) OR ("Hexachlorocyclohexane"[mh] AND (("Neoplasms"[mh] OR "Carcinogens"[mh] OR "Lymphoproliferative disorders"[mh] OR "Myeloproliferative disorders"[mh] OR "Toxicity Tests"[mh] OR ((cancer*[tiab] OR carcinogen*[tiab]) AND (risk*[tiab] OR health[tiab]) AND assessment*[tiab]) OR "Mutagens"[mh] OR "Mutagenicity Tests"[mh] OR "Chromosome Aberrations"[mh] OR "DNA Damage"[mh] OR "DNA Repair"[mh] OR "DNA Replication/drug effects"[mh] OR "DNA/drug effects"[mh] OR "DNA/metabolism"[mh] OR "Genomic Instability"[mh] OR "Salmonella typhimurium/drug effects"[mh] OR "Salmonella typhimurium/genetics"[mh] OR "Sister Chromatid Exchange"[mh] OR strand-break*[tiab]))) OR Hexachlorocyclohexane/pharmacology[majr]) AND (2020/10/01:3000[mhda] OR 2020:3000[dp])) OR ((("gamma-Lindane"[tw] OR "1,2,3,4,5,6-Hexachlorocyclohexane"[tw] OR "alpha-Benzene hexachloride"[tw] OR "alpha-Hexachlorocyclohexane"[tw] OR "beta-Benzene hexachloride"[tw] OR "beta-hexachlorocyclohexane"[tw] OR "delta-Benzene hexachloride"[tw] OR "delta-Benzenehexachloride"[tw] OR "delta-Hexachlorocyclohexane"[tw] OR "gamma-Benzene

Table B-2. Database Query Strings

Database search date	Query string
	hexachloride"[tw] OR "gamma-Hexachlorocyclohexane"[tw] OR "1,2,3,4,5,6-Hexachlorocyclohexane"[tw] OR "A-Hexachlorocyclohexane"[tw] OR "alpha-Benzene hexachloride"[tw] OR "alpha-Benzenehexachloride"[tw] OR "alpha-Hexachlorocyclohexane"[tw] OR "alpha-Lindane"[tw] OR "Aalindan"[tw] OR "Aficide"[tw] OR "Agrocide"[tw] OR "Agronexit"[tw] OR "Ameisentod"[tw] OR "Aparasin"[tw] OR "Aphtiria"[tw] OR "Aplidal"[tw] OR "Arbitex"[tw] OR "B-Hexachlorocyclohexane"[tw] OR "Benzene hexachloride"[tw] OR "Benzenehexachloride"[tw] OR "beta-1,2,3,4,5,6-Hexachlorocyclohexane"[tw] OR "beta-Hexachloro-cyclohexane"[tw] OR "beta-Hexachlorocyclohexane"[tw] OR "beta-Lindane"[tw] OR "Ben-Hex"[tw] OR "Benhexol"[tw] OR "Bexol"[tw] OR "Celanex"[tw] OR "Chloresene"[tw] OR "Codechine"[tw] OR "Cyclohexane, 1,2,3,4,5,6-hexachloro-"[tw] OR "delta-Benzene hexachloride"[tw] OR "delta-Benzenehexachloride"[tw] OR "delta-Hexachlorocyclohexane"[tw] OR "delta-Lindane"[tw] OR "Devoran"[tw] OR "Entomoxan"[tw] OR "Forst-Nexen"[tw] OR "Gallogama"[tw] OR "Gamacarbatox"[tw] OR "Gamacid"[tw] OR "Gamacide"[tw] OR "Gamaphex"[tw] OR "Gamene"[tw] OR "Gamiso"[tw] OR "gamma Benzene hexachloride"[tw] OR "gamma-1,2,3,4,5,6-Hexachlorocyclohexane"[tw] OR "gamma-Benzenehexachloride"[tw] OR "gamma-Benzohexachloride"[tw] OR "gamma-Hexachlorcyclohexanum"[tw] OR "gamma-Hexachloro-cyclohexane"[tw] OR "gamma-Hexachlorobenzene"[tw] OR "gamma-Hexachlorocyclohexane"[tw] OR "Gammalin"[tw] OR "Gammaterr"[tw] OR "Gammexane"[tw] OR "Geobilan"[tw] OR "Gexane"[tw] OR "HEXACHLORCYCLOHEXANE"[tw] OR "Hexachloro-cyclohexane"[tw] OR "Hexachlorocyclohexane"[tw] OR "hexachlorocyclohexanes"[tw] OR "Hexachlor"[tw] OR "Hexachlorcyclohexan"[tw] OR "Heclotox"[tw] OR "Hexachloran"[tw] OR "Hexachlorane"[tw] OR "Hexaverm"[tw] OR "Hexcidum"[tw] OR "Hexicide"[tw] OR "Hexyclan"[tw] OR "Hilbeech"[tw] OR "Hortex"[tw] OR "Hungaria L 7"[tw] OR "Hungaria L-7"[tw] OR "Jacutin"[tw] OR "Kokotine"[tw] OR "Kwell"[tw] OR "Lindane"[tw] OR "Lasochron"[tw] OR "Lendine"[tw] OR "Lentox"[tw] OR "Lidenal"[tw] OR "Lindafor"[tw] OR "Lindagam"[tw] OR "Lindagrain"[tw] OR "Lindagranox"[tw] OR "Lindan"[tw] OR "Lindanum"[tw] OR "Lindapoudre"[tw] OR "Lindatox"[tw] OR "Lindosep"[tw] OR "Lintox"[tw] OR "Linvur"[tw] OR "Lorexane"[tw] OR "Mszycol"[tw] OR "Neo-Scabicidol"[tw] OR "Nexol E"[tw] OR "Nexol-E"[tw] OR "Nicochloran"[tw] OR "Novigam"[tw] OR "Omnitox"[tw] OR "Ovadziak"[tw] OR "Owadziak"[tw] OR "Pedraczak"[tw] OR "Pflanzol"[tw] OR "Quellada"[tw] OR "Technical HCH"[tw] OR "technical grade HCH"[tw] OR "Scabene"[tw] OR "Silvanol"[tw] OR "Spritz-Rapidin"[tw] OR "Spritzlindane"[tw] OR "Spruehpflanzol"[tw] OR "Streunex"[tw] OR "α-Benzohexachloride"[tw] OR "α-Hexachlorocyclohexane"[tw] OR "α-Lindane"[tw] OR "β-1,2,3,4,5,6-Hexachlorocyclohexane"[tw] OR "β-Benzene hexachloride"[tw] OR "β-Hexachlorocyclohexane"[tw] OR "β-Lindane"[tw] OR "γ-1,2,3,4,5,6-Hexachlorocyclohexane"[tw] OR "γ-Benzene hexachloride"[tw] OR "γ-Benzohexachloride"[tw] OR "γ-Hexachlorobenzene"[tw] OR "γ-Hexachlorocyclohexane"[tw] OR "γ-Lindane"[tw] OR "δ-Benzene hexachloride"[tw] OR "δ-Hexachlorocyclohexane"[tw] OR "δ-Lindane"[tw] OR "alpha BHC"[tw] OR "alpha-BHC"[tw] OR "alpha-HCH"[tw] OR "beta BHC"[tw] OR "beta-BHC"[tw] OR "beta-HCH"[tw] OR "delta BHC"[tw] OR "delta-BHC"[tw] OR "delta-HCH"[tw] OR "Gamma-BHC"[tw] OR "gamma-HCH"[tw] OR "Nexit Stark"[tw] OR "Nexit-stark"[tw] OR "α-BHC"[tw] OR "α-HCH"[tw] OR "β-666"[tw] OR "β-BHC"[tw] OR "β-HCH"[tw] OR "γ-666"[tw] OR "γ-BHC"[tw] OR "γ-HCH"[tw] OR "δ-BHC"[tw] OR "δ-HCH"[tw] OR "total BHC"[tw] OR "α-666"[tw] OR "δ-666"[tw] OR "BHC-gamma"[tw] OR "α-Benzenehexachloride"[tw] OR "Detox 25"[tw] OR "Dol Granule"[tw] OR "ENT 7,796"[tw] OR "TAP 85"[tw] OR "Ameisenmittel merck"[tw] OR "Arcotal S"[tw] OR "Bentox 10"[tw] OR "Benzene-1,2,3,4,5,6-hexachloride"[tw] OR "beta-Hexachlorobenzene"[tw] OR "Detmol Extract"[tw] OR "Fenoform forte"[tw] OR "Gamma-mean 400"[tw] OR "Geolin G

Table B-2. Database Query Strings

Database search date	Query string
	3"[tw] OR "Hungaria L7"[tw] OR "Mglawik L"[tw] OR "Milbol 49"[tw] OR "Nexen FB"[tw] OR "sang-gamma"[tw] OR "sang gamma"[tw] OR "Submar"[tw] OR "Verindal Ultra"[tw] OR "HCH, technical grade"[tw] OR "beta-Hexachlorobenzene"[tw] OR "β-Hexachlorobenzene"[tw]) NOT medline[sb]) AND (2020/10/01:3000[crdt] OR 2020/10/01:3000[edat] OR 2020:3000[dp])) OR ("Hexachlorocyclohexane"[mh] AND 2022/04/01:3000[mhda])
	"A-HCCH"[tw] OR "BHC alpha"[tw] OR "BHC beta"[tw] OR "BHC delta"[tw] OR "BHC-alpha"[tw] OR "BHC-beta"[tw] OR "BHC-delta"[tw] OR "D-BHC"[tw] OR "D-HCCH"[tw] OR "Dolmix"[tw] OR "ENT 9,232"[tw] OR "HCH (mixed isomers)"[tw] OR "Tri-6 Dust No. 30"[tw] OR "(1.alpha.,2.alpha.,3.beta.,4.alpha.,5.alpha.,6.beta.)-1,2,3,4,5,6-Hexachlorocyclohexane"[tw] OR "(1.alpha.,2.alpha.,3.beta.,4.alpha.,5.beta.,6.beta.)1,2,3,4,5,6-Hexachlorocyclohexane"[tw] OR "(1.alpha.,2.beta.,3.alpha.,4.beta.,5.alpha.,6.beta.)-1,2,3,4,5,6-hexachloro-cyclohexane"[tw] OR "(1alpha,2alpha,3alpha,4beta,5alpha,6beta)-1,2,3,4,5,6-Hexachlorocyclohexane"[tw] OR "(1alpha,2alpha,3beta,4alpha,5beta,6beta)-1,2,3,4,5,6-Hexachlorocyclohexane"[tw] OR "(1alpha,2beta,3alpha,4beta,5alpha,6beta)-1,2,3,4,5,6-Hexachlorocyclohexane"[tw] OR "(1r,2r,3r,4r,5r,6r)-1,2,3,4,5,6-Hexachlorocyclohexane"[tw] OR "(1R,2R,3R,4R,5S,6S)-1,2,3,4,5,6-Hexachlorocyclohexane"[tw] OR "(1R,2S,3r,4R,5S,6r)-1,2,3,4,5,6-Hexachlorocyclohexane"[tw] OR "(1R,2S,3r,4R,5S,6s)-1,2,3,4,5,6-Hexachlorocyclohexane"[tw] OR "alpha-hexachlorcyclohexane"[tw] OR "1,2,3,4,5,6-G-HEXACHLOROCYCLOHEXANE"[tw] OR "1,2,3,4,5,6-Hexachloro(1a,2a,3a,4b,5a,6b)cyclohexane"[tw] OR "1,2,3,4,5,6-Hexachloro(1a,2b,3a,4b,5a,6b)cyclohexane"[tw] OR "1,2,3,4,5,6-Hexachloro-(1.alpha.,2.alpha.,3.alpha.,4.beta.,5.alpha.,6.beta.) cyclohexane"[tw] OR "1-alpha,2-alpha,3-alpha,4-beta,5-alpha,6-beta-Hexachlorocyclohexane"[tw] OR "1-alpha,2-beta,3-alpha,4-beta,5-alpha,6-beta-Hexachlorocyclohexane"[tw] OR "1a,2a,3b,4a,5b,6b-Hexachlorocyclohexane"[tw] OR "1α,2α,3β,4α,5α,6β)-1,2,3,4,5,6-Hexachlorocyclohexane"[tw] OR "(1α,2α,3α,4β,5α,6.β)-1,2,3,4,5,6-Hexachlorocyclohexane"[tw] OR "(1α,2α,3α,4β,5α,6β)-1,2,3,4,5,6-Hexachlorcyclohexan"[tw] OR "(1α,2α,3α,4β,5α,6β)-1,2,3,4,5,6-hexachlorocyclohexane"[tw] OR "(1α,2α,3α,4β,5α,6β)-1,2,3,4,5,6-hexaclorociclohexano"[tw] OR "(1α,2α,3β,4α,5α,6β)-1,2,3,4,5,6-Hexachlorocyclohexane"[tw] OR "(1α,2α,3β,4α,5β,6β)-1,2,3,4,5,6-Hexachlorcyclohexan"[tw] OR "(1α,2α,3β,4α,5β,6β)-1,2,3,4,5,6-Hexachlorocyclohexane"[tw] OR "(1α,2α,3β,4α,5β,6β)-1,2,3,4,5,6-hexaclorociclohexano"[tw] OR "(1α,2β,3α,4β,5α,6β)-1,2,3,4,5,6-Hexachlorcyclohexan"[tw] OR "(1α,2β,3α,4β,5α,6β)-1,2,3,4,5,6-Hexachlorocyclohexane"[tw] OR "(1α,2β,3α,4β,5α,6β)-1,2,3,4,5,6-hexaclorociclohexano"[tw] OR "A-Benzene hexachloride"[tw] OR "ALPHA-1,2,3,4,5,6-HEXACHLORCYCLOHEXAN"[tw] OR "alpha-1,2,3,4,5,6-Hexachlorocyclohexane"[tw] OR "alpha-Hexachloran"[tw] OR "alpha-Hexachlorane"[tw] OR "B-Benzene hexachloride"[tw] OR "Bercema-Spritz-Lindan 50"[tw] OR "Benzanex"[tw] OR "BETA-1,2,3,4,5,6-HEXACHLORCYCLOHEXAN"[tw] OR "beta-Hexachloran"[tw] OR "delta-(Aeeeee)-1,2,3,4,5,6-hexachlorocyclohexane"[tw] OR "Gamaxene"[tw] OR "Gamoline"[tw] OR "Gamtox"[tw] OR "Grammexene"[tw] OR "Gyben"[tw] OR "Hexablanc"[tw] OR "Hexachlorine cyclohexane"[tw] OR "Hexachlorzyklohexan"[tw] OR "Hexamul"[tw] OR "Hexapoudre"[tw] OR "Isatox"[tw] OR "Kanodane"[tw] OR "Lidano"[tw] OR "Prodactif"[tw] OR "Scabecid"[tw] OR "trans-alpha-Benzenehexachloride"[tw] OR "Trives-T"[tw] OR "BHC, d-"[tw] OR "BHC, total"[tw] OR "Cyclohexane,1,2,3,4,5,6-hexachloro-(1.alpha.,2.alpha.,3.beta.,4.alpha.,5.alpha.,6.beta.)-]"[tw] OR "Ciclohexano, 1,2,3,4,5,6-hexacloro-"[tw] OR "Cyclohexane, beta-1,2,3,4,5,6-

