HSIA
halogenated
solvents
industry
alliance, inc.

March 27, 2020

Environmental Protection Agency
Docket Center
1200 Pennsylvania Avenue, NW
Washington, DC 20460

Re: Docket No. EPA-HQ-OPPT-2019-0499

Dear Sirs:

The Halogenated Solvents Industry Alliance, Inc. (HSIA) represents producers and users of carbon tetrachloride (CTC). We offer these comments on EPA's draft Risk Evaluation for CTC, 85 Fed. Reg. 4658 (Jan. 27, 2020), developed under § 6(b) of the Toxic Substances Control Act (TSCA), as amended in June 2016 by the Frank R. Lautenberg Chemical Safety for the 21st Century Act ("Lautenberg Act").

I. Non-Compliance with TSCA § 26(h) and (i)

As EPA recognizes, TSCA § 26(h) and (i) require EPA to use scientific information, technical procedures, measures, methods, protocols, methodologies, and models consistent with the best available science and to base its decisions on the weight of the scientific evidence. TSCA § 6(b)(4)(F), as revised by the Lautenberg Act, requires that the risk evaluation, while it may not consider costs or other non-risk factors, must among other things:

- "integrate and assess available information on hazards and exposures for the conditions of use of the chemical substance, including information that is relevant to specific risks of injury to health or the environment and information on potentially exposed or susceptible subpopulations identified as relevant by the Administrator;"
- "take into account, where relevant, the likely duration, intensity, frequency, and number of exposures under the conditions of use of the chemical substance;" and
- "describe the weight of the scientific evidence for the identified hazard and exposure."

New TSCA § 26(h) requires for each risk evaluation (as "a decision based on science") that "the Administrator shall use scientific information, technical procedures, measures, methods, protocols, methodologies, or models, employed in a manner consistent with the best available science, and shall consider as applicable—

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(1) the extent to which the scientific information, technical procedures, measures, methods, protocols, methodologies, or models employed to generate the information are reasonable for and consistent with the intended use of the information;

(2) the extent to which the information is relevant for the Administrator's use in making a decision about a chemical substance or mixture;

(3) the degree of clarity and completeness with which the data, assumptions, methods, quality assurance, and analyses employed to generate the information are documented;

(4) the extent to which the variability and uncertainty in the information, or in the procedures, measures, methods, protocols, methodologies, or models, are evaluated and characterized; and

(5) the extent of independent verification or peer review of the information or of the procedures, measures, methods, protocols, methodologies, or models."

Together, these new provisions indicate that a risk evaluation that supports a TSCA § 6 rule must be more robust than a screening level assessment. The draft Risk Evaluation, while commendable for its use of systematic review, continues to rely on the same methodology that EPA has followed for 40 years, as evidenced *inter alia* by its references to the 2005 Guidelines for Carcinogen Risk Assessment[1] and the 2010 IRIS review of CTC.[2] The following, drawn from a landmark report by the National Academy of Sciences,[3] are among the basic default concepts that underlie this methodology:

- Laboratory animals are a surrogate for humans in assessing cancer risks; positive cancer-bioassay results in laboratory animals are taken as evidence of a chemical's cancer-causing potential in humans.
- Humans are as sensitive as the most sensitive animal species, strain, or sex evaluated in bioassay with appropriate study-design characteristics.
- Agents that are positive in long-term animal experiments and also show evidence of promoting or carcinogenic activity should be considered as complete carcinogens.
- Benign tumors are surrogates for malignant tumors, so benign and malignant tumors are added in evaluating wither a chemical is carcinogenic and in assessing its potency.
- Chemicals act like radiation at low exposures (doses) in inducing cancer, i.e., intake of even one molecule of a chemical has an associated probability for cancer induction that can be calculated, so the appropriate model for relating exposure-response relationships is the linearized multistage model.

[1] EPA Guidelines for Carcinogen Risk Assessment and Supplemental Guidance for Assessing Susceptibility from Early-Life Exposure to Carcinogens, 70 Fed. Reg. 17765 (April 7, 2005) (hereafter the "Cancer Guidelines" or the "Guidelines").

[2] EPA Integrated Risk Information System (IRIS) Review of Toxicological Information on Carbon Tetrachloride (2010) (hereafter the "CTC IRIS Assessment").

[3] National Academy of Sciences, *Science and Judgment in Risk Assessment* (NRC/NAS, 1994).

- Important biological parameters, including the rate of metabolism of chemicals, in hums and laboratory animals are related to body surface area. When extrapolating metabolic data from laboratory animals to humans, one may use the relationship of surface area in the test species to that in humans in modifying the laboratory animal data.
- A given unit of intake of a chemical has the same effect, regardless of the time of its intake; chemical intake is integrated over time, irrespective of intake rate and duration.
- Individual chemicals act independently of other chemicals in inducing cancer when multiple chemicals are taken into the body; when assessing the risks associated with exposures to mixtures of chemicals, one treats the risks additively.

EPA's current quantitative risk assessment methodology (QRA) for carcinogens was first used by the Food & Drug Administration (FDA) in the early 1970s. As developed by FDA and EPA, the methodology incorporated generic policy choice default assumptions and policy-based choice of analytic procedures adopted in the then state-of-the-science of carcinogenesis.[4] The impact of these generic policy chosen default options can be seen in a chart prepared in 1984 by Elizabeth Anderson, then the Director of EPA's Office of Health and Environmental Assessment, illustrating the risk-enhancing impact of default options.[5]

The Anderson chart lists six default options that date from the 1970s (and are still in the current guidelines) and compares them to alternatives that could result in a *reduction* in risk estimates by a factor of 16 to 10,800:

Factor	Range of possible reduction in estimated cancer risk
A. Weight vs surface area	2-12
B. Maximum or average likelihood vs upper 95% confidence	2-3
C. Malignant tumors vs malignant plus benign tumors	1-2
D. Average animal sensitivity vs most sensitive animal	2-5
E. Pharmacodynamics vs effective dose	1-6
F. Risks at shorter than equilibrium buildup time	2-5
Total	16-10,800

[4] Much of this discussion is drawn from Barnard, Scientific Method and Risk Assessment, *Reg. Tox. Pharmacol*. 19, 211-218 (1994).

[5] Anderson, E., Use of Risk Assessment in the Evaluation of Public Health Impacts of Toxic Chemicals, Lecture series on *"Risk Analysis in Environmental Health with Emphasis on Carcinogenesis*," Harvard School of Public Health, September 18-20, 1984.

It should be noted that in characterizing the upper confidence limit value generated by the current methodology, EPA did not refer to the impact on the risk estimate of the policy chosen dose-response model, the linearized multistage model (LMS). Alternative models would give risk values several orders of magnitude lower than the LMS model. The best characterization of the plausible upper confidence level estimate generated by the LMS appears in the predecessor to the 2005 Guidelines:

> "Such an estimate, however, does not give a realistic prediction of risk. The true value may be as low as zero. The range of risks, described by the upper limit given by the chosen model and the lower limit which may be as low as zero, should be explicitly stated."[6]

The current risk assessment procedures, although described by EPA as weight-of-the-evidence, involve in fact a mixture of a description of the data which is then used to select those parts of the data for statistical analysis with the analysis limited or constrained by the policy choice of default assumptions and analytic procedures. The data are summarized in the risk assessment document; however, the criteria for interpretation and analysis are policy choices resulting in the regulatory use of an upper confidence limit value calculated using only a selected part of the data. This is not in accordance with TSCA § 26(h) and (i).

The Guidelines recognize that there may be scientific advances not consistent with the policy-based assumptions and the Guidelines accordingly authorize departure in certain cases from the policy default options. In practice the strength of the policy choices has been so strong that departure rarely occurred. Below we describe some of the reasons that a departure is authorized, indeed necessary, in the case of CTC.

II. Limitations of CTC Cancer Risk Assessment

In the Risk Evaluation EPA classifies carbon tetrachloride in accordance with the Cancer Guidelines as "likely to be carcinogenic to humans" based on: "(1) inadequate evidence of carcinogenicity in humans and (2) sufficient evidence in animals by oral and inhalation exposure, *i.e.*, hepatic tumors in multiple species (rat, mouse, and hamster) and pheochromocytomas (adrenal gland tumors) in mice." The CTC cancer classification and the calculation of the Inhalation Unit Risk (IUR) has not fundamentally changed from the 2010 CTC IRIS assessment. This repetition of the 2010 CTC IRIS assessment for the Risk Evaluation does not fulfill the requirements of the Lautenberg Act with the use of the best available science and decisions based on the weight of the scientific evidence.

