

UPDATING EPA'S 2010 TOXICITY ASSESSMENT OF CHLOROPRENE

OUTLINE OF PRESENTATION

- Objectives of Ramboll Environ's evaluation of the IRIS Toxicological Review of Chloroprene
- Overview of the IRIS Toxicological Review
- Key NRC recommendations for improving IRIS methods
- Epidemiological studies of chloroprene exposed workers and cancer risks
- Toxicological Evidence of carcinogenicity and MOA
- Uncertainties and assumptions in the current IUR
- Reconciling large differences between mice and humans
- Summary and conclusions
- Next steps: deriving an IUR based on integration of current published evidence

RAMBOLL ENVIRON'S EVALUATION OF THE IRIS TOXICOLOGY REVIEW OF CHLOROPRENE - OBJECTIVES

- Independent critical review and integration of the epidemiological, toxicological and mechanistic evidence published to date on chloroprene as a human carcinogen
- Evaluation of the IRIS Toxicological Review of Chloroprene (US EPA, 2010), and especially the IUR, in light of NRC guidance for improved IRIS evaluations and new publications

RAMBOLL ENVIRON'S EVALUATION OF THE IRIS TOXICOLOGICAL REVIEW OF CHLOROPRENE - FINDINGS

Updating the IUR is warranted based on several key points:

1. Epidemiological evidence may have been misinterpreted
 - Epidemiology insufficient to conclude that chloroprene causes cancer in humans
 - Data are inadequate for determining risk estimates
2. Published IUR included influential adjustments and multiple upward rounding
3. Reliance on animal evidence for risk estimates requires fuller consideration of evidence of profound interspecies differences
 - Tumor incidence indicates differences in sensitivity
 - New studies clearly demonstrate toxicokinetic differences
 - Current best practices would indicate validated PBPK modeling (as used with VCM)

OVERVIEW OF 2010 TOXICOLOGICAL REVIEW

- Review was finalized and published in 2010 (US EPA, 2010)
- EPA classified chloroprene as “likely to be a human carcinogen”
- Basis for EPA’s carcinogenicity classification:
 - Results from the National Toxicology Program (NTP, 1998) chronic inhalation bioassay;
 - Associations noted between chloroprene exposure and liver cancer in four of nine studies;
 - Limited evidence of an association between chloroprene exposure and lung cancer;
 - Proposed mutagenic mode of action; and
 - Analogies drawn between chloroprene, 1,3-butadiene and vinyl chloride monomer
- EPA established an IUR of $5 \times 10^{-4} (\mu\text{g}/\text{m}^3)^{-1}$ in the Toxicological Review

https://cfpub.epa.gov/ncea/iris2/chemicalLanding.cfm?substance_nmbr=1021

NRC (2011, 2014) NOTABLE RECOMMENDATIONS FOR IMPROVING THE IRIS PROCESS

Greater documentation and transparency needed regarding

- Inclusion and exclusion criteria
- How studies are evaluated: quality assessment and weighting
 - Impact of study methods
 - Validity of exposure metrics or surrogates

Better integration of data across all lines of scientific evidence

- Is there consistency across lines of evidence?
- What are the plausible biological explanations for inconsistencies, *e.g.*, toxicokinetic differences across species that impact dose-response relationships?

APPLICATION OF NRC RECOMMENDATIONS IN THE RE-EVALUATION OF CHLOROPRENE TOXICITY DEMONSTRATE NEED TO UPDATE IUR

- Revisiting the epidemiological evidence
 - Marsh et al. (2007 a,b) is the most robust epidemiology study of chloroprene exposed workers
 - No excess liver or respiratory cancers; no notable association with chloroprene exposure
- Revisiting evidence supporting a mutagenic MOA
 - Standard *in vivo* genotoxicity tests are negative
 - Evidence of point mutations is inconsistent in *in vitro* assays
 - Evidence suggests a cytotoxic MOA as an alternative to a genotoxic MOA
- Toxicokinetic evidence, including new published data
 - Totality of published, peer-reviewed evidence, including recent analyses using a validated PBPK model, highlight the need to address differences in toxicokinetics, especially if an IUR is based on mouse data

Integration of the evidence strongly suggests that the responses relied on for the IUR are unique to the mouse. Absent strong, affirmative epidemiological evidence of cancer risk, employing an approach that quantitatively addresses differences between the mouse and human is critical.