Table B-2. Database Query Strings

Database search date	Query string
	hexachloro-"[tw] OR "Cyclohexane, delta-1,2,3,4,5,6-hexachloro-"[tw] OR "Cyclohexane, l,2,3,4,5,6-hexachloro-, (1alpha,2alpha,3beta,4alpha,5beta,6beta)-"[tw] OR "1,2,3,4,5,6-Benzenehexachloride"[tw] OR "1,2,3,4,5,6-Hexachlorohexane"[tw] OR "D-Benzene hexachloride"[tw] OR "D-Hexachlorocyclohexane"[tw] OR "delta-1,2,3,4,5,6-Hexachlorocyclohexane"[tw] OR "α-1,2,3,4,5,6-Hexachlorocyclohexane"[tw] OR "α-Hexachloran"[tw] OR "α-Hexachlorane"[tw] OR "α-Hexachlorcyclohexane"[tw] OR "β-Hexachloran"[tw] OR "γ-Hexachloran"[tw] OR "γ-Hexachlorane"[tw] OR "δ-1,2,3,4,5,6-Hexachlorocyclohexane"[tw]
NTRL	
06/2023	Date limit 2020-2023 Search Titles OR Keywords; "Hexachlorocyclohexane" OR "Benzene hexachloride" OR "Lindane" OR "Hexachlorane" OR "Benzenehexachloride"
Toxcenter	
06/2023	FILE 'TOXCENTER' ENTERED AT 16:26:48 ON 12 MAY 2023 L1 35805 SEA 319-84-6 OR 319-85-7 OR 319-86-8 OR 58-89-9 OR 608-73-1 L2 35767 SEA L1 NOT TSCATS/FS L3 35033 SEA L2 NOT PATENT/DT L4 1214 SEA L3 AND ED>=20201001 ACTIVATE TOXQUERY/Q --------- L7 QUE (CHRONIC OR IMMUNOTOX? OR NEUROTOX? OR TOXICOKIN? OR BIOMARKER? OR NEUROLOG?) L8 QUE (PHARMACOKIN? OR SUBCHRONIC OR PBPK OR EPIDEMIOLOGY/ST,CT, IT) L9 QUE (ACUTE OR SUBACUTE OR LD50# OR LD(W)50 OR LC50# OR LC(W)50) L10 QUE (TOXICITY OR ADVERSE OR POISONING)/ST,CT,IT L11 QUE (INHAL? OR PULMON? OR NASAL? OR LUNG? OR RESPIR?) L12 QUE ((OCCUPATION? OR WORKPLACE? OR WORKER?) AND EXPOS?) L13 QUE (ORAL OR ORALLY OR INGEST? OR GAVAGE? OR DIET OR DIETS OR DIETARY OR DRINKING(W)WATER?) L14 QUE (MAXIMUM AND CONCENTRATION? AND (ALLOWABLE OR PERMISSIBLE)) L15 QUE (ABORT? OR ABNORMALIT? OR EMBRYO? OR CLEFT? OR FETUS?) L16 QUE (FOETUS? OR FETAL? OR FOETAL? OR FERTIL? OR MALFORM? OR OVUM?) L17 QUE (OVA OR OVARY OR PLACENTA? OR PREGNAN? OR PRENATAL?) L18 QUE (PERINATAL? OR POSTNATAL? OR REPRODUC? OR STERIL? OR TERATOGEN?) L19 QUE (SPERM OR SPERMAC? OR SPERMAG? OR SPERMATI? OR SPERMAS? OR SPERMATOB? OR SPERMATOC? OR SPERMATOG?) L20 QUE (SPERMATOI? OR SPERMATOL? OR SPERMATOR? OR SPERMATOX? OR

Table B-2. Database Query Strings

Database search date	Query string
	SPERMATOZ? OR SPERMATU? OR SPERMI? OR SPERMO?) L21 QUE (NEONAT? OR NEWBORN? OR DEVELOPMENT OR DEVELOPMENTAL?) L22 QUE (ENDOCRIN? AND DISRUPT?) L23 QUE (ZYGOTE? OR CHILD OR CHILDREN OR ADOLESCEN? OR INFANT?) L24 QUE (WEAN? OR OFFSPRING OR AGE(W)FACTOR?) L25 QUE (DERMAL? OR DERMIS OR SKIN OR EPIDERM? OR CUTANEOUS?) L26 QUE (CARCINOG? OR COCARCINOG? OR CANCER? OR PRECANCER? OR NEOPLAS?) L27 QUE (TUMOR? OR TUMOUR? OR ONCOGEN? OR LYMPHOMA? OR CARCINOM?) L28 QUE (GENETOX? OR GENOTOX? OR MUTAGEN? OR GENETIC(W)TOXIC?) L29 QUE (NEPHROTOX? OR HEPATOTOX?) L30 QUE (ENDOCRIN? OR ESTROGEN? OR ANDROGEN? OR HORMON?) L31 QUE (OCCUPATION? OR WORKER? OR WORKPLACE? OR EPIDEM?) L32 QUE 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 OR L30 OR L31 --------- L42 643 SEA L4 AND L32 L44 71 SEA L42 AND MEDLINE/FS L45 125 SEA L42 AND BIOSIS/FS L46 447 SEA L42 AND CAPLUS/FS L47 0 SEA L42 NOT (MEDLINE/FS OR BIOSIS/FS OR CAPLUS/FS) L48 564 DUP REM L44 L45 L46 (79 DUPLICATES REMOVED) L*** DEL 71 S L42 AND MEDLINE/FS L*** DEL 71 S L42 AND MEDLINE/FS L49 71 SEA L48 L*** DEL 125 S L42 AND BIOSIS/FS L*** DEL 125 S L42 AND BIOSIS/FS L50 113 SEA L48 L*** DEL 447 S L42 AND CAPLUS/FS L*** DEL 447 S L42 AND CAPLUS/FS L51 380 SEA L48 L52 493 SEA (L49 OR L50 OR L51) NOT MEDLINE/FS D SCAN L52

Table B-3. Strategies to Augment the Literature Search

Source	Query and number screened when available
TSCATS via ChemView	
06/2023	Compounds searched: 319-84-6, 319-85-7, 319-86-8, 58-89-9, 608-73-1

Table B-3. Strategies to Augment the Literature Search

Source	Query and number screened when available
NTP	
06/2023	Date limit 2020-2023 "58-89-9" "Hexachlorocyclohexane" "Lindane" "Benzene hexachloride" "319-84-6" "319-85-7" "319-86-8" "608-73-1" "Hexachlorane" "Benzenehexachloride"
Regulations.gov	
06/2023	"Lindane" Hexachlorocyclohexane "Benzene hexachloride" "Hexachlorane" "Benzenehexachloride" "319-84-6" "319-85-7" "319-86-8" "58-89-9" "608-73-1"
NIH RePORTER	
07/2023	Search Criteria: Fiscal Year: Active Projects; Text Search: lindane OR Hexachlorocyclohexane OR Hexachlorocyclohexanes OR "Benzene hexachloride" OR Hexachlorane OR Benzenehexachloride (advanced); Limit to: Project Title, Project Terms, Project Abstracts Search Criteria: Fiscal Year: Active Projects; Text Search: "gamma-Lindane" OR "1,2,3,4,5,6-Hexachlorocyclohexane" OR "alpha-Benzene hexachloride" OR "alpha-Hexachlorocyclohexane" OR "beta-Benzene hexachloride" OR "beta-hexachlorocyclohexane" OR "delta-Benzene hexachloride" OR "delta-Benzenehexachloride" OR "delta-Hexachlorocyclohexane" OR "gamma-Benzene hexachloride" OR "gamma-Hexachlorocyclohexane" OR "1,2,3,4,5,6-Hexachlorocyclohexane" OR "A-Hexachlorocyclohexane" OR "alpha-Benzene hexachloride" OR "alpha-Benzenehexachloride" OR "alpha-Hexachlorocyclohexane" OR "alpha-Lindane" OR "Aalindan" OR "Aficide" OR "Agrocide" OR "Agronexit" OR "Ameisentod" OR "Aparasin" OR "Aphtiria" OR "Aplidal" OR "Arbitex" OR "B-Hexachlorocyclohexane" OR "Benzene hexachloride" OR "Benzenehexachloride" OR "beta-1,2,3,4,5,6-Hexachlorocyclohexane" OR "beta-Hexachloro-cyclohexane" OR "beta-Hexachlorocyclohexane" OR "beta-Lindane" OR "Ben-Hex" OR "Benhexol" OR "Bexol" OR "Celanex" OR "Chloresene" OR "Codechine" OR "Cyclohexane, 1,2,3,4,5,6-hexachloro-" OR "delta-Benzene hexachloride" OR "delta-Benzenehexachloride" OR "delta-Hexachlorocyclohexane" OR "delta-Lindane" OR "Devoran" OR "Entomoxan" OR "Forst-Nexen" OR "Gallogama" OR "Gamacarbatox" OR "Gamacid" OR "Gamacide" OR "Gamaphex" OR "Gamene" OR "Gamiso" OR "gamma Benzene hexachloride" OR "gamma-1,2,3,4,5,6-Hexachlorocyclohexane" OR "gamma-Benzenehexachloride" OR "gamma-Benzohexachloride" OR "gamma-Hexachlorcyclohexanum" OR "gamma-Hexachloro-cyclohexane" OR "gamma-Hexachlorobenzene" OR "gamma-Hexachlorocyclohexane" OR "Gammalin" OR "Gammaterr" OR "Gammexane" OR "Geobilan" OR "Gexane" OR "HEXACHLORCYCLOHEXANE" OR "Hexachloro-cyclohexane" OR "Hexachlorocyclohexane" OR "hexachlorocyclohexanes" OR "Hexachlor" OR "Hexachlorcyclohexan" OR "Heclotox" OR "Hexachloran" OR "Hexachlorane" OR "Hexaverm" OR "Hexcidum" OR "Hexicide" OR "Hexyclan" OR "Hilbeech" OR "Hortex" OR "Hungaria L 7" OR "Hungaria L-7" OR "Jacutin" OR "Kokotine" OR "Kwell" OR "Lindane" OR "Lasochron" OR "Lendine" OR "Lentox" OR "Lidenal" OR "Lindafor" OR

Table B-3. Strategies to Augment the Literature Search

Source	Query and number screened when available
	"Lindagam" OR "Lindagrain" OR "Lindagranox" OR "Lindan" OR "Lindanum" OR "Lindapoudre" OR "Lindatox" OR "Lindosep" OR "Lintox" OR "Linvur" OR "Lorexane" OR "Mszycol" OR "Neo-Scabicidol" OR "Nexol E" OR "Nexol-E" OR "Nicochloran" OR "Novigam" OR "Omnitox" OR "Ovadziak" OR "Owadziak" OR "Pedraczak" OR "Pflanzol" OR "Quellada" OR "Technical HCH" OR "technical grade HCH" OR "Scabene" OR "Silvanol" OR "Spritz-Rapidin" OR "Spritzlindane" OR "Spruehpflanzol" OR "Streunex" (advanced); Limit to: Project Title, Project Terms, Project Abstracts Search Criteria: Fiscal Year: Active Projects; Text Search: "α-Benzohexachloride" OR "α-Hexachlorocyclohexane" OR "α-Lindane" OR "β-1,2,3,4,5,6-Hexachlorocyclohexane" OR "β-Benzene hexachloride" OR "β-Hexachlorocyclohexane" OR "β-Lindane" OR "γ-1,2,3,4,5,6-Hexachlorocyclohexane" OR "γ-Benzene hexachloride" OR "γ-Benzohexachloride" OR "γ-Hexachlorobenzene" OR "γ-Hexachlorocyclohexane" OR "γ-Lindane" OR "δ-Benzene hexachloride" OR "δ-Hexachlorocyclohexane" OR "δ-Lindane" OR "alpha BHC" OR "alpha-BHC" OR "alpha-HCH" OR "beta BHC" OR "beta-BHC" OR "beta-HCH" OR "delta BHC" OR "delta-BHC" OR "delta-HCH" OR "Gamma-BHC" OR "gamma-HCH" OR "Nexit Stark" OR "Nexit-stark" OR "α-BHC" OR "α-HCH" OR "β-666" OR "β-BHC" OR "β-HCH" OR "γ-666" OR "γ-BHC" OR "γ-HCH" OR "δ-BHC" OR "δ-HCH" OR "total BHC" OR "α-666" OR "δ-666" OR "BHC-gamma" OR "α-Benzenehexachloride" OR "Detox 25" OR "Dol Granule" OR "ENT 7,796" OR "TAP 85" OR "Ameisenmittel merck" OR "Arcotal S" OR "Bentox 10" OR "Benzene-1,2,3,4,5,6-hexachloride" OR "beta-Hexachlorobenzene" OR "Detmol Extract" OR "Fenoform forte" OR "Gamma-mean 400" OR "Geolin G 3" OR "Hungaria L7" OR "Mglawik L" OR "Milbol 49" OR "Nexen FB" OR "sang-gamma" OR "sang gamma" OR "Submar" OR "Verindal Ultra" OR "HCH, technical grade" OR "beta-Hexachlorobenzene" OR "β-Hexachlorobenzene" (advanced); Limit to: Project Title, Project Terms, Project Abstracts
Other	Identified throughout the assessment process

The 2023 results were:

- Number of records identified from PubMed, NTRL, and TOXCENTER (after duplicate removal): 673
- Number of records identified from other strategies: 18
- Total number of records to undergo literature screening: 691

B.1.2 Literature Screening

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

- 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.