In the 2010 CTC IRIS assessment, EPA concluded that there is insufficient information on the mode-of-action (MOA) of CTC for mouse liver tumors at low doses and the mouse pheochromocytomas to support a non-linear dose-response approach for assessing cancer risk. In spite of that conclusion, a majority (four out of six) of the EPA Science Advisory Committee

[6] 51 Fed. Reg. 33992 (Sept. 24, 1986).

review for the 2010 CTC IRIS assessment recommended that the CTC cancer risk should be preferably based on a non-linear threshold method.

> To quote directly from the IRIS response to reviewer comments: "Two reviewers considered it appropriate to present a linear low-dose extrapolation approach as an alternative approach, but that based on available evidence, the nonlinear method seems more appropriate." A fifth reviewer stated that use of a linear dose-response model is "difficult to defend and is not a preferable approach", [and a] "sixth reviewer did not agree that a linear assessment is justified for carbon tetrachloride." Even among the two reviewers who agreed that a low-dose linear approach was the "most clear, prudent and scientifically defensible approach", one of those reviewers noted that use of a nonlinear approach is "reasonable to consider", although noting that such an approach might use an additional, possibly 10-fold, uncertainty factor to assure protection of both cancer and non-cancer endpoints.

As discussed below, the Japan Bioassay Research Center (JBRC) rodent inhalation bioassays on which the IUR for CTC is based were not adequately evaluated by EPA in the Risk Evaluation, nor were new scientific data included in the Risk Evaluation that provide important evidence for a cytotoxic-proliferative mode of action (MOA) of CTC at low doses.

A. Use of Animal Data/Mode of Action

Liver Tumors

CTC has been tested for carcinogenicity by the inhalation route in two-year rat and mouse bioassays conducted at the JBRC. The data from these studies are available in an unpublished report[7] and in a scientific journal.[8] Male and female F344 rats and BDF1 mice were exposed by inhalation to 0, 5, 25, or 125 ppm CTC for 6 hours/day, 5 days/week for 104 weeks. In rats, the incidence of liver carcinomas and adenomas were significantly increased in both sexes at 125 ppm, but not at 5 or 25 ppm. Liver carcinomas were increased in the 25 ppm-exposed females, but the incidence was not statistically significantly different from the controls although it did exceed the range of historical control incidence from the testing laboratory. Chronic liver toxicity was also seen in the male and female rats at 25 and 125 ppm, but not at 5 ppm. Thus, liver tumors in rats only occurred at exposure concentrations that also induce liver toxicity.

[7] JBRC, Subchronic inhalation toxicity and carcinogenicity studies of carbon tetrachloride in F344 rats and BDF1 mice (Studies Nos. 0020, 0021, 0043, and 0044). Kanagawa, Japan Industrial Safety and Health Association, Japan Bioassay Research Center, Kanagawa, Japan. Unpublished report to the Ministry of Labor. Hirasawa Hadano Kanagawa, 257 Japan (1998).

[8] Nagano, K, Sasaki, T, Umeda, Y, Nishizawa, T, Ikawa, N, Ohbayashi, H, Arito, H, Yamamato, S, Fukishima, S, Inhalation carcinogenicity and chronic toxicity of carbon tetrachloride in rats and mice, *Inhal. Toxicol.* 19: 1089-1103 (2007b).

In mice, there was a significant increase in liver adenomas and carcinomas in both sexes at 25 and 125 ppm. As reported in the Nagano *et al.* cancer bioassay paper, the incidence of liver adenomas was increased in the 5 ppm-exposed female mice (8/49 or 16.3%) compared to the controls (2/50 or 4%). Nagano *et al.* considered this difference in liver adenoma incidence to be statistically significant by the Fisher exact test ($p \leq 0.05$, two-tailed test). The published paper also reported that "the tumor incidence exceeded the upper range of the JBRC historical control data (43 cases [5.1%] in 849 female Crj:BDF1 mice in seventeen 2-yr inhalation studies that have been conducted in the JBRC during the 17-yr period from 1990 to 2006, with a maximum incidence of 12% in a single study." Chronic liver toxicity was seen in the male and female mice at 25 ppm and 125 ppm, but not at 5 ppm.

A major consideration of EPA in the 2010 IRIS assessment and in the Risk Evaluation to not accept a cytotoxic-proliferative MOA for CTC carcinogenicity was that the Nagano *et al.* cancer bioassay showed "An [statistically significant] increased incidence of hepatocellular adenomas occurred in the low-exposure [5 ppm] female mouse in the absence of nonneoplastic liver toxicity." However, this conclusion by EPA is based on an interpretation of the data that is not the current practice by the National Toxicology Program (NTP).[9] The NTP uses what is known as the "Haseman Rule" which tests the significant differences in tumor incidence between the control and dose groups at 0.05 for rare tumors and at 0.01 for common tumors [liver adenomas are considered a common tumor in this strain of mice because the background incidence is >1%]. The JBRC (1998) unpublished study report lists a p value of 0.0634 from a Fisher exact test (two-tail) for the statistical difference in liver adenomas between the 5 ppm female mice and the controls, which is in agreement with the $p \leq 0.05$ listed in the Nagano *et al.* cancer bioassay. However, based on the "Haseman Rule", the increased incidence of liver adenomas in the 5 ppm female mice is not statistically significant at $p \leq 0.01$ and therefore should not be considered treatment-related.

Additional evidence that that the 5 ppm exposure concentration in the JBRC study is a No-Observed- Effect-Level for liver tumors is the personal communication between Kasuke Nagano (JBRC) to Mary Manibusan (EPA) mentioned in the CTC IRIS assessment on the historical control data in Crj:BDF1 mice from 20 studies at JBRC. JBRC reported a mean incidence of 5.2% and a range of 2 to 10%, which, when compared to a 4% incidence of liver adenomas in the female mice controls, suggests that the incidence of liver adenomas was unusually low in control mice in the CTC study, thus exaggerating the statistical difference between the 5 ppm females and the controls. The wide range in the historical control range for liver adenomas may also indicate that background rates for these tumors are highly variable.

EPA also did not include in the Risk Evaluation a scientific study published since the 2010 IRIS assessment that provides important evidence for a non-linear MOA for CTC carcinogenicity at low doses. Uehara *et al.* (2013) published a study that investigated whether fibrosis promotes

[9] Kissling, GE, Haseman, JK, Zeigler, E, Proper interpretation of chronic toxicity studies and their statistics: A critique of "Which level of evidence does the US National Toxicology Program provide? Statistical considerations using the Technical Report 578 on *Ginkgo biloba* as an example", Toxicol Lett. 237: 161-164 (2015).

hepatocellular carcinomas (HCC) via a mechanism involving liver stem cell activation.[10] Groups of male $B6C3F_1$ mice (14 days old) were given vehicle control or a single intraperitoneal injection of 1 mg/kg diethylnitrosamine. At eight weeks of age the mice were administered twice per week either vehicle control or 0.2 ml/kg (318 mg/kg) CTC for 9 or 14 weeks. The mice that received CTC only showed clear evidence of liver toxicity and by 22 weeks of age, increased incidences of liver adenomas and carcinomas. Lipid peroxidation was significantly increased in the mouse livers at 17 and 22 weeks of age, as determined by 4-hydroxynonenal (4-HNE) staining. Most importantly, the number of 8-oxo-deoxyguanine adducts in liver DNA was not increased in the CTC-treated mice at either time point compared to the controls (8-oxo-dG/10^6 dG at 17 weeks: 2.3 ± 1.1 vs.1.6 ± 0.5, controls; 8-oxo-dG/10^6 dG at 22 weeks: 1.2 ± 0.4 vs. 1.2 ± 1.2, controls). These findings show that there was no secondary DNA damage associated with CTC radical-induced lipid peroxidation and/or cytotoxicity at the time points measured at a relatively low dose of CTC that also resulted in liver tumors in mice. This lack of concordance between DNA adducts and cellular oxidative stress in liver tumor-bearing mice dosed with CTC provides critical evidence supporting a cytotoxic-proliferative (non-linear) MOA for CTC carcinogenicity at low doses. Hence, EPA's position in the Risk Evaluation of a low-dose linear MOA for liver tumors is untenable in light of the most-up-to-date scientific studies on carbon tetrachloride toxicity.