WEIGHT OF EPIDEMIOLOGICAL EVIDENCE SHOULD BE REEVALUATED

- Occupational cohort studies have been conducted around the world
 - U.S. and Western European cohorts (Pell 1978, Leet & Selevan 1982, Colonna & Leydevant 2001, Marsh et al. 2007 a,b)
 - Eastern European and Asian cohorts (Bulbulyan et al. 1998,1999, Li et al. 1989)
- US and Western European cohort studies are more robust
 - Pooled study (Marsh et al, 2007 a,b) is the largest and strongest
- Eastern European and Asian cohorts have significant limitations
 - Poor documentation of cohort enumeration and inadequate reference rates
 - Low statistical power and unstable relative risk estimates
 - Poor occupational exposure assessment, including identification and consideration of potentially consequential confounding factors

US and Western cohort studies warrant greater weight

KEY LIMITATIONS IN THE ARMENIAN, RUSSIAN AND CHINESE COHORTS

- Results are unstable and unreliable primarily due to small study populations in which the expected number of specific cancer deaths is often less than two.
- Inaccurate or inappropriate reference population rates leading to improper estimates of expected deaths.
- Additional methodological weaknesses including poor characterization of chloroprene and other chemical and lifestyle exposures.
- Inadequate consideration of other causes of liver and lung cancers prevalent outside of the US and western Europe.
 - China: high rates of liver cancer due to hepatitis B viral infection and aflatoxin exposure
 - Armenia and Russia: High prevalence and levels of cigarette smoking and alcohol consumption
- No updates have been conducted of the Chinese, Russian, or Armenian cohorts to clarify or confirm the mixed and unstable findings.

MARSH ET AL. (2007) STUDY FINDINGS SHOULD BE GIVEN GREATEST WEIGHT

USEPA Criteria	US Study (Marsh et al., 2007)				Other Studies			
	Kentucky ¹	North Ireland ¹	Louisiana ¹	France-M ¹	Armenia ²	France-It ³	Russia ⁴	China ⁵
Clear objectives	H‡	H	H	H	H	H-M	H	M
Comparison groups	H	H-M	H-M	M	M	M	M-L	L
Exposure	H	H	H	H	M	M	L	L
Follow-up	H	H-M	H	H-M	M-L	M-L	M-L	M-L
Case ascertainment	H	H-M	H-M	H-M	M	M	M	H-M
Control of bias	H-M	H-M	H-M	M	M-L	M	M	M-L
Sample size	H	H	M	L	M-L	L	H-M	M-L
Data collection and evaluation	H	H	H	H	M	M	M-L	M-L
Adequate response	H	H	H	H	M	M	M	H-M
Documentation of results	H	H	H	H	M-L	M	M	L
Overall rank (1=best)	1	2	3	4	5	5	5	6

from Bukowski, 2009

‡ Subjective estimate of study quality for each specific criterion H=high, M=medium, L=low

1 – Marsh et al 2007; 2 – Bulbulyan et al 1999; 3 – Colonna et al 2001; 4 – Bulbulyan et al 1998; 5 – Li et al 1989

COMPARISON OF KEY CRITERIA ACROSS STUDIES

Key Criteria	US and Europe (Marsh <i>et al.</i> 2007)	Armenia (Bulbulyan <i>et al.</i> 1999)	Russia (Bulbulyan <i>et al.</i> 1998)	China (Li <i>et al.</i> 1989)
Sample Size	French, Irish and US 12,430 (KY ~200,000 py)	2,314	5,185	1,258
Follow-up	1949–2000	1979–1993	1979–1993	1969–1983
Exposure Assessment	Exposure modeling – 7 categories	Index (none, low, high)- before/after 1980	Index (none, med, high)- IH (inadequate) + job	High vs. low based on recall
Baseline rates	National, local plant area counties 1960–1994	Armenian rates 1980–1989	Moscow rates 1979–1993 or 1992–1993 (liver)	From “local area” 1973–1975 expected lung cancers: 0.4
Confounding	Used local rate comparisons; Low prevalence of other liver cancer risk factors	Alcohol use (high cirrhosis rates) and smoking prevalent	Alcohol use (high cirrhosis rates) and smoking; Co-exposure to VC	Hepatitis B and aflatoxin; Co-exposure to VC