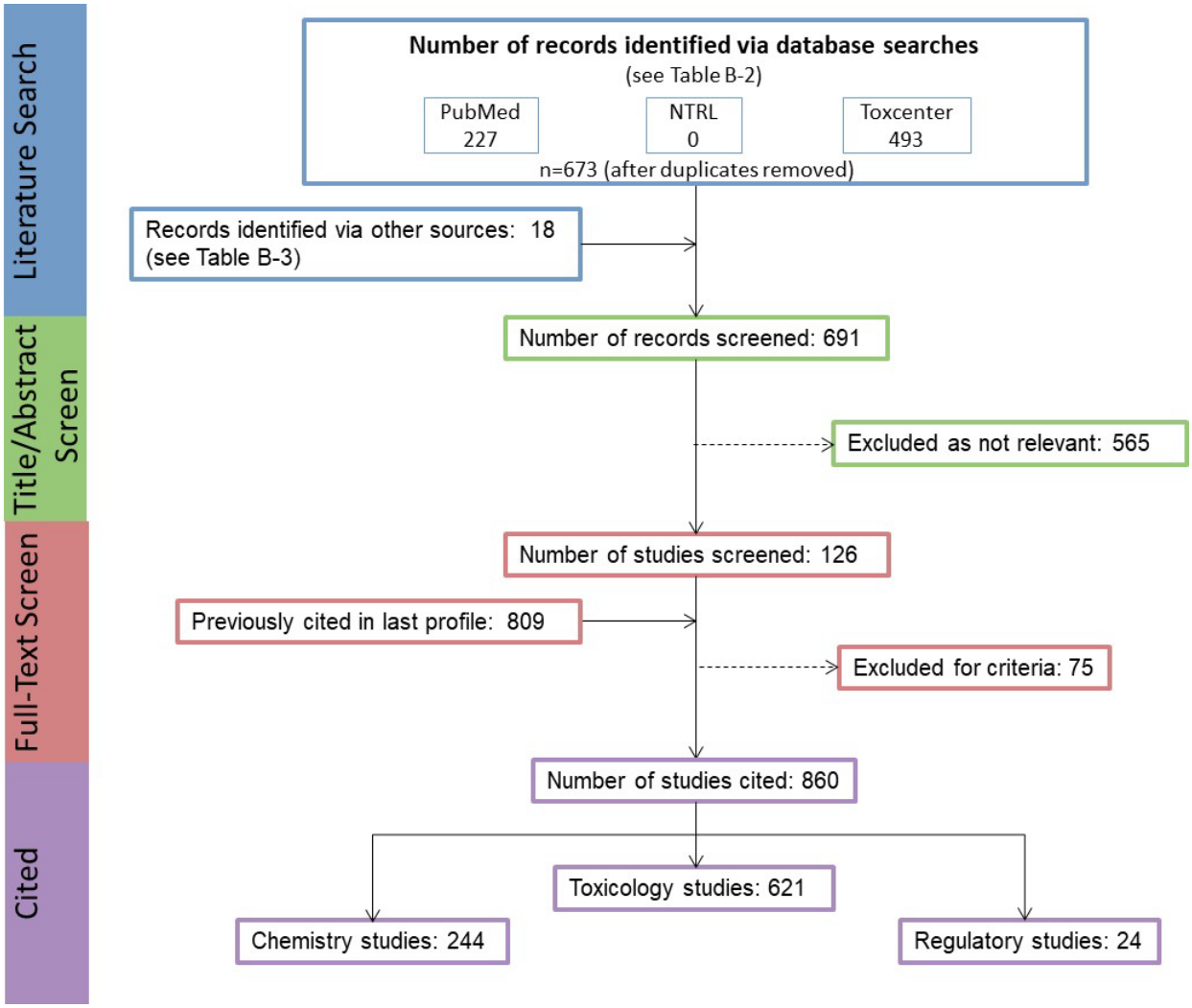
- Number of titles and abstracts screened: 691
- Number of studies considered relevant and moved to the next step: 129

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: 129
- Number of studies cited in the pre-public draft of the toxicological profile: 809
- Total number of studies cited in the profile: 860

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

Figure B-1. June 2023 Literature Search Results and Screen for HCH



APPENDIX C. FRAMEWORK FOR ATSDR'S SYSTEMATIC REVIEW OF HEALTH EFFECTS DATA FOR HCH

To increase the transparency of ATSDR's process of identifying, evaluating, synthesizing, and interpreting the scientific evidence on the health effects associated with exposure to HCH, ATSDR utilized a slight modification of NTP's Office of Health Assessment and Translation (OHAT) systematic review methodology (NTP 2013, 2015; Rooney et al. 2014). ATSDR's framework is an eight-step process for systematic review with the goal of identifying the potential health hazards of exposure to HCH:

- Step 1. Problem Formulation
- Step 2. Literature Search and Screen for Health Effects Studies
- Step 3. Extract Data from Health Effects Studies
- Step 4. Identify Potential Health Effect Outcomes of Concern
- Step 5. Assess the Risk of Bias for Individual Studies
- Step 6. Rate the Confidence in the Body of Evidence for Each Relevant Outcome
- Step 7. Translate Confidence Rating into Level of Evidence of Health Effects
- Step 8. Integrate Evidence to Develop Hazard Identification Conclusions

The systematic review for this profile is divided into four sections:

1. Steps 1, 2, and 3 for α-, β-, and γ-HCH (Sections C.1, C.2, and C.3)
2. Steps 4, 5, 6, 7, and 8 for α-HCH (Sections C.4, C.5, C.6, C.7, and C.8)
3. Steps 4, 5, 6, 7, and 8 for β-HCH (Sections C.9, C.10, C.11, C.12, and C.13)
4. Steps 4, 5, 6, 7, and 8 for γ-HCH (Sections C.14, C.15, C.16, C.17, and C.18)

C.1 PROBLEM FORMULATION

The objective of the toxicological profile and this systematic review was to identify the potential health hazards associated with inhalation, oral, or dermal/ocular exposure to HCH. The inclusion criteria used to identify relevant studies examining the health effects of HCH are presented in Table C-1.

Data from human and laboratory animal studies were considered relevant for addressing this objective. Human studies were divided into two broad categories: observational epidemiology studies and controlled exposure studies. The observational epidemiology studies were further divided: cohort studies (retrospective and prospective studies), population studies (with individual data or aggregate data), and case-control studies.

Table C-1. Inclusion Criteria for Identifying Health Effects Studies

Species
Human
Laboratory mammals
Route of exposure
Inhalation
Oral
Dermal (or ocular)
Parenteral (these studies will be considered supporting data)

Table C-1. Inclusion Criteria for Identifying Health Effects Studies

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
Cancer

C.2 LITERATURE SEARCH AND SCREEN FOR HEALTH EFFECTS STUDIES

A literature search and screen were conducted to identify studies examining the health effects of HCH. The literature search framework for the toxicological profile is discussed in detail in Appendix B.

C.2.1 Literature Search

As noted in Appendix B, the current literature search was intended to update the draft toxicological profile for HCH released for public comment in January 2023. See Appendix B for the databases searched and the search strategy.

A total of 691 records relevant to all sections of the toxicological profile were identified (after duplicate removal).

C.2.2 Literature Screening

As described in Appendix B, a two-step process was used to screen the literature search to identify relevant studies examining the health effects of HCH.

Title and Abstract Screen. In the Title and Abstract Screen step, 691 records were reviewed; 45 documents were considered to meet the health effects inclusion criteria in Table C-1 and were moved to the next step in the process.

Full Text Screen. In the second step in the literature screening process for the systematic review, a full text review of 299 health effect documents (documents identified in the update literature search and documents cited in older versions of the profile) was performed. From those 299 documents (446 studies), 64 documents (70 studies) were included in the qualitative review.

The epidemiological database for HCH is extensive. To facilitate the selection and inclusion of human studies of greater utility in assessing the hazards of HCH, only studies meeting the criteria below were included in the Toxicological Profile.

- Exposure was assessed for individuals, either using a biomarker or through detailed individual history (i.e., ecological study designs were excluded);
- The study presented an effect estimate specific to one or more HCH isomers;
- The statistical analysis of the association was multivariate, with consideration of at least one potential covariate. Studies limited to bivariate analyses (i.e., Pearson or Spearman correlation coefficients) were not included, nor were studies in which the analysis was limited to a comparison between blood concentrations in cases and controls;
- The health outcomes evaluated in the study were not mechanistic in nature (e.g., oxidative stress, genotoxicity);
- Case reports and case series were included if there was clear evidence of exposure to one or more HCH isomers.

C.3 EXTRACT DATA FROM HEALTH EFFECTS STUDIES

Relevant data extracted from the individual studies selected for inclusion in the systematic review were collected in customized data forms. A summary of the type of data extracted from each study is presented in Table C-2. For references that included more than one experiment or species, data extraction records were created for each experiment or species.

Table C-2. Data Extracted From Individual Studies

Citation
Chemical form
Route of exposure (e.g., inhalation, oral, dermal)
Specific route (e.g., gavage in oil, drinking water)
Species
Strain
Exposure duration category (e.g., acute, intermediate, chronic)
Exposure duration
Frequency of exposure (e.g., 6 hours/day, 5 days/week)
Exposure length
Number of animals or subjects per sex per group
Dose/exposure levels
Parameters monitored
Description of the study design and method
Summary of calculations used to estimate doses (if applicable)
Summary of the study results
Reviewer's comments on the study
Outcome summary (one entry for each examined outcome)

Table C-2. Data Extracted From Individual Studies

No-observed-adverse-effect level (NOAEL) value
Lowest-observed-adverse-effect level (LOAEL) value
Effect observed at the LOAEL value

A summary of the extracted data for each study is presented in the Supplemental Documents for HCH isomers and overviews of the results of the inhalation, oral, and dermal exposure studies are presented in Sections 2.2–2.18 of the profile and in the Levels Significant Exposures tables in Section 2.1 of the profile (Tables 2-1 through 2-2).

There was only a single study of δ-HCH, and inadequate data for derivation of an MRL, so a systematic review of the data for this isomer was not undertaken.

C.4 IDENTIFY POTENTIAL HEALTH EFFECT OUTCOMES OF CONCERN—α-HCH

Overviews of the potential health effect outcomes for α-HCH identified in human and animal studies are presented in Tables C-3 and C-4, respectively. There was a small number of human studies examining a handful of endpoints; the largest number of studies was devoted to developmental endpoints. The human studies used measures of α-HCH in blood or tissues to assess exposure, so the route is unknown; for the purpose of enumerations, these studies are considered to reflect oral exposure (e.g., through contaminated food). There were no inhalation or dermal animal studies of α-HCH, and very few oral studies. The available animal studies primarily examined liver effects and cancer. The most sensitive effect in animal studies were hepatic. Studies examining hepatic effects were carried through to Steps 4–8 of the systematic review. There were 70 studies (published in 64 documents) examining these potential outcomes carried through to Steps 4–8 of the systematic review. There were no human studies examining hepatic effects of α-HCH.

APPENDIX C

Table C-3. Overview of the Health Outcomes for α-Hexachlorocyclohexane Evaluated In Human Studies

	Body weight	Respiratory	Cardiovascular	Gastrointestinal	Hematological	Musculoskeletal	Hepatic	Renal	Dermal	Ocular	Endocrine	Immunological	Neurological	Reproductive	Developmental	Other Noncancer	Cancer
Inhalation studies																	
Cohort																	
Case control																	
Population																	
Case series																	
Oral studies																	
Cohort																	
Case control								2 1				1 1	3 2		4 3	2	5 3
Population								2			4 1	1		3 1	3 2	1	
Case series																	
Dermal studies																	
Cohort																	
Case control																	
Population																	
Case series																	

Number of studies examining endpoint	0	1	2	3	4	5–9	≥10
Number of studies reporting outcome	0	1	2	3	4	5–9	≥10

APPENDIX C

Table C-4. Overview of the Health Outcomes for α-Hexachlorocyclohexane Evaluated in Experimental Animal Studies

	Body weight	Respiratory	Cardiovascular	Gastrointestinal	Hematological	Musculoskeletal	Hepatic	Renal	Dermal	Ocular	Endocrine	Immunological[a]	Neurological[a]	Reproductive[a]	Developmental	Other Noncancer	Cancer
Inhalation studies																	
Acute-duration																	
Intermediate-duration																	
Chronic-duration																	
Oral studies																	
Acute-duration	1						4									1	
							1									1	
Intermediate-duration	1						10						2				6
							7										6
Chronic-duration	1						2	1									2
	1						2	1									2
Dermal studies																	
Acute-duration																	
Intermediate-duration																	
Chronic-duration																	

Number of studies examining endpoint	0	1	2	3	4	5–9	≥10
Number of studies reporting outcome	0	1	2	3	4	5–9	≥10

[a]Number of studies examining endpoint includes studies evaluating histopathology, but not evaluating function.

C.5 ASSESS THE RISK OF BIAS FOR INDIVIDUAL STUDIES—α-HCH

C.5.1 Risk of Bias Assessment—α-HCH

The risk of bias of individual studies was assessed using OHAT's Risk of Bias Tool (NTP 2015). The risk of bias questions for observational epidemiology studies, human-controlled exposure studies, and animal experimental studies are presented in Tables C-5, C-6, and C-7, respectively. Each risk of bias question was answered on a four-point scale:

- **Definitely low risk of bias (++)**
- **Probably low risk of bias (+)**
- **Probably high risk of bias (-)**
- **Definitely high risk of bias (– –)**

In general, "definitely low risk of bias" or "definitely high risk of bias" were used if the question could be answered with information explicitly stated in the study report. If the response to the question could be inferred, then "probably low risk of bias" or "probably high risk of bias" responses were typically used.

Table C-5. Risk of Bias Questionnaire for Observational Epidemiology Studies

Selection bias
Were the comparison groups appropriate?
Confounding bias
Did the study design or analysis account for important confounding and modifying variables?
Attrition/exclusion bias
Were outcome data complete without attrition or exclusion from analysis?
Detection bias
Is there confidence in the exposure characterization?
Is there confidence in outcome assessment?
Selective reporting bias
Were all measured outcomes reported?

Table C-6. Risk of Bias Questionnaire for Human-Controlled Exposure Studies

Selection bias
Was administered dose or exposure level adequately randomized?
Was the allocation to study groups adequately concealed?
Performance bias
Were the research personnel and human subjects blinded to the study group during the study?
Attrition/exclusion bias
Were outcome data complete without attrition or exclusion from analysis?
Detection bias
Is there confidence in the exposure characterization?
Is there confidence in outcome assessment?

Table C-6. Risk of Bias Questionnaire for Human-Controlled Exposure Studies

Selective reporting bias
Were all measured outcomes reported?

Table C-7. Risk of Bias Questionnaire for Experimental Animal Studies

Selection bias
Was administered dose or exposure level adequately randomized?
Was the allocation to study groups adequately concealed?
Performance bias
Were experimental conditions identical across study groups?
Were the research personnel blinded to the study group during the study?
Attrition/exclusion bias
Were outcome data complete without attrition or exclusion from analysis?
Detection bias
Is there confidence in the exposure characterization?
Is there confidence in outcome assessment?
Selective reporting bias
Were all measured outcomes reported?

After the risk of bias questionnaires were completed for the health effects studies, the studies were assigned to one of three risk of bias tiers based on the responses to the key questions listed below and the responses to the remaining questions.