There are a number of mechanisms in cells to defend against oxidative stress from reactive oxygen species (ROS). For example, the upregulation of stress-response genes is a common adaptive response induced in mammalian cells, many of which encode antioxidant defense enzymes.[11] Catalase and superoxide dismutase are endogenous antioxidant enzymes that neutralize ROS; endogenous glutathione (GSH) also plays a critical role as an antioxidant. Given the many antioxidant protective mechanisms in cells, it is biologically implausible that CTC causes liver tumors via a non-threshold (linear) dose-response MOA. A critical amount of the CTC metabolites or the reactive products of oxidative stress has to be present in order to overwhelm the cellular antioxidant defenses of the cell to reach the DNA. The study by Uehara *et al.* showed that oxidative stress and liver cell damage can occur in the absence of oxidative DNA damage at relatively low CTC doses.

In summary, the oral and inhalation carcinogenicity studies on CTC in rats, mice, and hamsters show increases in liver tumors at doses that also cause liver toxicity. While liver adenomas were increased in the 5 ppm-exposed female mice in the absence of liver toxicity, this increase was not statistically significant using the level of significance ($p \leq 0.01$) used by NTP and others; this increase may been artifactual due to an unusually low incidence of liver adenomas in the control mice. Therefore, 5 ppm should be considered a No-Observed-Effect-Concentration (NOEC) for both liver toxicity and liver cancer. EPA has concluded that "The available data for carbon tetrachloride do not support a conclusion that this compound induces cancer through a mutagenic mode of action." Based on a considerable number of scientific studies, the MOA can

[10] Uehara, T, Ainslie, GR, Kutanzi, K, Pogribny, IP, Muskhelishvilli, L, Izawa, T, Yamate, J, Kosyk, O, Shymonyak, S, Bradford, BU, Boorman, GA, Bataller, R, Rusyn. I, Molecular mechanisms of fibrosis-associated promotion of liver carcinogenesis, *Toxicol. Sci.* 132: 53-63 (2013).

[11] Klaunig, JE, Kamendulis, LM, Hocevar, BA, Oxidative stress and oxidative damage in carcinogenesis, *Toxicol. Pathol.* 38: 96-109 (2010).

be explained by the involvement of cytotoxicity and proliferation from the highly reactive radical metabolites of CTC. These metabolites are formed in the endoplasmic reticulum from the CYP2E1 enzymes which immediately react with cellular macromolecules, such as proteins and lipids in the near vicinity of the endoplasmic reticulum, causing cellular damage. These radical metabolites are not expected to reach the DNA in the nucleus of the cell because of their high reactivity. Lipid peroxidation has been shown to occur from the reaction of these radicals, particularly the trichloromethyl peroxy radical, with lipids releasing hydroxyl radicals and reactive aldehydes, such as 4-HNE. The hydroxyl radical has the potential to react with DNA and can form promutagenic 8-OH-dG adducts.[12] However, as noted above, oxidative DNA damage was not associated with CTC radical-induced lipid peroxidation and/or cytotoxicity. Thus, the best available science and the weight of the scientific evidence indicate that CTC is carcinogenic in the liver only via a MOA with a non-linear (threshold) dose-response.

Mouse pheochromocytomas

EPA based its IUR for CTC on the male mouse pheochromocytomas (adrenal medulla tumors) from the JBRC two-year carcinogenicity study and a low-dose linear extrapolation approach for estimating human cancer risk. The incidence of pheochromocytomas was significantly increased in the 25 and 125 ppm male mice and in the 125 ppm female mice. Pheochromocytomas were not increased in either the male or female rats. In the CTC IRIS assessment, EPA stated: "The application of a nonlinear approach for liver tumors is based on MOA information specific to that tumor type and therefore does not apply to pheochromocytomas for which the MOA is unknown. The RfD and RfC based on liver toxicity cannot be assumed to be protective for the potential cancer risk associated with carbon tetrachloride-induced pheochromocytomas." This argument by EPA, assumes that our understanding of the science is not adequate for a plausible MOA for the mouse pheochromocytomas while ignoring the fundamental aspects of CTC metabolism and cellular biology. Below are several key points suggesting similar low-dose threshold MOAs for both liver and adrenal medulla tumors:

- Adrenal medulla cells have the same basic cell structure as liver cells.
- CTC is expected to be metabolized to trichloromethyl and trichloromethyl peroxy radical metabolites in the ER.
- Reactive CTC radical mechanisms in adrenal medulla cells are expected to be similar to liver cells.
- Antioxidant defense mechanisms in adrenal medulla cells are expected to be similar to liver cells.
- Mutagenic MOA for tumors is not supported by genotoxicity data.

[12] Cheng, KC, Cahill, DS, Kasai, H, Nishimura, S, Loeb, LA, 8-Hydroxyguanine, an abundant form of oxidative DNA damage, cause G-T and A-C mutations, *J. Biol. Chem.* 267: 166-172 (1992).

As pheochromocytomas occurred in mice at exposure concentrations that also resulted in toxicity in liver cells, estimation of human cancer risk based on liver toxicity would be adequately protective for both tumor types.

B. Human and Animal Studies of Brain and Nervous System Tumors

The weight of the evidence from human epidemiology and animal studies show no increased risk of brain and nervous system tumors from carbon tetrachloride exposure.

Epidemiology Studies

The draft Risk Evaluation includes three epidemiological studies on brains tumors and one study on nervous system tumors published since the 2010 CTC IRIS assessment and which were considered acceptable based on the quality criteria in the systematic review. While a brief summary of the results of these studies (and others) was provided in Table 3-8 of the Risk Evaluation, there was no analysis of the studies, nor was any conclusion provided regarding the overall evidence for an association between these tumors and CTC exposure. EPA also incorrectly described Heck *et al.* (2013)[13] as a study of brain cancer, but it was actually of neuroblastomas, a childhood cancer arising from cells that form the sympathetic nervous system, which is not the brain.

These four epidemiology studies plus an additional study on astrocytic brain tumors by Heineman *et al*. (1994)[14] were reviewed by Dr. Carol Burns (see Attachment A). Considering the risk of bias, lack of consistency, and high contribution of chance and confounding, Dr. Burns concluded that these five studies do not show an increased risk of brain and nervous system tumors and carbon tetrachloride exposure. It is important to note in these small epidemiology studies of rare diseases and uncommon exposures that artificially high risk estimates can occur from random variability, resulting is a phenomenon of effect size magnification.[15] The results may be statistically significant but with very wide confidence intervals that indicate imprecision. This imprecision is seen in all five of the epidemiology studies, with the exception of the case-control study by Ruder *et al.* (2013)[16] which showed no association between brain tumors and CTC exposures.

[13] Heck, JE, Park, AS, Qiu, J, Cockburn, M, Ritz, B, An exploratory study of ambient air toxics exposure in pregnancy and the risk of neuroblastomas in offspring, *Environ. Res*. 127: 1-6 (2013).

[14] Heineman, EF, Cocco, P, Gomez, MR, Dosemeci, M, Stewart, PA, Hayes, RB, Zahm, SH, Thomas, TL, Blair, A, Occupational exposure to chlorinated aliphatic hydrocarbons and risk of astrocytic brain cancer, *A. J. Ind. Med.* 26: 155-169 (1994).

[15] EPA, Framework for Incorporating Human Epidemiologic & Incident Data in Risk Assessments for Pesticides, Office of Pesticide Programs (2016).

[16] Ruder AM, Yiin JH, Waters MA, *et al*., The Upper Midwest Health Study: Gliomas and Occupational Exposure to Chlorinated Solvents, *Occup. Environ. Med.* 70: 73-80 (2013).

Animal Studies

The animal toxicity data on CTC do not support brain tumors being a health concern. Very low levels of CYP2E1 enzyme have been detected in rat brain tissue,[17] indicating that the brain has some capacity to metabolize CTC. And, indeed, if rats are given a very high single oral dose (1,590 mg/kg) of CTC sufficient to overwhelm the antioxidant defense mechanisms in the cell, oxidative stress can occur in brain tissue.[18] It should be noted that saturation of overall CTC metabolism in the rat is thought to occur at much lower doses.[19] This study used only a single oral dose, so information on dose-response is lacking, including whether the effects in the brain can occur at lower doses than in the liver. Nevertheless, the acute oral dose of CTC used in the Ritash *et al.* study is orders of magnitude higher than doses than are expected to occur from realistic human exposure to CTC, and therefore the study is of questionable relevance to EPA's Risk Evaluation.