SUMMARY OF COHORT RESULTS SHOW INCONSISTENCIES

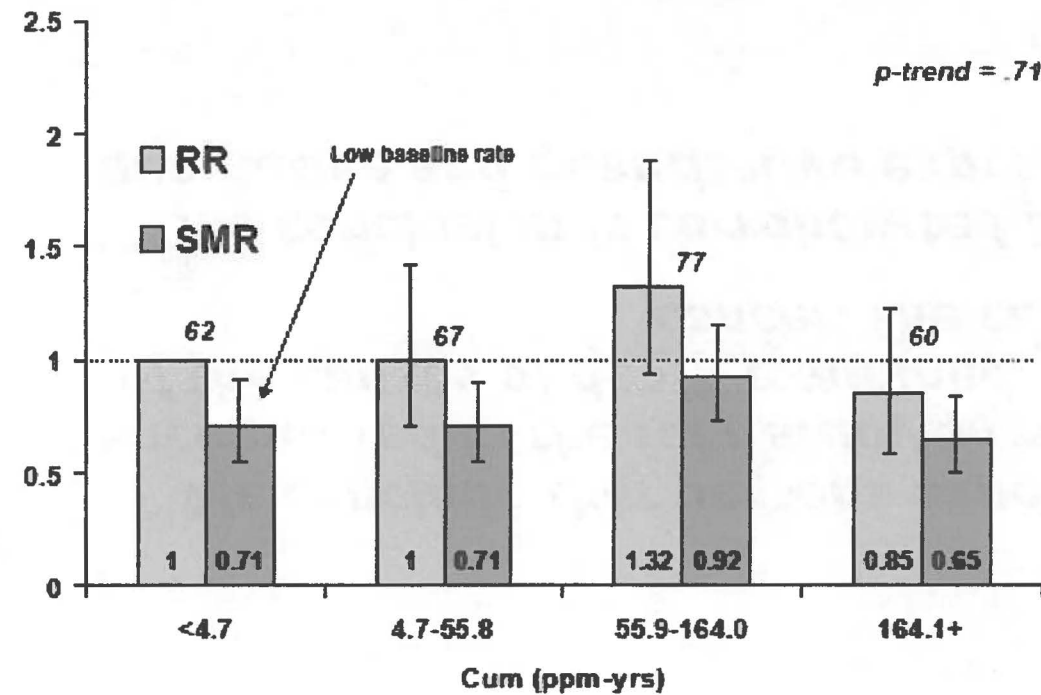
Cohort	Lung Cancer SMR (95% CI)	Liver Cancer SMR (95% CI)
Chinese	5.13 (0.62–18.5)	2.42 (0.89–5.3)
Russian	1.40 (0.9–2.0)	2.40 (1.1–4.3)
Armenian	0.53 (0.24–1.19)	3.27 (1.5–7.3)
French	0.47 (0.13–1.20)	0.56 (0.01–3.1)
Irish	0.78 (0.56–1.05)	0.24 (0.01–1.34)
US KY	0.75 (0.66–0.85)	0.90 (0.52–1.4)
US LA	0.55 (0.26–1.00)	0.00 (0.0–2.4)