- Is there confidence in the exposure characterization? (only relevant for observational studies)
- Is there confidence in the outcome assessment?
- Does the study design or analysis account for important confounding and modifying variables? (only relevant for observational studies)

First Tier. Studies placed in the first tier received ratings of "definitely low" or "probably low" risk of bias on the key questions **AND** received a rating of "definitely low" or "probably low" risk of bias on the responses to at least 50% of the other applicable questions.

Second Tier. A study was placed in the second tier if it did not meet the criteria for the first or third tiers.

Third Tier. Studies placed in the third tier received ratings of "definitely high" or "probably high" risk of bias for the key questions **AND** received a rating of "definitely high" or "probably high" risk of bias on the response to at least 50% of the other applicable questions.

The results of the risk of bias assessment for the α-HCH health effects studies (animal experimental studies) are presented in Table C-8. There were no observational epidemiology or human controlled experimental studies of α-HCH.

Table C-8. Summary of Risk of Bias Assessment for α-Hexachlorocyclohexane—Experimental Animal Studies

Reference	Risk of bias criteria and ratings										Risk of bias tier
	Selection bias		Performance bias		Attrition/ exclusion bias	Detection bias		Selective reporting bias	Other bias		
	Was administered dose or exposure level adequately randomized?	Was the allocation to study groups adequately concealed?	Were experimental conditions identical across study groups?	Were the research personnel blinded to the study group during the study?	Were outcome data complete without attrition or exclusion from analysis?	Is there confidence in the exposure characterization?	Is there confidence in the outcome assessment?*	Were all measured outcomes reported?	Did the study design or analysis account for important confounding and modifying variables?		
Outcome: Hepatic effects											
Oral intermediate exposure											
Sumida et al. 2007 (rat; 28 days)	–	–	+	+	–	+	+	++	NA		First
Ito et al. 1973 (mouse; 24 weeks)	–	–	+	+	+	++	+	++	NA		First
Ito et al. 1975 (rat; 24 or 48 weeks)	–	–	+	+	–	++	+	++	NA		First
Nagasaki et al. 1975 (rat; 24 weeks)	–	–	+	+	–	–	+	++	NA		Second
Nagasaki et al. 1975 (mouse; 24 weeks)	–	–	+	+	–	–	+	++	NA		Second
Nagasaki et al. 1975 (hamster; 24 weeks)	–	–	+	+	–	–	+	++	NA		Second
Tryphonas and Iverson 1983 (mouse; 50 weeks)	–	–	+	+	++	+	+	++	NA		First

Table C-8. Summary of Risk of Bias Assessment for α-Hexachlorocyclohexane—Experimental Animal Studies

Reference	Risk of bias criteria and ratings										Risk of bias tier
	Selection bias		Performance bias		Attrition/ exclusion bias	Detection bias		Selective reporting bias	Other bias		
	Was administered dose or exposure level adequately randomized?	Was the allocation to study groups adequately concealed?	Were experimental conditions identical across study groups?	Were the research personnel blinded to the study group during the study?	Were outcome data complete without attrition or exclusion from analysis?	Is there confidence in the exposure characterization?	Is there confidence in the outcome assessment?*	Were all measured outcomes reported?	Did the study design or analysis account for important confounding and modifying variables?		
Oral chronic exposure											
Fitzhugh et al. 1950 (rat; 72 weeks)	–	–	+	+	++	+	+	++	NA		First

++ = definitely low risk of bias; + = probably low risk of bias; – = probably high risk of bias; – – = definitely high risk of bias; na = not applicable

*Key question used to assign risk of bias tier

C.6 RATE THE CONFIDENCE IN THE BODY OF EVIDENCE FOR EACH RELEVANT OUTCOME—α-HCH

Confidence in the body of human and animal evidence were evaluated independently for each potential outcome. ATSDR did not evaluate the confidence in the body of evidence for carcinogenicity; rather, the Agency defaulted to the cancer weight-of-evidence assessment of other agencies including HHS, EPA, and IARC. The confidence in the body of evidence for an association or no association between exposure to HCH and a particular outcome was based on the strengths and weaknesses of individual studies. Four descriptors were used to describe the confidence in the body of evidence for effects or when no effect was found:

- **High confidence:** the true effect is highly likely to be reflected in the apparent relationship
- **Moderate confidence:** the true effect may be reflected in the apparent relationship
- **Low confidence:** the true effect may be different from the apparent relationship
- **Very low confidence:** the true effect is highly likely to be different from the apparent relationship

Confidence in the body of evidence for a particular outcome was rated for each type of study: case-control, case series, cohort, population, human-controlled exposure, and experimental animal. In the absence of data to the contrary, data for a particular outcome were collapsed across animal species, routes of exposure, and exposure durations. If species (or strain), route, or exposure duration differences were noted, then the data were treated as separate outcomes.

In the case of α-HCH, the only available type of study was experimental animal.

C.6.1 Initial Confidence Rating—α-HCH

In ATSDR's modification to the OHAT approach, the body of evidence for an association (or no association) between exposure to α-HCH and a particular outcome was given an initial confidence rating based on the key features of the individual studies examining that outcome. The presence of these key features of study design was determined for individual studies using four "yes or no" questions, which were customized for epidemiology, human controlled exposure, or experimental animal study designs. Separate questionnaires were completed for each outcome assessed in a study. The key features for observational epidemiology (cohort, population, and case-control) studies, human controlled exposure studies, and experimental animal studies are presented in Tables C-9, C-10, and C-11, respectively. The initial confidence in the study was determined based on the number of key features present in the study design:

- **High Initial Confidence:** Studies in which the responses to the four questions were "yes".
- **Moderate Initial Confidence:** Studies in which the responses to only three of the questions were "yes".
- **Low Initial Confidence:** Studies in which the responses to only two of the questions were "yes".
- **Very Low Initial Confidence:** Studies in which the response to one or none of the questions was "yes".

Table C-9. Key Features of Study Design for Observational Epidemiology Studies

Exposure was experimentally controlled
Exposure occurred prior to the outcome
Outcome was assessed on individual level rather than at the population level
A comparison group was used

Table C-10. Key Features of Study Design for Human Controlled Exposure Studies

A comparison group was used or the subjects served as their own control
A sufficient number of subjects were tested
Appropriate methods were used to measure outcomes (i.e., clinically-confirmed outcome versus self-reported)
Appropriate statistical analyses were performed and reported or the data were reported in such a way to allow independent statistical analysis

Table C-11. Key Features of Study Design for Experimental Animal Studies

A concurrent control group was used
A sufficient number of animals per group were tested
Appropriate parameters were used to assess a potential adverse effect
Appropriate statistical analyses were performed and reported or the data were reported in such a way to allow independent statistical analysis

The presence or absence of the key features and the initial confidence levels for studies of hepatic effects observed in the animal experimental studies are presented in Table C-12.

Table C-12. Presence of Key Features of Study Design for α-Hexachlorocyclohexane—Experimental Animal Studies

	Key feature				
Reference	Concurrent control group	Sufficient number of animals per group	Appropriate parameters to assess potential effect	Adequate data for statistical analysis	Initial study confidence
Outcome: Hepatic effects					
Oral intermediate exposure					
Sumida et al. 2007 (rat; 28 days)	Yes	No	Yes	Yes	Moderate
Ito et al. 1973 (mouse; 24 weeks)	Yes	Yes	Yes	Yes	High

Table C-12. Presence of Key Features of Study Design for α-Hexachlorocyclohexane—Experimental Animal Studies

Reference	Key feature: Concurrent control group	Key feature: Sufficient number of animals per group	Key feature: Appropriate parameters to assess potential effect	Key feature: Adequate data for statistical analysis	Initial study confidence
Ito et al. 1975 (rat; 24 or 48 weeks)	Yes	Yes	Yes	No	Moderate
Nagasaki et al. 1975 (rat; 24 weeks)	Yes	No	Yes	No	Low
Nagasaki et al. 1975 (mouse; 24 weeks)	Yes	Yes	Yes	No	Moderate
Nagasaki et al. 1975 (hamster; 24 weeks)	Yes	No	Yes	No	Low
Tryphonas and Iverson 1983 (mouse; 50 weeks)	Yes	Yes	Yes	Yes	High
Oral chronic exposure					
Fitzhugh et al. 1950 (rat; 72 weeks)	Yes	No	Yes	Yes	Moderate

A summary of the initial confidence ratings for each outcome is presented in Table C-13. If individual studies for a particular outcome and study type had different study quality ratings, then the highest confidence rating for the group of studies was used to determine the initial confidence rating for the body of evidence; any exceptions were noted in Table C-13.

Table C-13. Initial Confidence Rating for α-HCH Health Effects Studies

	Initial study confidence	Initial confidence rating
Outcome: Hepatic effects		
Oral intermediate exposure		
Animal studies		
Sumida et al. 2007 (rat; 28 days)	Moderate	High
Ito et al. 1973 (mouse; 24 weeks)	High	
Ito et al. 1975 (rat; 24 or 48 weeks)	Moderate	
Nagasaki et al. 1975 (rat; 24 weeks)	Low	
Nagasaki et al. 1975 (mouse; 24 weeks)	Moderate	
Nagasaki et al. 1975 (hamster; 24 weeks)	Low	
Tryphonas and Iverson 1983 (mouse; 50 weeks)	High	
Oral chronic exposure		
Animal studies		
Fitzhugh et al. 1950	Moderate	Moderate

C.6.2 Adjustment of the Confidence Rating—α-HCH

The initial confidence rating was then downgraded or upgraded depending on whether there were substantial issues that would decrease or increase confidence in the body of evidence. The nine properties of the body of evidence that were considered are listed below. The summaries of the assessment of the confidence in the body of evidence for hepatic effects are presented in Table C-14. If the confidence ratings for a particular outcome were based on more than one type of human study, then the highest confidence rating was used for subsequent analyses. An overview of the confidence in the body of evidence for all health effects associated with α-HCH exposure is presented in Table C-15.

Five properties of the body of evidence were considered to determine whether the confidence rating should be downgraded:

- **Risk of bias.** Evaluation of whether there is substantial risk of bias across most of the studies examining the outcome. This evaluation used the risk of bias tier groupings for individual studies examining a particular outcome. Below are the criteria used to determine whether the initial confidence in the body of evidence for each outcome should be downgraded for risk of bias:
    - No downgrade if most studies are in the risk of bias first tier
    - Downgrade one confidence level if most studies are in the risk of bias second tier
    - Downgrade two confidence levels if most studies are in the risk of bias third tier

- **Unexplained inconsistency.** Evaluation of whether there is inconsistency or large variability in the magnitude or direction of estimates of effect across studies that cannot be explained. Below are the criteria used to determine whether the initial confidence in the body of evidence for each outcome should be downgraded for unexplained inconsistency:
    - No downgrade if there is little inconsistency across studies or if only one study evaluated the outcome
    - Downgrade one confidence level if there is variability across studies in the magnitude or direction of the effect
    - Downgrade two confidence levels if there is substantial variability across studies in the magnitude or direct of the effect

- **Indirectness.** Evaluation of four factors that can affect the applicability, generalizability, and relevance of the studies:
    - Relevance of the animal model to human health—unless otherwise indicated, studies in rats, mice, and other mammalian species are considered relevant to humans
    - Directness of the endpoints to the primary health outcome—examples of secondary outcomes or nonspecific outcomes include organ weight in the absence of histopathology or clinical chemistry findings in the absence of target tissue effects
    - Nature of the exposure in human studies and route of administration in animal studies—inhalation, oral, and dermal exposure routes are considered relevant unless there are compelling data to the contrary
    - Duration of treatment in animal studies and length of time between exposure and outcome assessment in animal and prospective human studies—this should be considered on an outcome-specific basis

  Below are the criteria used to determine whether the initial confidence in the body of evidence for each outcome should be downgraded for indirectness:
    - No downgrade if none of the factors are considered indirect
    - Downgrade one confidence level if one of the factors is considered indirect

- Downgrade two confidence levels if two or more of the factors are considered indirect

- **Imprecision.** Evaluation of the narrowness of the effect size estimates and whether the studies have adequate statistical power. Data are considered imprecise when the ratio of the upper to lower 95% CIs for most studies is ≥10 for tests of ratio measures (e.g., odds ratios) and ≥100 for absolute measures (e.g., percent control response). Adequate statistical power is determined if the study can detect a potentially biologically meaningful difference between groups (20% change from control response for categorical data or risk ratio of 1.5 for continuous data). Below are the criteria used to determine whether the initial confidence in the body of evidence for each outcome should be downgraded for imprecision:
  - No downgrade if there are no serious imprecisions
  - Downgrade one confidence level for serious imprecisions
  - Downgrade two confidence levels for very serious imprecisions

- **Publication bias.** Evaluation of the concern that studies with statistically significant results are more likely to be published than studies without statistically significant results.
  - Downgrade one level of confidence for cases where there is serious concern with publication bias

Four properties of the body of evidence were considered to determine whether the confidence rating should be upgraded:

- **Large magnitude of effect.** Evaluation of whether the magnitude of effect is sufficiently large so that it is unlikely to have occurred as a result of bias from potential confounding factors.
  - Upgrade one confidence level if there is evidence of a large magnitude of effect in a few studies, provided that the studies have an overall low risk of bias and there is no serious unexplained inconsistency among the studies of similar dose or exposure levels; confidence can also be upgraded if there is one study examining the outcome, provided that the study has an overall low risk of bias

- **Dose response.** Evaluation of the dose-response relationships measured within a study and across studies. Below are the criteria used to determine whether the initial confidence in the body of evidence for each outcome should be upgraded:
  - Upgrade one confidence level for evidence of a monotonic dose-response gradient
  - Upgrade one confidence level for evidence of a non-monotonic dose-response gradient where there is prior knowledge that supports a non-monotonic dose-response and a non-monotonic dose-response gradient is observed across studies

- **Plausible confounding or other residual biases.** This factor primarily applies to human studies and is an evaluation of unmeasured determinants of an outcome such as residual bias towards the null (e.g., "healthy worker" effect) or residual bias suggesting a spurious effect (e.g., recall bias). Below is the criterion used to determine whether the initial confidence in the body of evidence for each outcome should be upgraded:
  - Upgrade one confidence level for evidence that residual confounding or bias would underestimate an apparent association or treatment effect (i.e., bias toward the null) or suggest a spurious effect when results suggest no effect

- **Consistency in the body of evidence.** Evaluation of consistency across animal models and species, consistency across independent studies of different human populations and exposure

scenarios, and consistency across human study types. Below is the criterion used to determine whether the initial confidence in the body of evidence for each outcome should be upgraded:

- Upgrade one confidence level if there is a high degree of consistency in the database

Table C-14. Adjustments to the Initial Confidence in the Body of Evidence

	Initial confidence	Adjustments to the initial confidence rating	Final confidence
Hepatic effects:			
Human studies	NA	NA	NA
Animal studies	High	+1 consistency, +1 magnitude	High

Table C-15. Confidence in the Body of Evidence for α-Hexachlorocyclohexane

	Confidence in body of evidence	
Outcome	Human studies	Animal studies
Hepatic	NA	High

C.7 TRANSLATE CONFIDENCE RATING INTO LEVEL OF EVIDENCE OF HEALTH EFFECTS—α-HCH

In the seventh step of the systematic review of the health effects data for HCH, the confidence in the body of evidence for specific outcomes was translated to a level of evidence rating. The level of evidence rating reflected the confidence in the body of evidence and the direction of the effect (i.e., toxicity or no toxicity); route-specific differences were noted. The level of evidence for health effects was rated on a five-point scale:

- **High level of evidence:** High confidence in the body of evidence for an association between exposure to the substance and the health outcome
- **Moderate level of evidence:** Moderate confidence in the body of evidence for an association between exposure to the substance and the health outcome
- **Low level of evidence:** Low confidence in the body of evidence for an association between exposure to the substance and the health outcome
- **Evidence of no health effect:** High confidence in the body of evidence that exposure to the substance is not associated with the health outcome
- **Inadequate evidence:** Low or moderate confidence in the body of evidence that exposure to the substance is not associated with the health outcome OR very low confidence in the body of evidence for an association between exposure to the substance and the health outcome

A summary of the level of evidence of health effects for HCH is presented in Table C-16.