Neither brain toxicity nor brain tumors have been reported in repeated dose toxicity studies on CTC. The rat and mouse 13-week and two-year inhalation studies by the JBRC did not find any treatment-related effects (cancer or non-cancer) associated with the brain or nervous system tissue. The JBRC studies were conducted according to OECD test guideline 453 issued in 1981 for "Combined Chronic Toxicity/Carcinogenicity Studies."[20] The OECD test guideline 453 requires that organ weights including the brain be collected from all animals, and that the brain (including sections of the cerebrum, cerebellum, and medulla/pons) "be preserved in the most appropriate fixation medium for both the type of tissue and the intended subsequent histopathological examination." The histopathologic examination will include the brain as specified in the test guidelines. Given that EPA considers the JBRC studies to be of high quality and the basis for its cancer risk assessment, it can be concluded that adequate data exist from animal studies to evaluate whether CTC exposure is associated with an increased incidence of brain tumors.

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[17] García-Suástegui, WA, Ramos-Chávez, LA, Rubio-Osornio, Calvillo-Velasco, M, Atzin-Méndez, JA, Guevara, J, Silva-Adaya, D, The role of CYP2E1 in the drug metabolism or bioactivation in the brain, *Oxid. Med. Cell. Longev. 2017*: 1-14 (2017).

[18] Ritash, KR, Suganya, A, Dileepkumar, HV, Rajashekar, Y, Shivanandappa, T, A single acute hepatotoxic dose of CCl_4 causes oxidative stress in the rat bran. *Toxicol. Reports* 2: 891-895 (2015).

[19]Reynolds, ES, Treinen, RJ, Farrish, HH, Metabolism of [14C] carbon tetrachloride to exhaled, excreted and bound metabolites, Biochem. Pharmacol. 33: 3363-3374 (1984); Agency for Toxic Substances and Disease Registry [ATSDR], Toxicological Profile for Carbon Tetrachloride, U.S., Department of Health and Human Services, Public Health Service (2005).

[20] Organization for Economic Cooperation and Development [OECD] Guideline for Testing of Chemicals 453: Combined Chronic Toxicity/Carcinogenicity Studies, Test, Adopted May 1981.

A

III. Exposure Assessment Limitations

A. Exposures of Non-Occupational Users (ONUs)

Monitoring data on workers at CTC production facilities were submitted to EPA as part of HSIA's comments on the CTC problem formulation document (EPA-HQ-OPPT-0733-0084). The data included personal breathing zone measurements from both workers and occupational non-users (ONUs). EPA did not note, however, that certain exposure groups (*i.e.*, process supervisors, electricians, utilities control board technicians) were indeed ONUs and wrongly concluded that exposure data for ONUs were unavailable. EPA instead estimated ONU exposures at production facilities using a "worst-case" approach in which ONUs are exposed at the central tendency exposure concentration (50th percentile) of workers. In response to this oversight, HSIA submitted to EPA ONU monitoring data of 17 breathing-zone full shift samples showing that exposures are below the detection limit (<0.063 to <0.21 ppm) (EPA-HQ-OPPT-2019-0499-0022). These data show that the potential exposure of ONUs to CTC at production facilities is extremely low, and certainly well below the "worst-case" approach used by EPA. HSIA reminds EPA to utilize these ONU monitoring data in its revision of the draft Risk Evaluation.

For estimating ONU exposure, there does not seem to be a clear science-based rationale for the use of the central tendency of the worker risk. The ratio of the high-end to central tendency values for exposures are in the range of 4-fold for manufacturing workers. Using the central tendency value as the ONU exposure estimate implies that ONU exposures are only approximately 4-fold lower than those of workers in the near field. As noted above this implication does not align with the data provided to EPA. To the extent there are residual data needs for ONUs a more appropriate approach to estimate the ONU exposures (in the absence of adequate ONU monitoring data) is the use of ONU-specific exposure models. For example, a cursory evaluation of near and mid-field plume model shows a large drop off in concentration with distance (see Table 1), further demonstrating that the approach included in the assessment is likely a significant overestimate of ONU exposure.

Table 1. Near-field Plume Concentration Estimates for Hypothetical Carbon Tetrachloride Inhalation Exposure Scenario[a]

Distance from Source (m)	Concentration (mg/m^3)	Fold Difference
0.1 (minimum in model)	29,600	-
0.25	8,090	3.7
0.5	2,260	13.1
1	631	46.9
2	176	168

[a] Estimated using the same high-end estimate for generation rate (3,738 m^3/min) and wind velocity (2.23 m/s) as provided in Appendix D of the Supplemental File: Occupational Exposure estimate.

Use of the same generation rate and air speed calculated for the Tank Truck and Railcar Loading and Unloading Release and Inhalation Exposure Model in the near-field plume model results in a nearly 50-fold reduction in concentration at a distance of 0.1 to 1 meter from the source. Since the calculated cancer risk for inhalation is close to the benchmark value, improvement of the assumptions regarding the mid- and far-field exposures would have a major impact on the risk characterization for cancer inhalation. Moreover, to the degree that ONUs are in close proximity to generation sources, the same respiratory protection requirements prescribed for workers would apply as is typical practice in industrial hygiene programs. These programs assign protective equipment requirements for work zones as part of the job hazard analysis method. Thus, the assumption of significant exposures in the absence of respiratory protection is not consistent with current industrial practice.

B. Industrial Hygiene Practices at CTC Production Facilities

HSIA also submitted a description of the industrial hygiene practices at CTC production facilities, including details on tasks by exposure groups and generalized PPE requirements (EPA-HQ-OPPT-2019-0499-0029). We would like to emphasize several important items:

- CTC production is in a closed system process unit not enclosed in a building. Sample and disposal are the only production tasks not in a closed system and are of short duration (15-30 min) and performed by trained employees wearing appropriate PPE.

- ONUs are required to wear the same PPE as a workers when present in the same immediate work environment requiring PPE use.

- Maintenance employees performing tasks outside the production area are in a barricaded area; must wear appropriate PPE; and undergo real-time monitoring to ensure the adequacy of the barricaded area.

C. Worker Dermal Exposure and Risk Characterization

POD for Chronic Dermal Exposures (Non-Cancer)

EPA uses Equation 3-2 (page 134 of the draft Risk Evaluation) to calculate a POD for chronic dermal exposures for a non-cancer endpoint. In this equation the dermal absorption factor is eliminated because an external inhalation exposure concentration is extrapolated to a dermal retained dose. Based on the information provided in the Risk Evaluation, the HED_{dermal} is 31.1 mg/m^3 x 1.25 m^3/hour x 8 hours/day x 0.63 retained inhaled dose fraction / 80 kg = 2.45 mg/kg-day. EPA seems to have used the percent value (63%) rather than the fraction (0.63), resulting in the HED_{dermal} being 100-fold greater or 245 mg/kg-day.

Calculation of the dermal cancer slope factor (CSF)

A similar calculation error has also been made for the dermal CSF: the adjustment for inhalation absorption should be 1/0.63, not 1/63. Thus, the correct dermal CSF is 8×10^{-2} per mg/kg-day. It is also unclear why EPA referred to a value of 0.8% to adjust the IUR for dermal

absorption (see page 149). The dermal CSF calculated in the Risk Evaluation is based on the retained dose from inhalation exposure and is used to calculate risk from retained dose from dermal exposure assuming 4% absorption. Use of 0.8% dermal absorption rather than the 4% value results in an additional 5-fold reduction in risk.

Risk Characterization (Dermal)

Using the corrected POD, the MOE values for central tendency and high-end exposure without gloves are 6.62 and 2.23, respectively, for chronic dermal exposures. These values fall below the benchmark Margin of Exposure (MOE) of 30-fold. However, for the relevant uses, such as in production facilities, it is anticipated that worker populations would be included in industrial hygiene programs that ensure appropriate PPE use and training (see Section III.B). Thus, with the protection factor of 20-fold for appropriate glove use both the central tendency and high-end are above a MOE of 30.