Source: Bukowski 2009

MARSH STUDY SHOWS NO LIVER OR LUNG CANCER RISKS

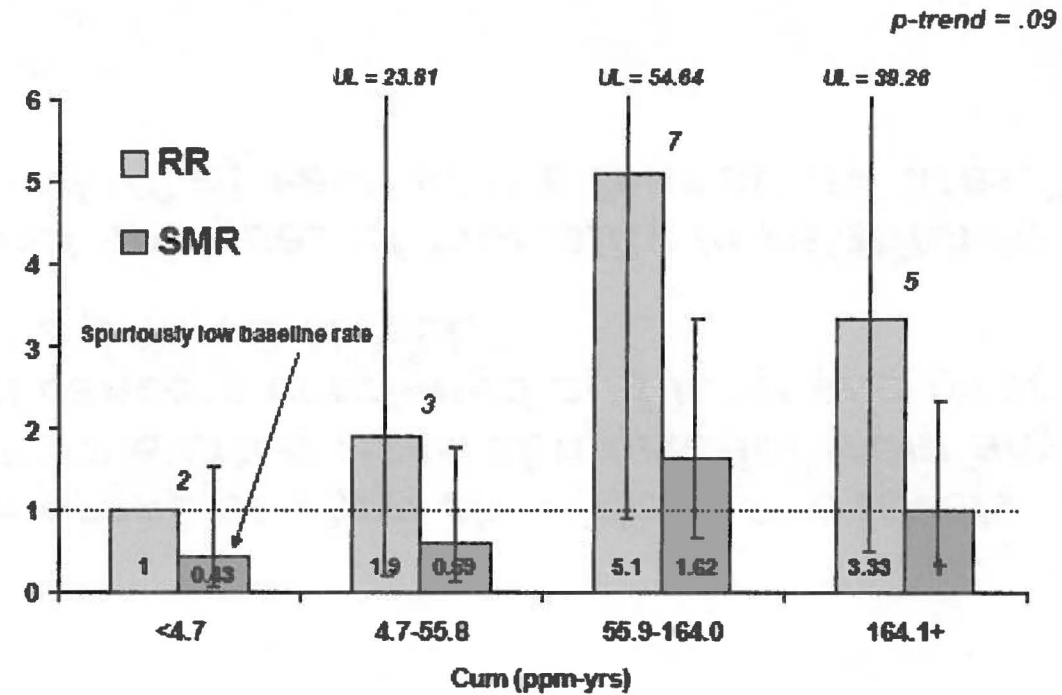
Marsh *et al.* (2007 a,b) study demonstrates no lung or liver cancer excess risk and no clear exposure-response (e.g., largest US plant)

Respiratory Cancer RRs and SMRs by Cumulative CD Exposure, Louisville



Number of observed deaths shown above bar
RRs also adjusted for gender

Liver Cancer RRs and SMRs by Cumulative CD Exposure, Louisville



Number of observed deaths shown above bar
RRs also adjusted for gender

MARSH ET AL. (2007) CONCLUSION

"We conclude that persons exposed to chloroprene or vinyl chloride at the levels encountered in the four study sites did not have elevated risks of mortality from any of the causes of death examined, including all cancers combined and lung and liver cancer, the cancer sites of a priori interest."

"This conclusion is corroborated by our detailed analyses of mortality in relation to qualitative and quantitative exposures to CD and VC at each of the four study sites."

G.M. Marsh et al. / Chemico-Biological Interactions 166 (2007) 285–300

EPIDEMIOLOGICAL SUMMARY

- A re-evaluation of the epidemiological evidence is warranted, including a critical review and synthesis.
- Marsh et al. (2007 a,b) should be weighted significantly more than studies from Asia, Russia and Armenia.
- A weight of evidence evaluation of the most robust studies does not demonstrate a causal association between chloroprene exposure and lung or liver cancers.

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EVIDENCE OF GENOTOXICITY IS INCONSISTENT AND SHOULD BE RECONSIDERED

Supporting

In Vitro

- Mutagenic in Ames test in two of four publications (Bartsch *et al.* 1979; Willems 1980); only aged chloroprene mutagenic
- (1-chloroethenyl)oxirane metabolite mutagenic in the Ames test without S9 mix using a pre-incubation test method (Himmelstein *et al.*, 2001)
- (1-chloroethenyl)oxirane metabolite of chloroprene reacts with nucleosides and double stranded DNA to form DNA adducts (Munter *et al.*, 2002; 2007a, b)

In Vivo

- Induced Sex Linked Recessive Lethals via feeding (*Drosophila melanogaster*) in one of two publications (Vogel 1979)
- Excess A to T transversions in oncogene codons in mouse lung and Harderian tumor cells (Sills *et al.*, 1999)

Weight of evidence not consistent with mutagenic MOA; based on NRC recommendations, alternative MOAs should be considered.