Table C-16. Level of Evidence of Health Effects for α-Hexachlorocyclohexane

Outcome	Confidence in body of evidence	Direction of health effect	Level of evidence for health effect
Human studies		NA	
Animal studies			
Hepatic	High	Health Effect	High

C.8 INTEGRATE EVIDENCE TO DEVELOP HAZARD IDENTIFICATION CONCLUSIONS—α-HCH

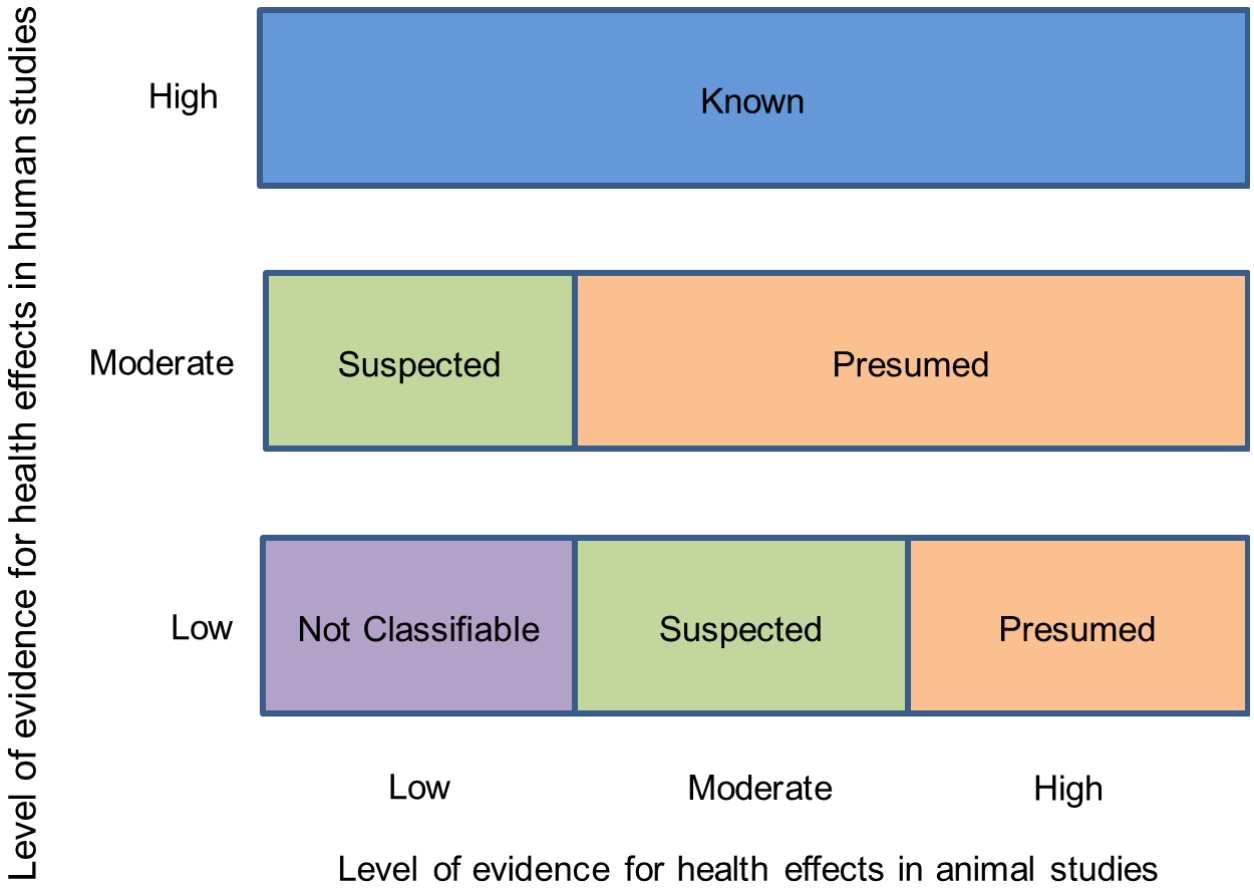
The final step involved the integration of the evidence streams for the human studies and animal studies to allow for a determination of hazard identification conclusions. For health effects, there were four hazard identification conclusion categories:

- **Known** to be a hazard to humans
- **Presumed** to be a hazard to humans
- **Suspected** to be a hazard to humans
- **Not classifiable** as to the hazard to humans

The initial hazard identification was based on the highest level of evidence in the human studies and the level of evidence in the animal studies; if there were no data for one evidence stream (human or animal), then the hazard identification was based on the one data stream (equivalent to treating the missing evidence stream as having low level of evidence). The hazard identification scheme is presented in Figure C-1 and described below:

- **Known:** A health effect in this category would have:
  - High level of evidence for health effects in human studies **AND** a high, moderate, or low level of evidence in animal studies.
- **Presumed:** A health effect in this category would have:
  - Moderate level of evidence in human studies **AND** high or moderate level of evidence in animal studies **OR**
  - Low level of evidence in human studies **AND** high level of evidence in animal studies
- **Suspected:** A health effect in this category would have:
  - Moderate level of evidence in human studies **AND** low level of evidence in animal studies **OR**
  - Low level of evidence in human studies **AND** moderate level of evidence in animal studies
- **Not classifiable:** A health effect in this category would have:
  - Low level of evidence in human studies **AND** low level of evidence in animal studies

Figure C-1. Hazard Identification Scheme



Other relevant data such as mechanistic or mode-of-action data were considered to raise or lower the level of the hazard identification conclusion by providing information that supported or opposed biological plausibility.

Two hazard identification conclusion categories were used when the data indicated that there may be no health effect in humans:

- **Not identified** to be a hazard in humans
- **Inadequate** to determine hazard to humans

If the human level of evidence conclusion of no health effect was supported by the animal evidence of no health effect, then the hazard identification conclusion category of "not identified" was used. If the human or animal level of evidence was considered inadequate, then a hazard identification conclusion category of "inadequate" was used. As with the hazard identification for health effects, the impact of other relevant data was also considered for no health effect data.

The hazard identification conclusions for α-HCH are listed below and summarized in Table C-17.

Presumed Health Effects

- Hepatic
  - No information on hepatic effects in humans exposed to α-HCH.
  - High evidence level in animals including increased liver weight and histopathological lesions after oral exposure to α-HCH (Fitzhugh et al. 1950; Ito et al. 1973, 1975; Nagasaki et al. 1975; Sumida et al. 2007; Tryphonas and Iverson 1983).
  - Plausible mechanism based on increased oxidative stress markers in livers of animals exposed to low oral doses *in vivo* (Barros et al. 1991).

Table C-17. Hazard Identification Conclusions for α-Hexachlorocyclohexane

Outcome	Hazard identification
Hepatic effects	Presumed health effect

C.9 IDENTIFY POTENTIAL HEALTH EFFECT OUTCOMES OF CONCERN—β-HCH

Overviews of the potential health effect outcomes for β-HCH identified in human and animal studies are presented in Tables C-18 and C-19, respectively. Human studies examined a wide range of outcomes, with more studies of endocrine (thyroid hormone levels) developmental outcomes, other noncancer endpoints (diabetes and metabolic perturbations) and cancer than other outcomes. The human studies used measures of β-HCH in blood or tissues to assess exposure, so the route is unknown; for the purpose of enumerations, these studies are considered to reflect oral exposure (e.g., through contaminated food). Animal studies are limited to oral exposures, and the endpoints examined were limited. The animal data show that the liver and nervous system are sensitive effects of exposure to β-HCH; studies examining these potential outcomes were carried through to Steps 4–8 of the systematic review. There were 15 studies (published in 15 documents) examining these potential outcomes carried through to Steps 4–8 of the systematic review.

Table C-18. Overview of the Health Outcomes for β-Hexachlorocyclohexane Evaluated in Human Studies

	Body weight	Respiratory	Cardiovascular	Gastrointestinal	Hematological	Musculoskeletal	Hepatic	Renal	Dermal	Ocular	Endocrine	Immunological	Neurological	Reproductive	Developmental	Other Noncancer	Cancer
Inhalation studies																	
Cohort																	
Case control																	
Population																	
Case series																	
Oral studies																	
Cohort	1 1		1 1										1	2 2	7 4	2 	3 1
Case cohort																1	1
Case control								2			1 1	1 1	5 4	4 4	9 4	7 5	25 11
Nested case control															1	1	1
Population	2 2	1	3 1				2 1	2			9 6	2 1	2 1	5 2	13 8	11 5	1 1
Case series																	
Dermal studies																	
Cohort																	
Case control																	
Population																	
Case series																	

Number of studies examining endpoint	0	1	2	3	4	5–9	≥10
Number of studies reporting outcome	0	1	2	3	4	5–9	≥10

Table C-19. Overview of the Health Outcomes for β-Hexachlorocyclohexane Evaluated in Experimental Animal Studies

	Body weight	Respiratory	Cardiovascular	Gastrointestinal	Hematological	Musculoskeletal	Hepatic	Renal	Dermal	Ocular	Endocrine	Immunological[a]	Neurological[a]	Reproductive[a]	Developmental	Other Noncancer	Cancer
Inhalation studies																	
Acute-duration																	
Intermediate-duration																	
Chronic-duration																	
Oral studies																	
Acute-duration	1				2		3	1				1	2			1	
							2	1					2			1	
Intermediate-duration	2				1		4	1				3	2	3			1
	1				1		4	1				3	2	3			
Chronic-duration	1						1	1									1
	1						1	1									1
Dermal studies																	
Acute-duration																	
Intermediate-duration																	
Chronic-duration																	

Number of studies examining endpoint	0	1	2	3	4	5–9	≥10
Number of studies reporting outcome	0	1	2	3	4	5–9	≥10

[a]Number of studies examining endpoint includes study evaluating histopathology, but not evaluating function.

C.10 ASSESS THE RISK OF BIAS FOR INDIVIDUAL STUDIES—β-HCH

C.10.1 Risk of Bias Assessment—β-HCH

The risk of bias of individual studies was assessed using OHAT's Risk of Bias Tool (NTP 2015). The risk of bias questions for observational epidemiology studies, human-controlled exposure studies, and animal experimental studies were presented above in Tables C-5, C-6, and C-7, respectively. As described in Section C.5.1, each risk of bias question was answered on a four-point scale and studies were assigned to one of three risk of bias tiers.

The results of the risk of bias assessment for the different types of β-HCH health effects studies (observational epidemiology and animal experimental studies) are presented in Tables C-20 and C-21, respectively.

Table C-20. Summary of Risk of Bias Assessment for β-Hexachlorocyclohexane—Observational Epidemiology Studies

Reference	Risk of bias criteria and ratings: Selection bias — Were the comparison groups appropriate?	Confounding bias — Did the study design or analysis account for important confounding and modifying variables?*	Attrition / exclusion bias — Were outcome data complete without attrition or exclusion from analysis?	Detection bias — Is there confidence in the exposure characterization?*	Detection bias — Is there confidence in the outcome assessment?*	Selective reporting bias — Were all measured outcomes reported?	Risk of bias tier
Outcome: Neurological effects							
Cohort							
Medehouenou et al. 2019	++	++	+	++	++	++	First
Case-control							
Petersen et al. 2008	+	–	++	++	+	++	Second
Richardson et al. 2009, 2011	–	+	++	++	++	++	First
Singh et al. 2012, 2013, 2014	+	+	++	++	++	++	First
Xu et al. 2022	+	+	++	++	++	++	First
Zhang et al. 2021	+	+	++	++	++	++	First
Cross-sectional							
Kim et al. 2015	++	++	+	++	++	++	First
Steenland et al. 2014	–	+	+	++	++	++	First

Table C-20. Summary of Risk of Bias Assessment for β-Hexachlorocyclohexane—Observational Epidemiology Studies

Reference	Risk of bias criteria and ratings							Risk of bias tier
	Selection bias	Confounding bias	Attrition / exclusion bias	Detection bias		Selective reporting bias		
	Were the comparison groups appropriate?	Did the study design or analysis account for important confounding and modifying variables?*	Were outcome data complete without attrition or exclusion from analysis?	Is there confidence in the exposure characterization?*	Is there confidence in the outcome assessment?*	Were all measured outcomes reported?		
Outcome: Hepatic effects								
Cross-sectional								
Arrebola et al. 2014	++	++	–	++	++	++		First
Freire et al. 2015	++	++	–	++	++	++		First

++ = definitely low risk of bias; + = probably low risk of bias; – = probably high risk of bias; – – = definitely high risk of bias; NA = not applicable

*Key question used to assign risk of bias tier

Table C-21. Summary of Risk of Bias Assessment for β-Hexachlorocyclohexane—Experimental Animal Studies

Reference	Risk of bias criteria and ratings									
	Selection bias		Performance bias		Attrition/ exclusion bias	Detection bias		Selective reporting bias	Other bias	
	Was administered dose or exposure level adequately randomized?	Was the allocation to study groups adequately concealed?	Were experimental conditions identical across study groups?	Were the research personnel blinded to the study group during the study?	Were outcome data complete without attrition or exclusion from analysis?	Is there confidence in the exposure characterization?	Is there confidence in the outcome assessment?*	Were all measured outcomes reported?	Did the study design or analysis account for important confounding and modifying variables?	Risk of bias tier
Outcome: Hepatic effects										
Oral intermediate exposure										
Van Velsen et al. 1986 (rat; 13 weeks)	–	–	+	+	++	++	+	++	NA	First
Hanada et al. 1973 (mouse; 32 weeks)	–	–	+	+	+	–	+	++	NA	First
Ito et al. 1973 (mouse; 24 weeks)	–	–	+	+	+	++	+	++	NA	First
Ito et al. 1975 (rat; 24–48 weeks)	–	–	+	+	–	++	+	++	NA	First
Oral chronic exposure										
Fitzhugh et al. 1950 (rat; 107 weeks)	–	–	+	+	++	+	+	++	NA	First