For the cancer risk estimate, the corrected dermal slope factor is 8×10^{-2} per mg/kg-day as retained dose. Based on the estimated dermal chronic retained dose for cancer of 0.1 mg/kg-day for central tendency and 0.39 mg/kg-day high end, the corresponding risk estimates are 8×10^{-3} and 3×10^{-2}, respectively. Thus, as with the chronic non-cancer endpoint, appropriate glove use in a production facility with a protection factor of 20 would result in cancer risk close to the 1×10^{-4} for the central tendency and slightly above for the high end dermal exposures.

Considering the significant conservatism in the dermal exposure assumptions, the likely actual estimates for dermal cancer risk would be below the 1×10^{-4} benchmark. For most tasks that involve dermal exposure in relevant chemical manufacturing scenarios (*e.g.*, sampling a process line or hooking up a transfer line) there is no likely routine skin contact and certainly not hours each day. In most routine tasks with any liquid present, chemical protective gloves would be used. Any liquid spills will land on the outside of a glove and largely evaporate. The full hand surface (or two full hands) would never be covered with liquid under any normal routine scenario.

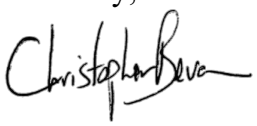
IV. Conclusion

In sum, the "applicable requirements of TSCA § 6," with which the Lautenberg Act mandates that a completed risk assessment must comply before it can support § 6 rulemaking, include taking into account exposure under the conditions of use, describing the weight of the scientific evidence for the identified hazard and exposure, the use of scientific information employed in a manner consistent with the best available science, the consideration of variability and uncertainty in the information, and consideration of the extent of independent verification or peer review of the information.

Regrettably, the draft Risk Evaluation does not fulfill the requirements of the Lautenberg Act. Its hazard assessment is not based on the best available science; its exposure assessment does not utilize all of the available occupational exposure information; and it does not reflect the current industrial hygiene practices in place at facilities where CTC is produced. To maintain the credibility of its regulatory efforts under TSCA, it is imperative

that EPA build upon the available information to construct a more realistic risk assessment before proceeding with rulemaking.

Sincerely,



Christopher Bevan, PhD, DABT
Director, Scientific Programs

Attachment

Burns Epidemiology Consulting, LLC

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Title: **Summary of carbon tetrachloride epidemiology studies of adult brain cancer and childhood neuroblastoma**

Date: March 23, 2020.

Client: HSIA, contact Christopher Bevan

Below are summary comments regarding 5 individual studies. Review comments of each study are provided in the Appendix.

Summary

The TSCA Risk Evaluation presents results for four studies published since 2010 of cancers of the nervous system within Table 3-8. In the accompanying Systematic Review Supplemental File: Data Quality Evaluation of Epidemiological Studies, the EPA evaluated the quality of these studies as well as the 1994 publication of risk of astrocytic brain cancer by Heineman et al. (1994). While the EPA reviewed each study across six domains with respect to quality and risk of bias, there was no discussion regarding causal inference or weight of evidence across studies. The nine viewpoints of Sir Bradford Hill provide useful context to evaluate a body of literature with respect to strength of association, consistency, and biological plausibility, for example (Hill 1965). Considering the risk of bias, lack of consistency and high contribution of chance and confounding, these 5 studies do not demonstrate an increased risk of cancers of the brain and nervous system and carbon tetrachloride.

One important viewpoint in evaluating causality is "strength of the association". As discussed by Fedak et al. (2015), the interpretation of "strength" goes beyond *magnitude* and should consider statistical and biological significance. Statistical testing is influenced by the sample size and number of exposed in the study population. Small studies of rare diseases like brain cancer, and uncommon exposures, such as chlorinated solvents, are subject to random variability resulting in artificially high-risk estimates. The phenomenon of effect size magnification has been recognized by the EPA's Office of Pesticide Programs in their 2016 Framework for Incorporating Human Epidemiologic & Incident Data in Risk Assessments for Pesticides (US Environmental Protection Agency 2016). These outcomes, while statistically significant, are often imprecise with very wide confidence intervals.

One means by which to quantify the lack of precision in low powered studies is to calculate the ratio of the upper and lower confidence limits (Poole 2001). The confidence interval (CI) ratio being used as an evaluation tool by the National Toxicology Program (Office of Health Assessment and Translation 2015). The NTP OHAT guidelines deems the risk of bias to be "very serious" if the CI ratio is >= 10. Other reviewers have considered the measures to be precise if the CI ratio is below 4 (Schinasi and Leon 2014).

The results for carbon tetrachloride and brain and nervous system cancers are shown in Table 1, with the CI ratio shown. Considering these concepts, the associations of cancers of the brain and nervous system as reviewed by the EPA and presented in the Table 1, are either imprecise or do not show an increased risk for the exposure estimate with the highest probably of exposure and controlling for potential confounding.

Study 1. Heineman et al., 1994. There is no increased risk of astrocytic brain cancer when limited to subjects with high probability of exposure (OR = 0.8) and when controlling for other solvents.

Study 2. Heck et al., 2013. This is a study of childhood neuroblastoma and *in utero* exposure based upon the maternal address. Note that this is a cancer nerve tissue, not of the brain. This should not be considered as a brain cancer with the other studies of adult occupational exposure to carbon tetrachloride. Although this study has more than 14,000 control subjects the risk estimates are imprecise and as noted by the EPA, the exposure assessment may be biased.

Study 3. Nelson et al., 2012. Based upon a single case with high occupational exposure this study of glioblastoma multiforme suffers from low statistical power and imprecision.

Study 4. Neta et al. 2012. This study of glioma and meningioma was inconclusive. As shown in Table 1, the choice of referent group is highly influential (OR = 1.1 when comparing to unexposed vs. OR = 7.1 when comparing to low exposed). The risk estimates when comparing high to low exposed are statistically significant but imprecise. No associated was observed for meningioma and carbon tetrachloride.

Study 5. Ruder et al. 2013. No increased risk was observed for gliomas and exposure to carbon tetrachloride. These results were not statistically significant, but the CI ratio was < 4, indicating precision.

Table 1: Comparisons of risk estimates and precision across studies

Reference	Exposure	N exposed cases	Risk estimate	95% CI	CI ratio
Study 1 Astrocytic brain cancer (Heineman et al. 1994)	Ever exposed	137	OR = 1.2	0.9 – 17	18.9
	High probability of exposure	13	OR = 0.8	0.4 – 1.9	4.8
	High intensity exposure vs. Low/Med (Table 3)	22	OR = 2.9	1.2 – 7.1	5.9
	High probability of exposure, controlling for other solvents (Table 4)	13	OR = 0.6	0.2 – 1.7	8.5
	High intensity exposure vs. Low/Med, with 21+ y duration, controlling for other solvents (Table 5)	NR	OR = 1.0	0.2 – 7.3	36.5
Study 2 Neuroblastoma (Heck et al. 2013)	Residence 5 km from monitor (Model 2)	40	OR = 2.65	1.07 – 6.53	6.10
	Residence 2.5 km from monitor (Model 2)	12	OR = 7.87	1.37 – 45.34	33.09
	Highest quartile compared to lowest	NR	OR = 8.85	1.19 – 66.0	55.46
Study 3 Glioblastoma multiforme (Nelson et al. 2012)	Exposed vs. unexposed	2	RR = 10.09	NR	NR
	High occupational exposure vs. no exposure	1	HR = 26.59	2.9 – 243.50	9.16
Study 4 Glioma and meningioma (Neta et al. 2012)	Probable exposure	15 gliomas	OR = 0.6	0.3 – 1.2	4.0
	Higher average weekly exposure vs. unexposed	11 gliomas	OR =1.1	0.5 – 2.4	4.8
	Higher average weekly exposure vs. low exposed	11 gliomas	OR = 7.1	1.1 – 45.2	41.1
	Higher average weekly exposure, controlled for lead and magnetic fields	11 gliomas	OR = 60.2	2.4 – 1533.8	639.1
	Probable exposure	7 meningiomas	OR = 0.6	0.1 – 2.9	29.0
Study 5 Glioma (Ruder et al. 2013)	Ever occupational exposure vs. Never; excluding proxy	141	OR = 0.82	0.64 – 1.06	1.66
	Ever occupational exposure vs. Never; excluding proxy; cumulative exposure	NR	OR = 0.98	0.96 – 1.00	1.04
	Ever occupational exposure vs. Never; (including proxy)	263	OR = 0.79	0.65 – 0.97	1.49