Not Supporting

In Vitro

- Not mutagenic in Ames test in two of four publications (NTP 1998; Zeiger *et al.* 1987); freshly distilled chloroprene not mutagenic (Westphal *et al.*, 1994)
- Not a point mutagen in V79 cells (Drevon and Kuroki, 1979)
- (1-chloroethenyl)oxirane metabolite did not induce micronuclei in V79 cells (Himmelstein *et al.* 2001)
- Ames and DNA adduct studies of (1-chloroethenyl) oxirane metabolite show specificity with G/C and not A/T bases (Koch *et al.*, 1994)

In Vivo

- Not directly DNA reactive in conventional NTP (1998) studies
- No induced Sex Linked Recessive Lethals by direct injection (*Drosophila melanogaster*) in one of two publications (Foureman *et al.*, 1994)
- Excess A to T transversions inconsistent with result from Ames and DNA adduct studies (Himmelstein *et al.* 2001; Munter *et al.* 2002)
- Dose related decrease in *ras* mutation induction in mouse lung cells conflicts with the dose related increase in lung tumors (Sills *et al.*, 1999)

EVIDENCE SUPPORTS AN ALTERNATIVE MOA BASED ON TARGET SITE TOXICITY

• ***Evidence for an alternative MOA***

- Toxicity and hyperplasia at tumor target sites (*e.g.*, nose, oral cavity, and/or bronchioles) in mice/rats indicates induced cell proliferation, leading to increased expression of pre-existing mutations (NTP, 1998)
- A to T transversions found in spontaneous mouse lung tumors (*e.g.*, Buzard, 1996)
- Dose-related mouse lung bronchiolar hyperplasia consistent with increased lung tumors (NTP, 1998)
- Oncogene mutation kinetics are inverse to tumor incidence (*e.g.*, Sills *et al.*, 1999)
- Absence of DNA interaction (other than toxicity) in NTP (1998) genotoxicity studies
- No chromosome damage or point mutation in cultured mammalian cells (Shelby 1990; NTP 1998)
- Toxicity studies show a progression of events consistent with alternative MOA (*e.g.*, Melnick *et al.*, 1996)

• ***Comparisons to 1,3-butadiene***

- Chloroprene genotoxicity profile differs considerably from known carcinogens such as 1,3-butadiene
- Greater tissue injury than butadiene in rodents points to role in increased tumor incidence (NTP, 1998)

There is considerable evidence to support an alternative MOA

UNCERTAINTIES IN THE CURRENT IUR DUE TO UNDERLYING ASSUMPTIONS

Step	IUR per ug/m ³	Basis	Resulting Increase in IUR		Cumulative Increase in IUR	
			Factor	%	Factor	%
Most sensitive endpoint/species (portal-of-entry DAF=1.7)	1.06 x 10 ⁻⁴	Lung tumors in female mice as a portal-of-entry effect				
Most sensitive endpoint/species (systemic lesion DAF=1)	1.8 x 10 ⁻⁴	Lung tumors in female mice as a systemic effect	1.7	70%		
Multiple tumor adjustment	2.7 x 10 ⁻⁴	Multiple tumors	1.5	50%		
Rounding	3 x 10 ⁻⁴	Rounding	1.1	10%	2.8	183%
Application of ADAF	4.5 x10 ⁻⁴	Adjustment (without rounding)	1.5	50%	4.2	324%
Application of ADAF	5 x 10 ⁻⁴	Adjustment (with rounding)	1.7	70%	4.8	372%

NEW DATA SUPPORT NEED TO USE CONTEMPORARY PBPK MODELING

- Studies published by Yang et al. (2012) and Thomas et al. (2013) provide support and validation for the PBPK models originally presented by Himmelstein et al. (2004).
- Allen et al. (2014) developed and applied a method that combines available PBPK models for chloroprene with a statistical maximum likelihood approach to evaluate differences in low-dose risk of respiratory system cancer across species.
 - Results from human and animal studies were used to assess the difference between risk estimates based on both external and internal dose metrics.
 - The results demonstrate that the internal dose metric (μmoles of metabolized chloroprene/g lung/day) provides the statistically equivalent human- and animal-based risk estimates.
 - In addition, Allen et al. (2014) conducted uncertainty analyses to address the question of cross-species pharmacodynamic differences.
- The findings from Allen et al. (2014) indicate that an IUR that incorporates cross-species differences in pharmacokinetics would be on the order of 100 times lower than the current IUR (based on lung tumors in mice).