Table C-21. Summary of Risk of Bias Assessment for β-Hexachlorocyclohexane—Experimental Animal Studies

Reference	Risk of bias criteria and ratings									Risk of bias tier
	Selection bias		Performance bias		Attrition/ exclusion bias	Detection bias		Selective reporting bias	Other bias	
	Was administered dose or exposure level adequately randomized?	Was the allocation to study groups adequately concealed?	Were experimental conditions identical across study groups?	Were the research personnel blinded to the study group during the study?	Were outcome data complete without attrition or exclusion from analysis?	Is there confidence in the exposure characterization?	Is there confidence in the outcome assessment?*	Were all measured outcomes reported?	Did the study design or analysis account for important confounding and modifying variables?	
Outcome: Neurological effects										
Oral acute exposure										
Van Velsen et al. 1986 (rat; 2 weeks)	–	–	+	+	++	++	+	++	NA	First
Cornacoff et al. 1988 (mouse; 1 week)	–	–	+	+	–	++	+	++	NA	First
Oral intermediate exposure										
Muller et al. 1981 (rat; 30 days)	–	–	+	+	–	–	+	++	NA	Second

++ = definitely low risk of bias; + = probably low risk of bias; – = probably high risk of bias; – – = definitely high risk of bias; NA = not applicable

*Key question used to assign risk of bias tier

C.11 RATE THE CONFIDENCE IN THE BODY OF EVIDENCE FOR EACH RELEVANT OUTCOME—β-HCH

As discussed in greater detail in Section C.6, confidences in the bodies of human and animal evidence were evaluated independently for each potential outcome for each type of study: case-control, case series, cohort, population, human-controlled exposure, and experimental animal. In the absence of data to the contrary, data for a particular outcome were collapsed across animal species, routes of exposure, and exposure durations. If species (or strain), route, or exposure duration differences were noted, then the data were treated as separate outcomes.

C.11.1 Initial Confidence Rating—β-HCH

As discussed in greater detail in Section C.6.1, the body of evidence for an association (or no association) between exposure to β-HCH and a particular outcome was given an initial confidence rating based on the key features of the individual studies examining that outcome. Refer to Tables C-9, C-10, and C-11, respectively, for the key features for observational epidemiology (cohort, population, and case-control) studies, human controlled exposure studies, and experimental animal studies.

The presence or absence of the key features and the initial confidence levels for studies examining neurological and hepatic effects observed in the observational epidemiology and animal experimental studies are presented in Tables C-22 and C-23, respectively.

Table C-22. Presence of Key Features of Study Design for β-Hexachlorocyclohexane—Observational Epidemiology Studies

Reference	Key features				Initial study confidence
	Controlled exposure	Exposure prior to outcome	Outcomes assessed on an individual level	Comparison group	
Outcome: Neurological effects					
Cohort					
Medehouenou et al. 2019	No	Yes	Yes	Yes	Moderate
Case-control					
Petersen et al. 2008	No	No	Yes	Yes	Low
Richardson et al. 2009, 2011	No	No	Yes	Yes	Low
Singh et al. 2012, 2013, 2014	No	No	Yes	Yes	Low
Xu et al. 2022	No	No	Yes	Yes	Low
Zhang et al. 2021	No	No	Yes	Yes	Low
Cross-sectional					
Kim et al. 2015	No	No	Yes	Yes	Low
Steenland et al. 2014	No	No	Yes	Yes	Low
Outcome: Hepatic effects					
Cross-sectional					
Arrebola et al. 2014	No	No	Yes	Yes	Low
Freire et al. 2015	No	No	Yes	Yes	Low

Table C-23. Presence of Key Features of Study Design for β-Hexachlorocyclohexane—Experimental Animal Studies

Reference	Key feature: Concurrent control group	Key feature: Sufficient number of animals per group	Key feature: Appropriate parameters to assess potential effect	Key feature: Adequate data for statistical analysis	Initial study confidence
Outcome: Hepatic effects					
Oral intermediate exposure					
Van Velsen et al. 1986 (rat; 13 weeks)	Yes	Yes	Yes	Yes	High
Hanada et al. 1973 (mouse; 32 weeks)	Yes	Yes	Yes	No	Moderate
Ito et al. 1973 (mouse; 24 weeks)	Yes	Yes	Yes	Yes	High
Ito et al. 1975 (rat; 48 weeks)	Yes	Yes	Yes	No	Moderate
Oral chronic exposure					
Fitzhugh et al. 1950 (rat; 107 weeks)	Yes	No	Yes	Yes	Moderate
Outcome: Neurological effects					
Oral acute exposure					
Van Velsen et al. 1986 (rat; 2 weeks)	Yes	Yes	Yes	No	Moderate
Cornacoff et al. 1988 (mouse; 1 week)	Yes	Yes	Yes	No	Moderate
Oral intermediate exposure					
Muller et al. 1981 (rat; 30 days)	Yes	Yes	Yes	Yes	High

A summary of the initial confidence ratings for each outcome is presented in Table C-24. If individual studies for a particular outcome and study type had different study quality ratings, then the highest confidence rating for the group of studies was used to determine the initial confidence rating for the body of evidence; any exceptions were noted in Table C-24.

Table C-24. Initial Confidence Rating for β-Hexachlorocyclohexane Health Effects Studies

	Initial study confidence	Initial confidence rating
Outcome: Neurological effects		
Oral acute exposure		
Animal studies		
Van Velsen et al. 1986	Moderate	Moderate
Cornacoff et al. 1988	Moderate	
Oral intermediate exposure		
Animal studies		
Muller et al. 1981	High	High

Table C-24. Initial Confidence Rating for β-Hexachlorocyclohexane Health Effects Studies

	Initial study confidence	Initial confidence rating
Oral chronic exposure		
Human studies		
Medehouenou et al. 2019	Moderate	Moderate
Singh et al. 2012, 2013, 2014	Low	
Petersen et al. 2008	Low	
Richardson et al. 2009, 2011	Low	
Kim et al. 2015	Low	
Steenland et al. 2014	Low	
Xu et al. 2022	Low	
Zhang et al. 2021	Low	
Outcome: Hepatic effects		
Oral intermediate exposure		
Animal studies		
Van Velsen et al. 1986 (rat; 13 weeks)	High	High
Hanada et al. 1973 (mouse; 32 weeks)	Moderate	
Ito et al. 1973 (mouse; 24 weeks)	High	
Ito et al. 1975 (rat; 48 weeks)	Moderate	
Oral chronic exposure		
Human studies		
Arrebola et al. 2014	Low	Low
Freire et al. 2015	Low	
Animal studies		
Fitzhugh et al. 1950	Moderate	Moderate

C.11.2 Adjustment of the Confidence Rating—β-HCH

The initial confidence rating was then downgraded or upgraded depending on whether there were substantial issues that would decrease or increase confidence in the body of evidence. The five properties of the body of evidence that were considered to determine whether the confidence rating should be downgraded and the four properties of the body of evidence that were considered to determine whether the confidence rating should be upgraded are described above in Section C.6.2. The summaries of the assessment of the confidence in the body of evidence for neurological and hepatic effects are presented in Table C-25. If the confidence ratings for a particular outcome were based on more than one type of human study, then the highest confidence rating was used for subsequent analyses. An overview of the confidence in the body of evidence for all health effects associated with β-HCH exposure is presented in Table C-26.

Table C-25. Adjustments to the Initial Confidence in the Body of Evidence

	Initial confidence	Adjustments to the initial confidence rating	Final confidence
Neurological effects			
Human studies	Moderate	+1 consistency, -1 indirectness	Moderate
Animal studies	High	-1 indirectness, +1 dose-response, +1 consistency	High
Hepatic effects			
Human studies	Low	-1 indirectness	Very low
Animal studies	High	+1 dose-response, +1 consistency	High

Table C-26. Confidence in the Body of Evidence for β-Hexachlorocyclohexane

	Confidence in body of evidence	
Outcome	Human studies	Animal studies
Neurological	Moderate	High
Hepatic	Very Low	High

C.12 TRANSLATE CONFIDENCE RATING INTO LEVEL OF EVIDENCE OF HEALTH EFFECTS—β-HCH

As described in Section C.7, the confidence in the body of evidence for specific outcomes was translated to a level of evidence rating. The level of evidence rating reflected the confidence in the body of evidence and the direction of the effect (i.e., toxicity or no toxicity); route-specific differences were noted.

A summary of the level of evidence of health effects for β-HCH is presented in Table C-27.

Table C-27. Level of Evidence of Health Effects for β-Hexachlorocyclohexane

Outcome	Confidence in body of evidence	Direction of health effect	Level of evidence for health effect
Human studies			
Neurological	Moderate	Health effect	Moderate
Hepatic	Very Low	No health effect	Inadequate
Animal studies			
Neurological	High	Health effect	High
Hepatic	High	Health effect	High

C.13 INTEGRATE EVIDENCE TO DEVELOP HAZARD IDENTIFICATION CONCLUSIONS– β-HCH

The final step involved the integration of the evidence streams for the human studies and animal studies to allow for a determination of hazard identification conclusions. Refer to Section C.8 for the four hazard identification conclusion categories for health effects, the hazard characterization scheme (see Figure C-1), and the hazard identification conclusion categories.

The hazard identification conclusions for β-HCH are listed below and summarized in Table C-28.

Presumed Health Effects

- Neurological
  - Moderate level of evidence in humans based on case-control studies reporting associations between serum β-HCH and risk of Parkinson and Alzheimer diseases (Petersen et al. 2008; Richardson et al. 2009, 2011; Singh et al. 2012, 2013, 2014; Xu et al. 2022) and a cross-sectional study showing an association between risk of cognitive deficits and β-HCH in blood (Kim et al. 2015).
  - High level of evidence in animals exposed orally based on clinical signs of neurotoxicity in rats and mice after acute durations (Cornacoff et al. 1988; Van Velsen et al. 1986) and reduced nerve conduction velocity in rats exposed for an intermediate duration (Muller et al. 1981). Clinical signs showed dose-related increase in severity.
  - Supported by evidence for neurological effects of γ-HCH in humans and animals (see Section 2.15).
- Hepatic
  - Very low level of evidence in humans based on two cross-sectional studies reporting no association between serum or adipose levels of β-HCH and hepatic clinical chemistry endpoints except for increased serum bilirubin in females (Arrebola et al. 2014; Freire et al. 2015).
  - High level of evidence in animals based on liver weight and histopathology changes in rats and mice exposed by dietary administration for intermediate and chronic durations (Fitzhugh et al. 1950; Hanada et al. 1973; Ito et al. 1973, 1975; Van Velsen et al. 1986).

Table C-28. Hazard Identification Conclusions for – β-HCHs

Outcome	Hazard identification
Neurological	Presumed health effect
Hepatic	Presumed health effect

C.14 IDENTIFY POTENTIAL HEALTH EFFECT OUTCOMES OF CONCERN—γ-HCH

Overviews of the potential health effect outcomes for γ-HCH identified in human and animal studies are presented in Tables C-29 and C-30, respectively. Most of the human studies evaluated developmental, reproductive, renal, endocrine, or cancer endpoints. Most of human studies of noncancer endpoints used measures of γ-HCH in blood or tissues to assess exposure, so the route is unknown; for the purpose of enumerations, these studies are considered to reflect oral exposure (e.g., through contaminated food). Studies of occupational exposure via pesticide application are considered to reflect primarily inhalation exposure. Most of the animal studies used oral administration, and the available studies examined comprehensive noncancer and cancer endpoints. The effects seen at the lowest doses in the animal studies were developmental and immune system effects. Studies examining these potential outcomes

were carried through to Steps 4–8 of the systematic review. There were 41 studies (published in 35 documents) examining these potential outcomes carried through to Steps 4–8 of the systematic review.

APPENDIX C

Table C-29. Overview of the Health Outcomes for γ-Hexachlorocyclohexane Evaluated In Human Studies

	Body weight	Respiratory	Cardiovascular	Gastrointestinal	Hematological	Musculoskeletal	Hepatic	Renal	Dermal	Ocular	Endocrine	Immunological	Neurological	Reproductive	Developmental	Other Noncancer	Caner
Inhalation studies																	
Cohort											1						1
											1						1
Case control																	
Population																	
Case series																	
Oral studies																	
Cohort								2				1	2	1	3		3
													2	1	1		1
Case cohort																	
Case control								2				1	2	2	6	3	20
								1				1	1	1	4	1	8
Population		1						2			4	2		4	2	2	
		1									1	1		2	1	1	
Case series																	
Meta analysis																	1
Dermal studies																	
Cohort																	
Case control																	
Population																	
Case series																	

Number of studies examining endpoint	0	1	2	3	4	5–9	≥10	
Number of studies reporting outcome	0	1	2	3	4	5–9	≥10	

APPENDIX C

Table C-30. Overview of the Health Outcomes for γ-Hexachlorocyclohexane Evaluated in Experimental Animal Studies

	Body weight	Respiratory	Cardiovascular	Gastrointestinal	Hematological	Musculoskeletal	Hepatic	Renal	Dermal	Ocular	Endocrine	Immunological[a]	Neurological[a]	Reproductive[a]	Developmental	Other Noncancer	Caner
Inhalation studies																	
Acute-duration							1	1					2				
													2				
Intermediate-duration	2	2			1		1	1		1							
Chronic-duration																	
Oral studies																	
Acute-duration	12	2	2	2	2		18	4			2	3	19	11	22		
	2				2		11	2				3	17	5	16		
Intermediate-duration	7	2	4		3		19	10			2	8	13	16	12	2	2
		1	3				15	7			2	7	12	13	9	2	1
Chronic-duration	3				1		3	2					1	1			4
	1						3	1					1	1			2
Dermal studies																	
Acute-duration	2	1							1	1			2				
	0	1								1			2				
Intermediate-duration	1	1					1	1	2			1	1				
	0	1					1	1	1				1				
Chronic-duration																	

Number of studies examining endpoint	0	1	2	3	4	5–9	≥10
Number of studies reporting outcome	0	1	2	3	4	5–9	≥10

[a]Number of studies examining endpoint includes study evaluating histopathology, but not evaluating function.

C.15 ASSESS THE RISK OF BIAS FOR INDIVIDUAL STUDIES—γ-HCH

C.15.1 Risk of Bias Assessment—γ-HCH

The risk of bias of individual studies was assessed using OHAT's Risk of Bias Tool (NTP 2015). The risk of bias questions for observational epidemiology studies, human-controlled exposure studies, and animal experimental studies were presented above in Tables C-5, C-6, and C-7, respectively. As described in Section C.5.1, each risk of bias question was answered on a four-point scale and studies were assigned to one of three risk of bias tiers.