	Ever occupational exposure vs. Never; including proxy; cumulative exposure	NR	OR = 0.98	0.96 – 0.99	1.03

NR: not reported; OR: odds ratio; HR: Hazard ratio; RR: Relative risk

Appendix 1. Reviews of Individual studies

Heineman et al. (1994) Occupational exposure to chlorinated aliphatic hydrocarbons and risk of astrocytic brain cancer

Abstract from publication: Chlorinated aliphatic hydrocarbons (CAHs) were evaluated as potential risk factors for astrocytic brain tumors. Job-exposure matrices for six individual CAHs and for the general class of organic solvents were applied to data from a case-control study of brain cancer among white men. The matrices indicated whether the CAHs were likely to have been used in each industry and occupation by decade (1920-1980) and provided estimates of probability and intensity of exposure for "exposed" industries and occupations. Cumulative exposure indices were calculated for each subject. Associations of astrocytic brain cancer were observed with likely exposure to carbon tetrachloride, methylene chloride, tetrachloroethylene, and trichloroethylene, but were strongest for methylene chloride. Exposure to chloroform or methyl chloroform showed little indication of an association with brain cancer. Risk of astrocytic brain tumors increased with probability and average intensity of exposure, and with duration of employment in jobs considered exposed to methylene chloride, but not with a cumulative exposure score. These trends could not be explained by exposures to the other solvents.

EPA review extract (from EPA 2020 Systematic Review Supplemental File: Data Quality Evaluation of Epidemiological Studies)

Domain	Metric	Rating
2 Exposure	4 Measurement of exposure	Low
	5 Exposure levels	Medium
	6 Temporality	Low
3 Outcome	7 Outcome measurement	Medium
	8 Reporting bias	Medium
1 – 6	Overall Quality Evaluation	Medium

Comments. The case control study by Heineman et al. (1994) was exploratory research based upon deaths that occurred in geographic areas presumed to have a high opportunity for occupational exposure to chlorinated aliphatic hydrocarbons. Subjects were identified from death certificates from the period 1978 – 1981. Controls were similarly selected, based on noncancer causes of death. Interviews were conducted with next of kin. The exposure period ranged from 1920 to 1980.

Limitations. There are several limitations to this study and the approach. 1) All of the exposure and lifestyle information was based upon interviews with a proxy (i.e. not the subject himself). There is likely to be incorrect recall especially for jobs in the distance past. 2) The data available on each job lacked specificity for unique solvents and poor temporal detail. As noted by the EPA, the exposure measurement was low quality due to the opportunity for co-exposure to several solvents. 3) The overall participation was poor. From 741 men who died of brain cancer only 300 (40.49%) next of kin were identified and participated. The participation rate for controls was slightly higher (N = 320, 43.18%). This reduces the sample size and may introduce bias if participation was influenced by perception of exposure.

Strengths. Certain analytical approaches of the research are robust. For example, the authors categorized exposure to carbon tetrachloride by the probability that the substance was used by a worker (low, medium, high). Reporting by probability permits the reader to evaluate results for the group with the highest confidence of exposure.

Results

Reference	Exposure	N exposed cases	Risk estimate	95% CI	CI ratio
(Heineman et al. 1994)	Ever exposed	137	OR = 1.2	0.9 – 17	18.9
	High probability of exposure	13	OR = 0.8	0.4 – 1.9	4.8
	High intensity exposure vs. Low/Med (Table 3)	22	OR = 2.9	1.2 – 7.1	5.9
	High probability of exposure, controlling for other solvents (Table 4)	13	OR = 0.6	0.2 – 1.7	8.5
	High intensity exposure vs. Low/Med, with 21+ y duration, controlling for other solvents (Table 5)	NR	OR = 1.0	0.2 – 7.3	36.5

WOE. The sample size is greatly reduced from "ever" exposed to "high probability" of exposed. The risk estimates for the high probability group are likely to be the highest quality. Other risk estimates of interest are those for which co-exposures to other solvents are controlled. Most of these analyses show no excess risk and are not statistically significant.
Based on these estimates, the evidence does not support an increased risk of brain cancer and carbon tetrachloride.

Heck et al. (2013) An exploratory study of ambient air toxics exposure in pregnancy and the risk of neuroblastoma in offspring

Abstract from publication:

Little is known about the etiology of neuroblastoma, the most common cancer in infancy. In this study, we examined maternal exposure to ambient air toxics in pregnancy in relation to neuroblastoma in the child. We ascertained all cases of neuroblastoma listed in the California Cancer Registry 1990-2007 that could be linked to a California birth certificate, and controls were selected at random from California birth records. Average air toxics exposures during pregnancy were determined based upon measures from community-based air pollution monitors. The study included 75 cases and 14,602 controls who lived with 5 km of an air pollution monitor, and we additionally examined results for those living within a smaller radius around the monitor (2.5 km). Logistic regression was used to determine the risk of neuroblastoma with one interquartile range increase in air toxic exposure. Neuroblastoma risk was increased with higher maternal exposure to carbon tetrachloride (OR=2.65, 95%CI 1.07, 6.53) and polycyclic aromatic hydrocarbons (OR=1.39, 95%CI 1.05, 1.84), particularly indeno(1,2,3-cd)pyrene and dibenz(a,h)anthracene. Hexavalent chromium was associated with neuroblastoma at the 5 km distance (OR=1.32, 95%CI 1.00, 1.74) but not at the 2.5 km distance. This is one of the first studies to report associations between neuroblastoma and these air toxics.

EPA review extract (from EPA 2020 Systematic Review Supplemental File: Data Quality Evaluation of Epidemiological Studies)

Domain	Metric	Rating
2 Exposure	4 Measurement of exposure	Low
	5 Exposure levels	Medium
	6 Temporality	High
3 Outcome	7 Outcome measurement	Medium
	8 Reporting bias	Medium
1 – 6	Overall quality rating	Medium

Comments. The case control study of Heck et al., (2013) is a record linkage study relying on data from the California Cancer Registry (to identify neuroblastoma cases), birth certificates (to identify controls and determine addresses) and air pollution monitors. The study included 75 cases and more than 14,000 controls.

Limitations. As noted by the EPA, exposure assessment was low quality. Exposure to an air pollutant was inferred based only upon residence at birth. The authors used data from the 39 statewide monitors of the California Air Resources Board's Air Toxics Program, available for the study period 1990 to 2007. These locations were linked to addresses provided on the birth certificate. It is notable that for the period 1990 – 1998, only zip code was available. This exposure assessment makes several assumptions:

- The mother lived at the same address during the entire pregnancy. [note the authors cite research that 9 – 30% of families may move during pregnancy]
- The mother did not spend time away from the address, (i.e. the site monitor) such as for school or employment.
- Exposure levels were constant throughout the measurement period.

Building on the limited assumption of residence and exposure, the authors' methods for calculating the mean concentrations were vague. The authors used at least one reading for each full month of the pregnancy, but it is not clear how a summary concentration was computed. No

information was provided for the actual concentrations of carbon tetrachloride (and other pollutants) over time or by location.
Sensitivity analysis would have added confidence to the results. As noted above the authors recognize in their discussion that the address on the birth certificate may not reflect the residence during pregnancy for up to 30% of women. Analytic techniques are available to model the impact of greater or lesser mobility upon the exposure-outcome models. With the disproportionate number of cases (N = 75) to controls (N = 14,602) a small change in exposure (more or less) to cases might have a large impact on the odds ratio. Lastly, the authors provide no results separately for births with addresses (born 1999 – 2007) and those based on zip code (born 1990 – 1998) to evaluate if this impacted the risk estimates.
Strengths. The EPA rated this study as Medium quality based upon adequate design, temporality, and monitoring data for specific exposures. Record linkage studies are not subject to participation rate and recall bias. By relying on existing records, the authors were able to have unlimited controls to increase statistical power and did not need to contact individual subjects. The exposure metrics were based on actual stationary monitors, omitting the need for self-reporting of exposure and/or job history. Further, with the use of monitors, concentrations were specific to carbon tetrachloride (vs. chlorinated solvents).
Table Results

Reference	Exposure	N exposed cases	Risk estimate	95% CI	CI ratio
(Heck et al. 2013)	Residence 5 km from monitor (Model 2)*	40	OR = 2.65	1.07 – 6.53	6.10
	Residence 2.5 km from monitor (Model 2)*	12	OR = 7.87	1.37 – 45.34	33.09
	Highest quartile compared to lowest	NR	OR = 8.85	1.19 – 66.0	55.46

WOE Conclusion. As noted by EPA, the study quality was Medium. Because of limitations in exposure assessment, it is likely that misclassification occurred. As shown in the Results table, the risk estimates are statistically significant and higher for residences closer to the monitor and in the highest quartile. However, there were only 12 exposed cases in the 2.5 km analysis and the results are imprecise. Further, it is unknown how the children born in the 1990 – 1998 period, for whom only zip code was available, were included in these analyses.
There are two additional studies of neuroblastoma and air pollutants that are informative. Thompson et al. (2008) evaluated nonspecific hazardous air pollutants (HAP)and 19 different childhood cancers. They reported a nonsignificant odds ratio of 1.2 (95% CI 0.8 – 1.6) for HAP and neuroblastoma. A study looking at parental occupational exposures reported no increased risk of carbon tetrachloride and neuroblastoma, OR 0.4, 95% CI 0.2 – 1.2) (De Roos et al. 2001).
The evidence from Heck et al. (2013) is inconclusive due to small number of exposed cases, poor precision in risk estimates, and low-quality exposure assessment.