CONSIDERATION OF RECENT SCIENCE RESULTS IN A SIGNIFICANTLY DIFFERENT IUR

IUR per ug/m³		Basis	Resulting Decrease in IUR
US EPA (2010)	1.81 x 10 ⁻⁴	Lung tumors in female mice as a systemic effect	
Allen et al. (2014)	1.86 x 10 ⁻⁶	PBPK dosimetric adjustment of lung tumors in female mice as a systemic effect	~100 fold Decrease

CHLOROPRENE IUR IS NOT CONSISTENT WITH SIMILAR, BUT KNOWN CARCINOGENS

Compound (Year of Review)	IUR per $\mu\text{g}/\text{m}^3$	Basis	PBPK adjustment	Carcinogenicity classification
Chloroprene (2010)	5×10^{-4}	Multiple tumors in mice, mutagenic MOA	No	Possibly Carcinogenic
1,3 Butadiene (2002a)	3×10^{-5}	Human occupational studies	No	Known Carcinogen
Benzene (2002b)	2×10^{-6} to 7.8×10^{-6}	Human occupational studies	No	Known Carcinogen
Vinyl Chloride (2000)	4.4×10^{-6}	Liver tumors in rats	Yes	Known Carcinogen
Tetrachloroethylene (2012)	2.6×10^{-7}	Liver tumors in mice	Yes	Likely to be Carcinogenic

PBPK adjusted IUR for chloroprene is in line with that of compounds that are known carcinogens; IUR for VCM is based on animal data, but with PBPK model dosimetric adjustments

BASES FOR RECONSIDERATION OF CURRENT IUR

- Epidemiological evidence is inadequate for risk assessment.
 - Most robust studies do not support a causal association between occupational exposures to chloroprene and cancer.
- Assumptions and uncertainties in current IUR are not supported by the science; rounding adds additional layers of conservatism.
- Risk assessment based on animal studies
 - Should be based on most sensitive species/end point because of assumption of a single MOA that should be protective of other tumors/sites.
 - Toxicokinetic data indicate toxicokinetic differences that are consistent with observed responses across species; data indicate mice are the most sensitive species due to differences in the metabolism of chloroprene and detoxification of the chloroprene metabolite.
 - PBPK modeling is best approach for extrapolating from animals to humans; a PBPK model is available and validated (Himmelstein et al., 2004; Yang et al. 2012; Allen et al. 2014).

If IUR is based on mouse data, adjustments for differences in pharmacokinetics (e.g., the use of PBPK modelling) are needed to obtain the most scientifically sound value consistent with the available evidence.

NEXT STEPS

- Follow-up call or meeting to examine key points in greater detail
- Provide support or documentation on questions related to our analyses
- Formal request for updating the IRIS review and risk numbers?

THANK YOU

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REFERENCES

- Allen BC, Van Landingham C, Yang Y, Youk AO, Marsh GM, Esmen N, Gentry PR, Clewell III HJ, Himmelstein MW. (2014) A constrained maximum likelihood approach to evaluate the impact of dose metric on cancer risk assessment: Application to b-chloroprene. *Regulatory Toxicology and Pharmacology* 70: 203–213.
- Bartsch H, Malaveille C, Barbin A, and Planche, G. (1979) Mutagenic and alkylating metabolites of halo-ethylenes, 5 chlorobutadienes and dichlorobutenes produced by rodent or human liver tissues. Evidence for oxirane formation by P450-linked microsomal monooxygenases. *Archives of Toxicology* 41(4):249–277.
- Bulbulyan MA, Margaryan AG, Ilychova SA, Astashevsky SV, Uloyan SM, Cogan VY, Colin D, Boffetta P, and Zaridze DG. (1999) Cancer incidence and mortality in a cohort of chloroprene workers from Armenia. *International Journal of Cancer* 81(1):31-33.
- Bulbulyan MA, Changuina OV, Zaridze DG, Astashevsky SV, Colin D, and Boffetta P. (1998) Cancer mortality among Moscow shoe workers exposed to chloroprene (Russia). *Cancer Causes and Control* 9(4):381-387
- Bukowski JA. (2009) Epidemiologic evidence for chloroprene carcinogenicity: Review of study quality and its application to risk assessment. *Risk Analysis* 29(9):1203-1216.
- Buzard GS. (1996) Studies of oncogene activation and tumor suppressor gene activation in normal and neoplastic rodent tissue. *Mutation Research* 365(1-3):43-58.
- Colonna M and Laydevant G. (2001) A cohort study of workers exposed to chloroprene in the department of Isère, France. *Chemico-Biological Interactions* 135-136:505-14.
- Drevon C and Kuroki T. (1979) Mutagenicity of vinyl chloride, vinylidene chloride and chloroprene in V79 Chinese hamster cells. *Mutation Research* 67(2):173–182.