The results of the risk of bias assessment for the different types of γ-HCH health effects studies (observational epidemiology and animal experimental studies) are presented in Tables C-31 and C-32, respectively.

Table C-31. Summary of Risk of Bias Assessment for γ-Hexachlorocyclohexane—Observational Epidemiology Studies

Reference	Risk of bias criteria and ratings						Risk of bias tier
	Selection bias	Confounding bias	Attrition / exclusion bias	Detection bias		Selective reporting bias	
	Were the comparison groups appropriate?	Did the study design or analysis account for important confounding and modifying variables?*	Were outcome data complete without attrition or exclusion from analysis?	Is there confidence in the exposure characterization?*	Is there confidence in the outcome assessment?*	Were all measured outcomes reported?	
Outcome: Developmental effects							
Cohort							
Fenster et al. 2006	++	+	–	+	++	++	First
Yang et al. 2021a	++	+	+	++	++	++	First
Garcia- Villarino et al. 2022	++	++	+	++	++	++	First
Case-control							
Fernandez et al. 2007	++	++	+	++	++	++	First
Mustafa et al. 2013	+	+	+	++	+	++	First
Sharma et al. 2012	+	+	+	++	++	++	First
Siddiqui et al. 2003	+	+	+	++	+	++	First
Yang et al. 2021b	++	+	+	++	++	++	First
Yin et al. 2021	++	+	+	++	++	++	First
Cross-sectional							
Fang et al. 2019a, 2019b	++	+	++	++	+	++	First
Freire et al. 2011	++	+	–	++	–	++	Second

APPENDIX C

Table C-31. Summary of Risk of Bias Assessment for γ-Hexachlorocyclohexane—Observational Epidemiology Studies

Reference	Risk of bias criteria and ratings						Risk of bias tier
	Selection bias	Confounding bias	Attrition / exclusion bias	Detection bias		Selective reporting bias	
	Were the comparison groups appropriate?	Did the study design or analysis account for important confounding and modifying variables?*	Were outcome data complete without attrition or exclusion from analysis?	Is there confidence in the exposure characterization?*	Is there confidence in the outcome assessment?*	Were all measured outcomes reported?	
Outcome: Immunological effects							
Cohort							
Landgren et al. 2009	++	++	++	+	++	++	First
Case-control							
Meng et al. 2016	+	–	++	+	++	++	Second

++ = definitely low risk of bias; + = probably low risk of bias; – = probably high risk of bias; – – = definitely high risk of bias; NA = not applicable

*Key question used to assign risk of bias tier

Table C-32. Summary of Risk of Bias Assessment for γ-Hexachlorocyclohexane—Experimental Animal Studies

Reference	Risk of bias criteria and ratings									
	Selection bias		Performance bias		Attrition/ exclusion bias	Detection bias		Selective reporting bias	Other bias	
	Was administered dose or exposure level adequately randomized?	Was the allocation to study groups adequately concealed?	Were experimental conditions identical across study groups?	Were the research personnel blinded to the study group during the study?	Were outcome data complete without attrition or exclusion from analysis?	Is there confidence in the exposure characterization?	Is there confidence in the outcome assessment?*	Were all measured outcomes reported?	Did the study design or analysis account for important confounding and modifying variables?	Risk of bias tier
Outcome: Developmental effects										
Oral acute exposure										
Dalsenter et al. 1997a (rat; once)	–	–	+	+	++	–	+	++	NA	First
Dalsenter et al. 1997b (rat; once)	–	–	+	+	–	+	+	++	NA	First
Dalsenter et al. 1997b (rat; LDs 8–14)	–	–	+	+	–	+	+	++	NA	First
Johri et al. 2008 (rat; once)	–	–	+	+	–	–	+	++	NA	Second
Khera et al. 1979 (rat; GDs 6–15)	–	–	+	+	+	+	+	++	NA	First
Palmer et al. 1978 (rat; GDs 6–15)	+	+	+	+	+	–	+	++	NA	First
Rivera et al. 1991 (rat; once)	+	+	+	+	–	+	+	++	NA	First
Rivera et al. 1998 (rat; once)	+	+	+	+	–	–	+	++	NA	First

Table C-32. Summary of Risk of Bias Assessment for γ-Hexachlorocyclohexane—Experimental Animal Studies

Reference	Risk of bias criteria and ratings									
	Selection bias		Performance bias		Attrition/ exclusion bias	Detection bias		Selective reporting bias	Other bias	
	Was administered dose or exposure level adequately randomized?	Was the allocation to study groups adequately concealed?	Were experimental conditions identical across study groups?	Were the research personnel blinded to the study group during the study?	Were outcome data complete without attrition or exclusion from analysis?	Is there confidence in the exposure characterization?	Is there confidence in the outcome assessment?*	Were all measured outcomes reported?	Did the study design or analysis account for important confounding and modifying variables?	Risk of bias tier
Rivera et al. 1998 (rat; PNDs 8–14)	–	–	+	+	–	–	–	++	NA	Third
Serrano et al. 1990 (rat; PNDs 8–10)	+	+	+	+	–	–	+	++	NA	First
Di Consiglio et al. 2009 (mouse; GDs 9–16)	+	+	+	+	+	–	+	++	NA	First
Hassoun and Stohs 1996a (mouse, once)	–	–	+	+	++	–	+	++	NA	First
La Sala et al. 2009 (mouse; 3 days)	+	+	+	+	–	–	+	++	NA	First
Maranghi et al. 2007 (mouse, GDs 91–6)	+	+	+	+	++	–	+	++	NA	First
Traina et al. 2003 (mouse; GDs 9–16)	+	+	+	+	++	–	+	++	NA	First
Palmer et al. 1978 (rabbit; GDs 6–18)	+	+	+	+	+	–	+	++	NA	First
Oral intermediate exposure										
Breton et al. 2005 (rat; ~21 weeks (2-generation, premating–PND 98)	–	–	+	+	–	–	+	++	NA	First

Table C-32. Summary of Risk of Bias Assessment for γ-Hexachlorocyclohexane—Experimental Animal Studies

Reference	Risk of bias criteria and ratings									
	Selection bias		Performance bias		Attrition/ exclusion bias	Detection bias		Selective reporting bias	Other bias	
	Was administered dose or exposure level adequately randomized?	Was the allocation to study groups adequately concealed?	Were experimental conditions identical across study groups?	Were the research personnel blinded to the study group during the study?	Were outcome data complete without attrition or exclusion from analysis?	Is there confidence in the exposure characterization?	Is there confidence in the outcome assessment?*	Were all measured outcomes reported?	Did the study design or analysis account for important confounding and modifying variables?	Risk of bias tier
EPA 1991a (rat; 2-generation, 70 days prior to mating until sacrifice)	+	+	+	+	+	+	++	++	NA	First
EPA 1999c (rat; GD 6–LD 10)	–	–	+	+	++	++	+	++	NA	First
Johri et al. 2007 (rat; GDs 521)	+	+	+	+	–	–	+	++	NA	First
Johri et al. 2008 (rat; GDs 5–21 and PND 45)	+	+	+	+	–	–	+	++	NA	First
Matsuura et al. 2005 (rat; ~10 weeks (2-generation; premating–PND 21))	++	+	+	+	+	++	+	++	NA	First
Sauviat et al. 2005 (rat; ~13 weeks)	–	–	+	+	–	–	+	++	NA	Second
Srinivasan et al. 1991 (rat; GDs 0–21, LDs 1–28)	–	–	+	+	+	–	+	++	NA	First

Table C-32. Summary of Risk of Bias Assessment for γ-Hexachlorocyclohexane—Experimental Animal Studies

Reference	Risk of bias criteria and ratings: Selection bias: Was administered dose or exposure level adequately randomized?	Selection bias: Was the allocation to study groups adequately concealed?	Performance bias: Were experimental conditions identical across study groups?	Performance bias: Were the research personnel blinded to the study group during the study?	Attrition/ exclusion bias: Were outcome data complete without attrition or exclusion from analysis?	Detection bias: Is there confidence in the exposure characterization?	Detection bias: Is there confidence in the outcome assessment?*	Selective reporting bias: Were all measured outcomes reported?	Other bias: Did the study design or analysis account for important confounding and modifying variables?	Risk of bias tier
Srivastava et al. 2019 (rat, GDs 5–21)	–	–	+	+	–	–	–	++	NA	Third
Seiler et al. 1994 (rabbit; 12–15 weeks, 3 days/week)	–	–	+	+	–	+	+	++	NA	First
Outcome: Immunological effects										
Oral acute exposure										
Mediratta et al. 2008 (rat; 14 days)	+	+	+	+	++	–	+	++	NA	First
Hong and Boorman 1993 (mouse; 10 days)	+	+	+	+	–	–	+	++	NA	First
Hong and Boorman 1993 (mouse; 3 days)	+	+	+	+	–	–	+	++	NA	First
Oral intermediate exposure										
Koner et al. 1998 (rat; 8 weeks)	+	+	+	+	–	++	+	++	NA	First

Table C-32. Summary of Risk of Bias Assessment for γ-Hexachlorocyclohexane—Experimental Animal Studies

Reference	Risk of bias criteria and ratings									Risk of bias tier
	Selection bias		Performance bias		Attrition/ exclusion bias	Detection bias		Selective reporting bias	Other bias	
	Was administered dose or exposure level adequately randomized?	Was the allocation to study groups adequately concealed?	Were experimental conditions identical across study groups?	Were the research personnel blinded to the study group during the study?	Were outcome data complete without attrition or exclusion from analysis?	Is there confidence in the exposure characterization?	Is there confidence in the outcome assessment?*	Were all measured outcomes reported?	Did the study design or analysis account for important confounding and modifying variables?	
Mediratta et al. 2008 (rat; 21 days)	+	+	+	+	++	–	+	++	NA	First
Meera et al. 1992 (mouse; 24 weeks)	–	–	+	+	–	+	+	++	NA	First

++ = definitely low risk of bias; + = probably low risk of bias; – = probably high risk of bias; – – = definitely high risk of bias; NA = not applicable

*Key question used to assign risk of bias tier

C.16 RATE THE CONFIDENCE IN THE BODY OF EVIDENCE FOR EACH RELEVANT OUTCOME—γ-HCH

As discussed in greater detail in Section C.6, confidences in the bodies of human and animal evidence were evaluated independently for each potential outcome for each type of study: case-control, case series, cohort, population, human-controlled exposure, and experimental animal. In the absence of data to the contrary, data for a particular outcome were collapsed across animal species, routes of exposure, and exposure durations. If species (or strain), route, or exposure duration differences were noted, then the data were treated as separate outcomes.

C.16.1 Initial Confidence Rating—γ-HCH

As discussed in greater detail in Section C.6.1, the body of evidence for an association (or no association) between exposure to γ-HCH and a particular outcome was given an initial confidence rating based on the key features of the individual studies examining that outcome. Refer to Tables C-9, C-10, and C-11, respectively, for the key features for observational epidemiology (cohort, population, and case-control) studies, human controlled exposure studies, and experimental animal studies.

The presence or absence of the key features and the initial confidence levels for studies examining developmental and immune system effects in the observational epidemiology and animal experimental studies are presented in Tables C-33 and C-34, respectively.

Table C-33. Presence of Key Features of Study Design for γ-Hexachlorocyclohexane—Observational Epidemiology Studies

Reference	Key features				Initial study confidence
	Controlled exposure	Exposure prior to outcome	Outcomes assessed on an individual.	Comparison group	
Outcome: Developmental effects					
Cohort					
Fenster et al. 2006	No	Yes	Yes	Yes	Moderate
Yang et al. 2021a	No	Yes	Yes	Yes	Moderate
Garcia- Villarino et al. 2022	No	Yes	Yes	Yes	Moderate
Case-control					
Fernandez et al. 2007	No	No	Yes	Yes	Low
Mustafa et al. 2013	No	No	Yes	Yes	Low
Sharma et al. 2012	No	No	Yes	Yes	Low
Siddiqui et al. 2003	No	No	Yes	Yes	Low
Yang et al. 2021b	No	No	Yes	Yes	Low
Yin et al. 2021	No	No	Yes	Yes	Low
Cross-sectional					
Fang et al. 2019a, 2019b	No	No	Yes	Yes	Low

Table C-33. Presence of Key Features of Study Design for γ-Hexachlorocyclohexane—Observational Epidemiology Studies

Reference	Key features				Initial study confidence
	Controlled exposure	Exposure prior to outcome	Outcomes assessed on an individual.	Comparison group	
Outcome: Immunological effects					
Cohort					
Landgren et al. 2009	No	Yes	Yes	Yes	Moderate
Case-control					
Meng et al. 2016	No	No	Yes	Yes	Low
Cross-sectional					
Wang et al. 2021a	No	No	Yes	Yes	Low

Table C-34. Presence of Key Features of Study Design for γ-Hexachlorocyclohexane—Experimental Animal Studies

Reference	Key feature				Initial study confidence
	Concurrent control group	Sufficient number of animals per group	Appropriate parameters to assess potential effect	Adequate data for statistical analysis	
Outcome: Developmental effects					
Oral acute exposure					
Dalsenter et al. 1997a (rat; once)	Yes	Yes	Yes	Yes	High
Dalsenter et al. 1997b (rat; once)	Yes	Yes	Yes	Yes	High
Dalsenter et al. 1997b (rat; LDs 8–14)	Yes	Yes	Yes	Yes	High
Johri et al. 2008 (rat; once)	Yes	Yes	Yes	Yes	High
Khera et al. 1979 (rat; GDs 6–15)	Yes	Yes	Yes	Yes	High
Palmer et al. 1978 (rat; GDs 6–15)	Yes	Yes	Yes	Yes	High
Rivera et al. 1991 (rat; once)	Yes	No	Yes	Yes	Moderate
Rivera et al. 1998 (rat; once)	Yes	No	Yes	Yes	Moderate
Rivera et al. 1998 (rat; PNDs 8–14)	Yes	No	Yes	Yes	Moderate
Serrano et al. 1990 (rat; PNDs 8–10)	Yes	Yes	Yes	No	Moderate
Di Consiglio et al. 2009 (mouse; GDs 9–16)	Yes	No	Yes	Yes	Moderate
Hassoun and Stohs 1996a (mouse, once)	Yes	Yes	Yes	Yes	High
La Sala et al. 2009 (mouse; 3 days)	Yes	No	Yes	Yes	Moderate

Table C-34. Presence of Key Features of Study Design for γ-Hexachlorocyclohexane—Experimental Animal Studies