Nelson et al. (2012): Potential risk factors for incident glioblastoma multiforme: The Honolulu Heart Program and Honolulu-Asia Aging Study.

Abstract from publication:

Glioblastoma multiforme (GBM) is the most common adult primary malignant brain tumor. Ninety percent of adult GBM patients die within 24 months after diagnosis. The etiology of GBM is unknown. The Honolulu Heart Program (HHP) and Honolulu-Asia Aging Study (HAAS) are prospective, cohort studies of cardiovascular and neurodegenerative disease based on 8,006 Japanese-American men followed since 1965. The Japan Hawaii Cancer Study provides data on incident cancer cases in the HHP/HAAS cohort. We used data from these studies to obtain epidemiologic information about GBM. GBM cases were identified by searching the 1965-1998 databases using International Classification of Diseases (ICD-9) codes. Nine histologically confirmed GBM cases, 58-80 years old, were identified. The incidence rate was 6.2/100,000 person-years. Records of each case were reviewed. Selected variables from the first three examinations (1965-1968; 1968-1970; 1971-1974) were used to identify potential candidate GBM risk factors. A multivariate Cox proportional hazards model showed sugar intake and occupational exposure to carbon tetrachloride were independently and significantly associated with development of GBM.

EPA review extract (from EPA 2020 Systematic Review Supplemental File: Data Quality Evaluation of Epidemiological Studies)

Domain	Metric	Rating
2 Exposure	4 Measurement of exposure	Low
	5 Exposure levels	Medium
	6 Temporality	Medium
3 Outcome	7 Outcome measurement	High
	8 Reporting bias	High
1 – 6	Overall quality rating	Medium

Comments. The retrospective cohort study of Japanese American men who died between 1974 and 1995 by Nelson et al. (2012) is based upon occupational histories collected during periodic examinations. All subjects were part of a cohort study of 8006 men followed for about 30 years. Cases were determined from histologic exams. Exposure was determined by industrial hygienists' review of the occupation/industry combination. Note that while this analysis was published in 2012, the data are based on deaths that occurred 17 to 38 years prior, and putative exposure in the far distance past.

Limitations. The statistical power was low due to the small number of glioblastoma multiforme cases, N = 9. Only two cases had probable exposure to carbon tetrachloride. As noted by EPA's review, the exposure measurement is low quality. The authors note that exposed occupations included dry cleaners, firemen, chemists, machinists, and radio/TV repairmen but do not elaborate more on the nature of exposure to solvents in these jobs.

Strengths. The data were collected prospectively before the subjects were ill. This reduces the problem of information bias and low participation rates.

Results

Reference	Exposure	N exposed cases	Risk estimate	95% CI	CI ratio
(Nelson et al. 2012)	Exposed vs. unexposed	2	RR = 10.09	NR	NR
	High occupational exposure vs. no exposure	1	HR = 26.59	2.9 – 243.50	9.16

WOE. As noted by EPA, the study quality was Medium with an adequate design, high outcome ascertainment and a specific exposure metric. Because of limitations in exposure assessment, it is likely that misclassification occurred. As shown in the Results table, the risk estimates are statistically significant but imprecise. The Hazard Ratio is based on a single exposed case.
The evidence from Nelson et al. (2012) is inconclusive due to small number of exposed cases, poor precision in risk estimates, and low-quality exposure assessment.

Neta et al. (2012): Occupational exposure to chlorinated solvents and risks of glioma and meningioma in adults

Abstract from publication:

OBJECTIVES: Chlorinated solvents are classified as probable or possible carcinogens. It is unknown whether exposure to these agents increases the risk of malignant or benign brain tumours. Our objective was to evaluate associations of brain tumour risk with occupational exposure to six chlorinated solvents (i.e., dichloromethane, chloroform, carbon tetrachloride, 1,1,1-trichloroethane, trichloroethylene and perchloroethylene). METHODS: 489 glioma cases, 197 meningioma cases and 799 controls were enrolled in a hospital-based case-control study conducted at three U.S.A. hospitals in Arizona, Massachusetts and Pennsylvania. Information about occupational history was obtained through a detailed in person interview that included job-specific modules of questions such that the interview was tailored to each individual's particular work history. An industrial hygienist assessed potential solvent exposure based on this information and an exhaustive review of the relevant industrial hygiene literature. Unconditional logistic regression models were used to calculate OR and 95% CI for each solvent for ever/never, duration, cumulative, average weekly and highest exposure. RESULTS: Overall, we found no consistent evidence of an increased risk of glioma or meningioma related to occupational exposure to the six chlorinated solvents evaluated. There was some suggestion of an association between carbon tetrachloride and glioma in analyses restricted to exposed subjects, with average weekly exposure above the median associated with increased risk compared with below the median exposure (OR = 7.1, 95% CI 1.1 to 45.2). CONCLUSIONS: We found no consistent evidence for increased brain tumour risk related to chlorinated solvents.

EPA review extract (from EPA 2020 Systematic Review Supplemental File: Data Quality Evaluation of Epidemiological Studies)

Domain	Metric	Rating
2 Exposure	4 Measurement of exposure	Low
	5 Exposure levels	Medium
	6 Temporality	High
3 Outcome	7 Outcome measurement	High
	8 Reporting bias	High
1 – 6	Overall quality rating	High

Comments. The study of Neta et al. (2012) is a case control study of 484 glioma cases, 197 meningioma cases and 797 controls. Both groups of cases and controls were identified between 1994 and 1998 from Boston, Massachusetts hospitals. Exposure was assessed by industrial hygienists' interpretations of the lifetime occupational histories from 1930 to 1998. The probability, frequency and intensity of exposure to carbon tetrachloride was also assessed by decade. Note that while this analysis was published in 2012, the data are based on diagnoses that occurred 14 - 18 years prior, and putative exposure(s) are in the far distance past.
Limitations. Differential information bias may have occurred from the cases being more motivated to contribute detailed occupational information. As noted by EPA's review, the exposure measurement is low quality. Exposure to carbon tetrachloride was based upon the job history and not any information solvents from the study subjects.
Strengths. The cases were identified and enrolled in the study very quickly. The study was of incident cases not deaths. This reduced the number of proxy interviews required due to death or illness. Note that 16% of glioma, 8% of meningioma cases and 3% of controls were proxy interviews. The authors conducted sensitivity analyses to test various hypotheses and reran different statistical models.

Results

Reference	Exposure	N exposed cases	Risk estimate	95% CI	CI ratio
(Neta et al. 2012)	Probable exposure	15 gliomas	OR = 0.6	0.3 – 1.2	4.0
	Higher average weekly exposure vs. unexposed	11 gliomas	OR =1.1	0.5 – 2.4	4.8
	Higher average weekly exposure vs. low exposed	11 gliomas	OR = 7.1	1.1 – 45.2	41.1
	Higher average weekly exposure, controlled for lead and magnetic fields	11 gliomas	OR = 60.2	2.4 – 1533.8	639.1
	Probable exposure	7 meningiomas	OR = 0.6	0.1 – 2.9	29.0

WOE. The authors conducted analyses in two ways. One, using unexposed as the referent and another using low exposed as a referent. Their rationale was provided that "unexposed persons may be substantially different from exposed persons in ways that cannot be adjusted for in our analysis." However, they do not discuss how or why this may occur.
As noted by EPA, despite a low score of exposure assessment, the overall study quality was High with an adequate design, high outcome ascertainment and a specific exposure metric. Because of limitations in exposure assessment, it is likely that misclassification occurred. As shown in the Results table, choice of referent group is highly influential. The risk estimates when comparing high to low exposed are statistically significant but imprecise. No associated was observed for meningioma and carbon tetrachloride.
The evidence from Nelson et al. (2012) is inconclusive due to small number of exposed cases, poor precision in risk estimates, and low-quality exposure assessment.