REFERENCES (CONT.)

- Foureman P, Mason JM, Valencia R, Zimmering S. (1994) Chemical mutagenesis testing in drosophila. X. Results of 70 coded chemicals tested for the National Toxicology Program. *Environmental and Molecular Mutagenesis* 23(3):208-227.
- Himmelstein MW, Carpenter SC, and Hinderliter PM. (2004a) Kinetic modeling of betachloroprene metabolism: I. In vitro rates in liver and lung tissue fractions from mice, rats, hamsters, and humans. *Toxicological Sciences* 79(1):18-27.
- Himmelstein MW, Carpenter SC, Evans MV, Hinderliter PM, and Kenyon EM. (2004b) Kinetic modeling of beta-chloroprene metabolism: II. The application of physiologically based modeling for cancer dose response analysis. *Toxicological Sciences* 79(1):28-37.
- Himmelstein MW, Gladnick NL, Donner EM, Synder RD, and Valentine R. (2001) In vitro genotoxicity testing of (1-chloroethenyl)oxirane, a metabolite of beta-chloroprene. *Chemico-Biological Interactions* 135-136:703-713.
- Koch WH, Henrikson EN, Kupchella E, and Cebula TA. (1994) Salmonella typhimurium strain TA100 differentiates several classes of carcinogens and mutagens by base substitution specificity. *Carcinogenesis* 15(1):79-88.
- Leet TL and Selevan SG. (1982) Mortality analysis of workers exposed to chloroprene. Cincinnati: National Institute for Occupational Safety and Health.
- Li SQ, Dong QN, Liu YQ, and Liu YG. (1989) Epidemiologic study of cancer mortality among chloroprene workers. *Biomedical and Environmental Sciences* 2(2):141-149.
- Marsh GM, Youk AO, Buchanich JM, Cunningham M, Esmen NA, Hall TA, and Phillips ML. (2007a) Mortality patterns among industrial workers exposed to chloroprene and other substances: I. General mortality patterns. *Chemico-Biological Interactions* 166(1-3):285-300.
- Marsh GM, Youk AO, Buchanich JM, Cunningham M, Esmen NA, Hall TA, and Phillips ML. (2007b) Mortality patterns among industrial workers exposed to chloroprene and other substances: II. Mortality in relation to exposure. *Chemico-Biological Interactions* 166(1-3):301-316.

REFERENCES (CONT.)

Melnick RL, Elwell MR, Roycroft JH, Chou BJ, Ragan HA, and Miller RA. (1996) Toxicity of inhaled chloroprene (2-chloro-1,3-butadiene) in F344 rats and B6C3F(1) mice. *Toxicology* 108(1-2):79-91.

Munter T, Cottrell L, Ghai R, Golding BT, and Watson WP. (2007a) The metabolism and molecular toxicology of chloroprene. *Chemico-Biological Interactions* 166(1-3):323-331.

Munter T, Cottrell L, Ghai R, Golding BT, and Watson WP. (2007b). The metabolism and molecular toxicology of chloroprene. *Chemico-Biological Interactions* 168(2):169.

Munter T, Cottrell L, Hill S, Kronberg L, Watson WP, and Golding BT. (2002) Identification of adducts derived from reactions of (1-chloroethenyl)oxirane with nucleosides and calf thymus DNA. *Chemical Research in Toxicology* 15(12):1549-1560.