Reference	Key feature: Concurrent control group	Key feature: Sufficient number of animals per group	Key feature: Appropriate parameters to assess potential effect	Key feature: Adequate data for statistical analysis	Initial study confidence
Maranghi et al. 2007 (mouse, GDs 91–6)	Yes	Yes	Yes	Yes	High
Traina et al. 2003 (mouse; GDs 9–16)	Yes	Yes	Yes	Yes	High
Palmer et al. 1978 (rabbit; GDs 6–18)	Yes	Yes	Yes	Yes	High
Oral intermediate exposure					
Breton et al. 2005 (rat; ~21 weeks, 2-generation, premating–PND 98)	Yes	No	Yes	Yes	Moderate
EPA 1991a (rat; 2-generation, 70 days prior to mating until sacrifice)	Yes	Yes	Yes	Yes	High
EPA 1999c (rat; GD 6–LD 10)	Yes	Yes	Yes	Yes	High
Johri et al. 2007 (rat; GDs 5–21)	Yes	Yes	Yes	Yes	High
Johri et al. 2008 (rat; GDs 5–21 and PND 45)	Yes	Yes	Yes	Yes	High
Matsuura et al. 2005 (rat; ~10 weeks, 2-generation; premating–PND 21)	Yes	Yes	Yes	Yes	High
Sauviat et al. 2005 (rat; ~13 weeks)	Yes	No	Yes	Yes	Moderate
Srinivasan et al. 1991 (rat; GDs 0–21, LDs 1–28)	Yes	No	Yes	Yes	Moderate
Srivastava et al. 2019 (rat, GDs 5–21)	Yes	Yes	Yes	Yes	High
Seiler et al. 1994 (rabbit; 12–15 weeks, 3 days/week)	Yes	No	Yes	Yes	Moderate
Outcome: Immunological effects					
Oral acute exposure					
Mediratta et al. 2008 (rat; 14 days)	Yes	Yes	Yes	Yes	High
Hong and Boorman 1993 (mouse; 10 days)	Yes	Yes	Yes	Yes	High
Hong and Boorman 1993 (mouse; 3 days)	Yes	Yes	Yes	Yes	High
Oral intermediate exposure					
Koner et al. 1998 (rat; 8 weeks)	Yes	No	Yes	Yes	Moderate
Mediratta et al. 2008 (rat; 21 days)	Yes	No	Yes	Yes	Moderate
Meera et al. 1992 (mouse; 24 weeks)	Yes	Yes	Yes	Yes	High

A summary of the initial confidence ratings for each outcome is presented in Table C-35. If individual studies for a particular outcome and study type had different study quality ratings, then the highest confidence rating for the group of studies was used to determine the initial confidence rating for the body of evidence; any exceptions were noted in Table C-35.

Table C-35. Initial Confidence Rating for γ-Hexachlorocyclohexane Health Effects Studies

	Initial study confidence	Initial confidence rating
Outcome: Developmental effects		
Oral acute exposure		
Animal studies		
Dalsenter et al. 1997a	High	High
Dalsenter et al. 1997b	High	
Dalsenter et al. 1997b	High	
Johri et al. 2008	High	
Khera et al. 1979	High	
Palmer et al. 1978	High	
Rivera et al. 1991	Moderate	
Rivera et al. 1998	Moderate	
Rivera et al. 1998	Moderate	
Serrano et al. 1990	Moderate	
Di Consiglio et al. 2009	Moderate	
Hassoun and Stohs 1996a	High	
Hassoun and Stohs 1996a	Moderate	
La Sala et al. 2009	High	
Maranghi et al. 2007	High	
Traina et al. 2003	High	
Palmer et al. 1978	High	
Oral intermediate exposure		
Animal studies		
Breton et al. 2005	Moderate	High
EPA 1991a	High	
EPA 1999c	High	
Johri et al. 2007	High	
Johri et al. 2008	High	
Matsuura et al. 2005	High	
Sauviat et al. 2005	Moderate	
Srinivasan et al. 1991	Moderate	
Srivastava et al. 2019	High	
Seiler et al. 1994	Moderate	
Oral chronic exposure		
Human studies		
Fenster et al. 2006	Moderate	Moderate

Table C-35. Initial Confidence Rating for γ-Hexachlorocyclohexane Health Effects Studies

	Initial study confidence	Initial confidence rating
Yang et al. 2021a	Moderate	
Garcia-Villarino et al. 2022	Moderate	
Fernandez et al. 2007	Low	
Mustafa et al. 2013	Low	
Sharma et al. 2012	Low	
Siddiqui et al. 2003	Low	
Fang et al. 2019a, 2019b	Low	
Yang et al. 2021b	Low	
Yin et al. 2021	Low	
Outcome: Immunological effects		
Oral acute exposure		
Animal studies		
Mediratta et al. 2008	High	
Hong and Boorman 1993	High	High
Hong and Boorman 1993	High	
Oral intermediate exposure		
Animal studies		
Koner et al. 1998	Moderate	
Mediratta et al. 2008	Moderate	High
Meera et al. 1992	High	
Oral chronic exposure		
Human studies		
Landgren et al. 2009	Moderate	Moderate
Meng et al. 2016	Low	

C.16.2 Adjustment of the Confidence Rating—γ-HCH

The initial confidence rating was then downgraded or upgraded depending on whether there were substantial issues that would decrease or increase confidence in the body of evidence. The five properties of the body of evidence that were considered to determine whether the confidence rating should be downgraded and the four properties of the body of evidence that were considered to determine whether the confidence rating should be upgraded are described above in Section C.6.2. The summaries of the assessment of the confidence in the body of evidence for developmental and immune system effects are presented in Table C-36. If the confidence ratings for a particular outcome were based on more than one type of human study, then the highest confidence rating was used for subsequent analyses. An overview of the confidence in the body of evidence for all health effects associated with γ-HCH exposure is presented in Table C-37.

Table C-36. Adjustments to the Initial Confidence in the Body of Evidence

	Initial confidence	Adjustments to the initial confidence rating	Final confidence
Developmental effects			
Human studies	Moderate	-1 imprecision	Low
Animal studies	High	+1 consistency	High
Immunological effects			
Human Studies	Moderate	-1 risk of bias	Low
Animal Studies	High	+1 consistency	High

Table C-37. Confidence in the Body of Evidence for γ-Hexachlorocyclohexane

	Confidence in body of evidence	
Outcome	Human studies	Animal studies
Developmental	Low	High
Immune	Low	High

C.17 TRANSLATE CONFIDENCE RATING INTO LEVEL OF EVIDENCE OF HEALTH EFFECTS—γ-HCH

As described in Section C.7, the confidence in the body of evidence for specific outcomes was translated to a level of evidence rating. The level of evidence rating reflected the confidence in the body of evidence and the direction of the effect (i.e., toxicity or no toxicity); route-specific differences were noted.

A summary of the level of evidence of health effects for γ-HCH is presented in Table C-38.

Table C-38. Level of Evidence of Health Effects for γ-Hexachlorocyclohexane

Outcome	Confidence in body of evidence	Direction of health effect	Level of evidence for health effect
Human studies			
Developmental	Low	Health effect	Low
Immunological	Low	Health effect	Low
Animal studies			
Developmental	High	Health effect	High
Immunological	High	Health effect	High

C.18 INTEGRATE EVIDENCE TO DEVELOP HAZARD IDENTIFICATION CONCLUSIONS—γ-HCH

The final step involved the integration of the evidence streams for the human studies and animal studies to allow for a determination of hazard identification conclusions. Refer to Section C.8 for the four hazard identification conclusion categories for health effects, the hazard characterization scheme (see Figure C-1), and the hazard identification conclusion categories.

The hazard identification conclusions for γ-HCH are listed below and summarized in Table C-39.

Presumed Health Effects

- Developmental
  - Low level of evidence in humans based on associations between γ-HCH in maternal or fetal blood (or tissue) and intrauterine growth retardation/fetal growth retardation in small case-control studies in India (Sharma et al. 2012; Siddiqui et al. 2003), decreased gestational age and increased preterm birth in a cross-sectional study in China (Fang et al. 2019a, 2019b) and a case-control study in India (Mustafa et al. 2013), and cryptorchidism or hypospadias in a nested case-control study in Spain (Fernandez et al. 2007).
  - High level of evidence in animals based on studies in a variety of species exposed to γ-HCH for acute or intermediate durations during gestation or postnatal development demonstrating effects on a wide range of developmental endpoints, including birth outcomes (Beard et al. 1997; EPA 1991a, 1999c; Hassoun and Stohs 1996a; Matsuura et al. 2005; Sauviat et al. 2005) and development of the male and female reproductive tracts (Agrahari et al. 2019; Dalsenter et al. 1997a, 1997b; Di Consiglio et al. 2009; La Sala et al. 2009; Maranghi et al. 2007; Matsuura et al. 2005; Traina et al. 2003), central nervous system (Albertson et al. 1985; Breton et al. 2005; EPA 1999c; Johri et al. 2007, 2008; Rivera et al. 1991, 1998; Srivastava et al. 2019), heart (Sauviat et al. 2005), liver (Srinivasan et al. 1991), and thymus and spleen (Hassoun et al. 1996; Matsuura et al. 2005).
- Immunological
  - Low level of evidence in humans based on association between asthma and plasma levels of γ-HCH in children (Meng et al. 2016) and no evidence for increased prevalence of monoclonal gammopathy of undetermined significance in cohort of male pesticide applicators followed for 9 years (Landgren et al. 2009).
  - High level of evidence in animals based on acute- and intermediate-duration studies of γ-HCH administered orally to rats, mice, rabbits, and sheep showing suppression of the immune system (Banerjee et al. 1996; Desi et al. 1978; Dewan et al. 1980; Khurana et al. 1999; Koner et al. 1998; Mediratta et al. 2008; Meera et al. 1992) and effects on thymus, spleen, and lymph node weights or histology (Hong and Boorman 1993; Meera et al. 1992).

Table C-39. Hazard Identification Conclusions for γ-Hexachlorocyclohexane

Outcome	Hazard identification
Developmental	Presumed health effect
Immunological	Presumed health effect

APPENDIX D. 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 D-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 D-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-duration 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 (1)

Figure key[a] (3)	Species (strain) No./group (4)	Exposure parameters (5)	Doses (mg/kg/day) (5)	Parameters monitored (6)	Endpoint (7)	NOAEL (mg/kg/day) (8)	Less serious LOAEL (mg/kg/day) (9)	Serious LOAEL (mg/kg/day) (9)	Effect
CHRONIC EXPOSURE (2)									
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 (10)									
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									

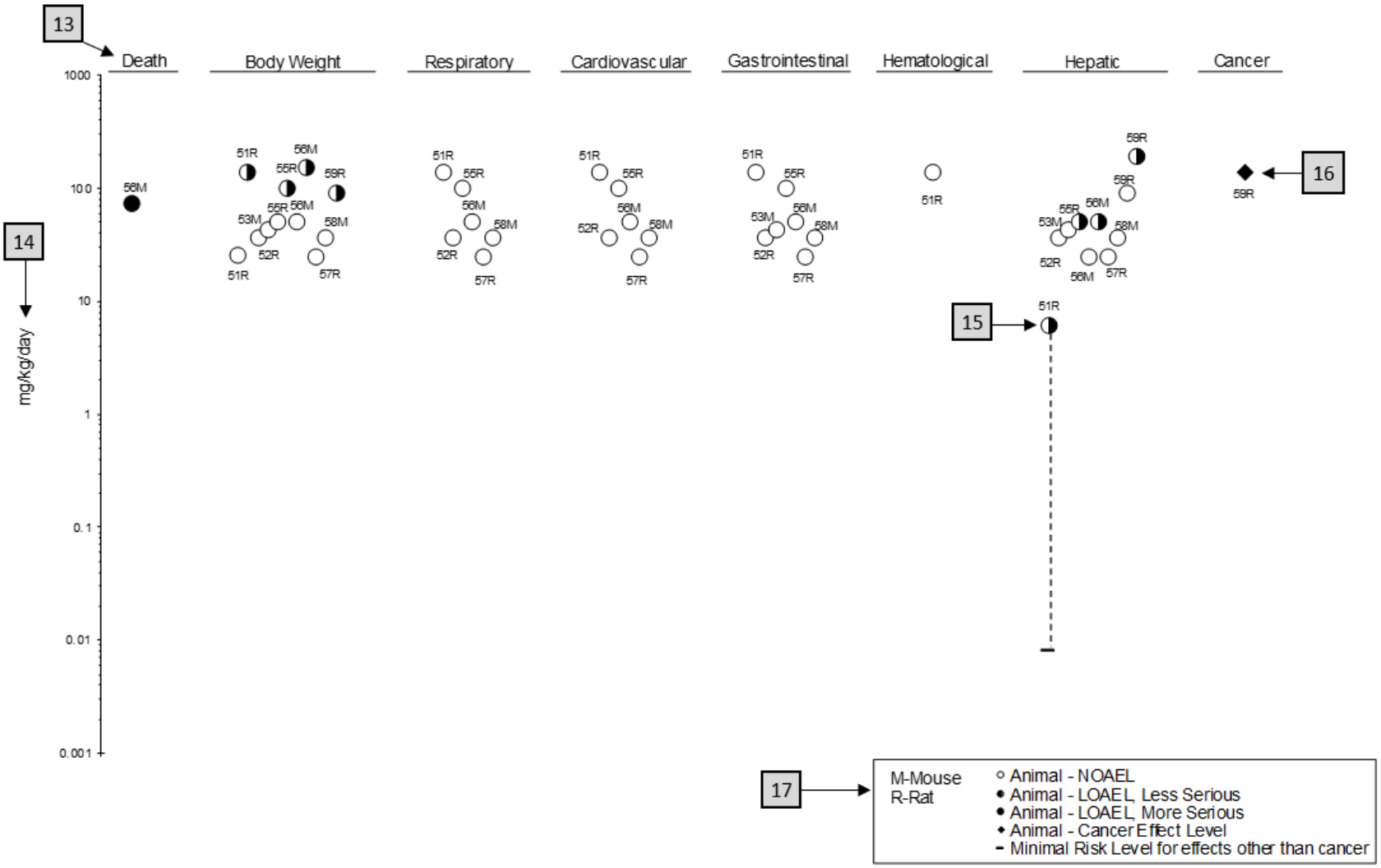
[a]The number corresponds to entries in Figure 2-x.

(11) [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 D

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



APPENDIX E. 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:

Clinical Briefs and Overview discuss health effects and approaches to patient management in a brief/factsheet style. They 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 F. 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 malignant 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 LOAEL—Indicates a minimal adverse effect or a reduced capacity of an organ or system to absorb additional toxic stress that does not necessarily lead to the inability of the organ or system to function normally.

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.

Serious LOAEL—A dose that evokes failure in a biological system and can lead to morbidity or mortality.

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 G. 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 Substances 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