Ruder et al. (2013): The Upper Midwest Health Study: gliomas and occupational exposure to chlorinated solvents

Abstract from the publication

OBJECTIVES: Occupational exposure to chlorinated aliphatic solvents has been associated with an increased cancer risk, including brain cancer. However, many of these solvents remain in active, large-volume use. We evaluated glioma risk from non-farm occupational exposure (ever/never and estimated cumulative exposure) to any of the six chlorinated solvents--carbon tetrachloride, chloroform, methylene chloride, trichloroethylene, tetrachloroethylene or 1,1,1--trichloroethane-among 798 cases and 1175 population-based controls, aged 18-80 years and non-metropolitan residents of Iowa, Michigan, Minnesota and Wisconsin. Methods Solvent use was estimated based on occupation, industry and era, using a bibliographic database of published exposure levels and exposure determinants. Unconditional logistic regression was used to calculate ORs adjusted for frequency matching variables age group and sex, and age and education. Additional analyses were limited to 904 participants who donated blood specimens (excluding controls reporting a previous diagnosis of cancer) genotyped for glutathione-S-transferases GSTP1, GSTM3 and GSTT1. Individuals with functional GST genes might convert chlorinated solvents crossing the blood-brain barrier into cytotoxic metabolites. RESULTS: Both estimated cumulative exposure (ppm-years) and ever exposure to chlorinated solvents were associated with decreased glioma risk and were statistically significant overall and for women. In analyses comparing participants with a high probability of exposure with the unexposed, no associations were statistically significant. Solvent-exposed participants with functional GST genes were not at increased risk of glioma. CONCLUSIONS: We observed no associations of glioma risk and chlorinated solvent exposure. Large pooled studies are needed to explore the interaction of genetic pathways and environmental and occupational exposures in glioma aetiology.

EPA review extract (from EPA 2020 Systematic Review Supplemental File: Data Quality Evaluation of Epidemiological Studies)

Domain	Metric	Rating
2 Exposure	4 Measurement of exposure	Medium
	5 Exposure levels	Medium
	6 Temporality	Medium
3 Outcome	7 Outcome measurement	High
	8 Reporting bias	High
1 – 6	Overall quality rating	High

Comments. The Upper Midwest Health Study by Ruder et al. (2013) is a case control study of 798 cases of glioma diagnosed 1989 – 1992 and 1175 population-based controls. Interviews were conducted with participants or their proxies regarding past jobs. Uniquely (i.e. compared to other studies of brain cancer and occupation), the investigators asked about exposure to solvents, "on which jobs and for how many hours a week these exposures occurred." Industrial hygienists reviewed the responses and also considered other exposure determinants such as ventilation and proximity.

Limitations. All exposure information was collected retrospectively, with a high proportion from proxies. The focus of the study was on agricultural exposures and the participants may have forgotten relevant exposed jobs. The estimates of job-based exposures to carbon tetrachloride were based upon models reported in the literature. As the authors noted, they were unable to determine if their study participants' experiences were consistent with these estimates.

Strengths. The study was based upon confirmed incident cases of glioma (vs. cases from death certificates). The authors stratified their results by respondent type (i.e. proxy) so that

information bias, if present, could be quantified. There were a large number of exposed cases permitting sufficient statistical power to evaluate solvent exposures. Genotypes for glutathione-S-transferase were evaluated to test for genetic susceptibility.

Reference	Exposure	N exposed cases	Risk estimate	95% CI	CI ratio
(Ruder et al. 2013)	Ever occupational exposure vs. Never; excluding proxy	141	OR = 0.82	0.64 – 1.06	1.66
	Ever occupational exposure vs. Never; excluding proxy; cumulative exposure	NR	OR = 0.98	0.96 – 1.00	1.04
	Ever occupational exposure vs. Never; (including proxy)	263	OR = 0.79	0.65 – 0.97	1.49
	Ever occupational exposure vs. Never; including proxy; cumulative exposure	NR	OR = 0.98	0.96 – 0.99	1.03

WOE. As noted by EPA, the overall study quality was High with an adequate design, high outcome ascertainment and a specific exposure metric. No increase in exposure to carbon tetrachloride was observed in any analysis of glioma.

The evidence from Ruder et al. (2013) suggests there is no association of occupational exposure to carbon tetrachloride and glioma.

REFERENCES

De Roos, A. J., A. F. Olshan, K. Teschke, C. Poole, D. A. Savitz, J. Blatt, M. L. Bondy, and B. H. Pollock. 2001. 'Parental occupational exposures to chemicals and incidence of neuroblastoma in offspring', *Am J Epidemiol*, 154: 106-14.

Fedak, Kristen M., Autumn Bernal, Zachary A. Capshaw, and Sherilyn Gross. 2015. 'Applying the Bradford Hill criteria in the 21st century: how data integration has changed causal inference in molecular epidemiology', *Emerging Themes in Epidemiology*, 12: 14.

Heck, J. E., A. S. Park, J. Qiu, M. Cockburn, and B. Ritz. 2013. 'An exploratory study of ambient air toxics exposure in pregnancy and the risk of neuroblastoma in offspring', *Environ Res*, 127: 1-6.

Heineman, E. F., P. Cocco, M. R. Gomez, M. Dosemeci, P. A. Stewart, R. B. Hayes, S. H. Zahm, T. L. Thomas, and A. Blair. 1994. 'Occupational exposure to chlorinated aliphatic hydrocarbons and risk of astrocytic brain cancer', *Am J Ind Med*, 26: 155-69.

Hill, A. B. 1965. 'The Environment and disease: Association or causation?', *Proc R Soc Med*, 58: 295-300.

Nelson, J. S., C. M. Burchfiel, D. Fekedulegn, and M. E. Andrew. 2012. 'Potential risk factors for incident glioblastoma multiforme: the Honolulu Heart Program and Honolulu-Asia Aging Study', *J Neurooncol*, 109: 315-21.

Neta, G., P. A. Stewart, P. Rajaraman, M. J. Hein, M. A. Waters, M. P. Purdue, C. Samanic, J. B. Coble, M. S. Linet, and P. D. Inskip. 2012. 'Occupational exposure to chlorinated solvents and risks of glioma and meningioma in adults', *Occup Environ Med*, 69: 793-801.

Office of Health Assessment and Translation, OHAT. 2015. "Handbook for conducting a literature-based health assessment using OHAT approach for systematic review and evidence integration." In, edited by Division of the National Toxicology Progam.

Poole, C. 2001. 'Low P-values or narrow confidence intervals: which are more durable?', *Epidemiology*, 12: 291-4.

Ruder, A. M., J. H. Yiin, M. A. Waters, T. Carreon, M. J. Hein, M. A. Butler, G. M. Calvert, K. E. Davis-King, P. A. Schulte, J. S. Mandel, R. F. Morton, D. J. Reding, K. D. Rosenman, P. A. Stewart, and Group Brain Cancer Collaborative Study. 2013. 'The Upper Midwest Health Study: gliomas and occupational exposure to chlorinated solvents', *Occup Environ Med*, 70: 73-80.

Schinasi, L., and M. E. Leon. 2014. 'Non-Hodgkin Lymphoma and Occupational Exposure to Agricultural Pesticide Chemical Groups and Active Ingredients: A Systematic Review and Meta-Analysis', *International Journal of Environmental Research and Public Health*, 11: 4449-527.

Thompson, J. A., S. E. Carozza, and L. Zhu. 2008. 'Geographic risk modeling of childhood cancer relative to county-level crops, hazardous air pollutants and population density characteristics in Texas', *Environ Health*, 7: 45.

US Environmental Protection Agency, USEPA. 2016. "Framework for Incorporating Human Epidemiologic & Incident Data in Risk Assessments for Pesticides." In, edited by Office of Pesticide Programs, 51.