NRC (National Research Council). (2014). Review of US EPA's Integrated Risk Information System (IRIS) Process. Washington, DC: National Academies Press.

NRC (National Research Council). (2011). Review of the Environmental Protection Agency's Draft IRIS Assessment of Formaldehyde. Washington, DC: National Academies Press.

NTP (National Toxicology Program). (1998) Toxicology and carcinogenesis studies of chloroprene (CAS No. 126-99-8) in F344 rats and B6C3F1 mice (inhalation studies). Public Health Service, U.S. department of Health and Human services; NTP TR-467. Available from the National Institute of Environmental Health Sciences, Research Triangle Park, NC and online at [http://ntp.niehs.nih.gov/ntp/htdocs/LT\)rpts/tr467.pdf](http://ntp.niehs.nih.gov/ntp/htdocs/LT)rpts/tr467.pdf)

Pell S. (1978) Mortality of workers exposed to chloroprene. *Journal of Occupational Medicine* 20(1):21-29.

Shelby MD. (1990) Results of NTP-sponsored mouse cytogenetic studies on 1,3-butadiene, isoprene, and chloroprene. *Environmental Health Perspectives* 86:71-73.

REFERENCES (CONT.)

- Sills RC, Hong HL, Melnick RL, Boorman GA, and Devereux TR. (1999) High frequency of codon 61 K-ras A-->T transversions in lung and Harderian gland neoplasms of B6C3F1 mice exposed to chloroprene (2-chloro-1,3-butadiene) for 2 years, and comparisons with the structurally related chemicals isoprene and 1,3-butadiene. *Carcinogenesis* 20(4):657–662.
- Thomas RS, Himmelstein, MW, Clewell HJ, 3rd, Yang Y, Healy E, Black MB, Andersen ME. (2013) Cross-Species Transcriptomic Analysis of Mouse and Rat Lung Exposed to Chloroprene. *Toxicological Sciences* 131(2): 629–640. doi:10.1093/toxsci/kfs314
- U.S EPA (Environmental Protection Agency). (2002b). Toxicological Review of Benzene. In support of Summary Information on the Integrated Risk Information System (IRIS). Washington, DC.
- U.S EPA (Environmental Protection Agency). (2002a). Toxicological Review of 1,3-Butadiene. In support of Summary Information on the Integrated Risk Information System (IRIS). Washington, DC.
- US EPA (Environmental Protection Agency). (2010) Toxicological Review of Chloroprene. In support of Summary Information on the Integrated Risk Information System (IRIS). Washington, DC.
- US EPA (Environmental Protection Agency). (2012). Toxicological Review of Tetrachloroethylene. In Support of Summary Information on the Integrated Risk information System (IRIS). Washington, DC.
- US EPA (Environmental Protection Agency). (2000). Toxicological Review of Vinyl Chloride. In Support of Summary Information on the Integrated Risk information System (IRIS). Washington, DC.
- Vogel E. (1979) Mutagenicity of chloroprene, 1-chloro-1,3-trans-butadiene, 1-4-dichlorobutene-2 and 1,4-dichloro-2,3-epoxybutane in *Drosophila melanogaster*. *Mutation Research* 67(4):377–381.
- .

REFERENCES (CONT.)

Westphal GA, Blaszkewicz M, Leutbecher M, Muller A, Hallier E, and Bolt HM. (1994) Bacterial mutagenicity of 2-chloro-1,3-butadiene (chloroprene) caused by decomposition products. *Archives of Toxicology* 68(2):79-84.

Willems MI. (1980) Evaluation of β -chloroprene and four chloroprene dimers in the Ames test by atmospheric exposure of the tester strains. Final report No. R-6392 by Central Institute for Nutrition and Food research for the Joint Industry Committee on Chloroprene.

Yang Y, Himmelstein MW, and Clewell HJ. (2012) Kinetic modeling of β -chloroprene metabolism: Probabilistic in vitro-in vivo extrapolation of metabolism in the lung, liver and kidneys of mice, rats and humans *Toxicology in Vitro* 26:1047-1055