Exhibit 19

UNITED STATES DISTRICT COURT

NORTHERN DISTRICT OF CALIFORNIA

IN RE: ROUNDUP PRODUCTS LIABILITY LITIGATION	MDL No. 2741 Case No. 16-md-02741-VC
This document relates to: ALL ACTIONS	

EXPERT REPORT OF DR. CHRISTOPHER J. PORTIER IN SUPPORT OF GENERAL CAUSATION ON BEHALF OF PLAINTIFFS

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Expert Report
Christopher J. Portier, Ph.D.

Charge

Glyphosate acid is a colorless, odorless, crystalline solid. Glyphosate is the term used to describe the salt that is formulated by combining the deprotonated glyphosate acid and a cation (isopropylamine, ammonium, or sodium). This expert report is intended to review the available scientific evidence relating to the potential of glyphosate and glyphosate-based formulations (GBFs), including Roundup®, to cause Non-Hodgkin's Lymphoma (NHL) in humans.

Qualifications

I received an undergraduate degree in mathematics in 1977 from Nicholls State University and a Master's degree and Ph.D. in biostatistics from the University of North Carolina School of Public Health in 1979 and 1981 respectively. My Ph.D. thesis addressed the optimal way to design a two-year rodent carcinogenicity study to assess the ability of a chemical to cause cancer[1, 2]; the optimal dosing pattern from my thesis is still used by most researchers. My first employment following my doctoral degree was a joint appointment at the National Institute of Environmental Health Sciences (NIEHS) and the National Toxicology Program (NTP) to conduct research on the design and analysis of experiments generally employed in toxicology. After 5 years with NIEHS/NTP, I developed my own research group which eventually became the Laboratory of Quantitative and Computational Biology and then the Laboratory of Computational Biology and Risk Assessment (LCBRA). One highlight during this period was the development of the Poly-3 Test for survival adjustment of data from two-year carcinogenicity studies in rodents[3, 4]; this test is used as the main method of analysis of these studies by the NTP and many others. We also did a complete analysis of the historical controls animals from the NTP studies[5, 6]. The LCBRA focused on the application of computational tools to identify chemicals that are toxic to humans, to develop tools for understanding the mechanisms underlying those toxicities and to quantify the risks to humans associated with these toxicities. The main toxicological focus of the LCBRA was cancer and my laboratory developed many methods for applying multistage models to animal cancer data and implemented the use of these models in several experimental settings[7-19]. In my last few years at the NIEHS/NTP, my research focus expanded to the development of tools for evaluating the response of complex experimental and human systems to chemicals[20-24] and the name of the laboratory shifted to Environmental Systems Biology.

Over my 32 years with the NIEHS/NTP, I was involved in numerous national priority issues that went beyond my individual research activities. After Congress asked NIEHS to work with the Vietnamese government to address the hazards associated with Agent Orange use during the Vietnamese War, I was given the responsibility of working with

my counterparts in Vietnam to build a research program in this area[25]. Congress also tasked NIEHS with developing a research program (EMF-RAPID) to address concerns about the risks to humans from exposure to power lines and to report back to Congress on what we found. I was in charge of evaluating all research developed under this program and was responsible for the final recommendations to Congress on this issue[26-28].

While at the NIEHS/NTP, I also had administrative positions that relate to my qualifications. From 2000 to 2006 I was the Director of the Environmental Toxicology Program (ETP) at NIEHS. The ETP included all of the toxicology research laboratories within the NIEHS Intramural Research Program. It was my responsibility to ensure the research being done was pertinent to the mission of the NIEHS, addressing high priority concerns about toxic substances and human health and that the NIEHS had adequate resources to complete this research.

During this time I was also Associate Director of the NTP, a position in which I was the scientific and administrative director of the NTP (The Director of the NTP was also the NIEHS Director and gave me complete autonomy in the management and science of the NTP). These two positions were historically always combined at the NIEHS and the NTP so that one person was in charge of all toxicological research at the NIEHS/NTP. The NTP is the world's largest toxicology program, routinely having 15 to 25 active two-year carcinogenicity studies, numerous genetic toxicology studies and many other toxicological studies being conducted at any given time. The NTP two-year carcinogenicity studies and their technical reports are also considered the "gold standard" of cancer studies due to their extreme high quality, their tremendous utility in evaluating human health hazards and the rigor and transparency they bring to the evaluation of the data. All data from NTP two-year cancer studies are publicly available including data on individual animals and images from the pathology review of each animal. The NTP is also home to the Report on Carcinogens, the US Department of Health and Human Services official list of what is known or reasonably anticipated to be carcinogenic to humans. It was my responsibility to decide what items eventually went onto this list while I was Associate Director of the NTP. In 2006, I became an Associate Director of the NIEHS, a senior advisor to the director and the director of the Office of Risk Assessment Research (ORAR). ORAR focused on stimulating new research areas on the evaluation of health risks from the environment and addressed major risk assessment issues on behalf of the NIEHS/NTP. For example, in this capacity, I lead a multiagency effort to understand the health risks to humans from climate change and to develop a research program in this area[29].

I left the NIEHS/NTP in 2010 to become the Director of the National Center for Environmental Health (NCEH) at the Centers for Disease Control and Prevention and simultaneously Director of the Agency for Toxic Substances and Disease Registry (ATSDR). NCEH does research and supports activities aimed at reducing the impact of environmental hazards on public health. One well-respected research effort of the NCEH is the National Biomonitoring Program. This program tests for the presence of hundreds of chemicals in human blood and urine in a national sample of people in the

United States. ATSDR advices the Environmental Protection Agency (EPA) and communities on the potential health impacts from toxic waste dump sites (superfund sites). ATSDR is required by law to produce ToxProfiles. These are comprehensive reviews of the scientific literature for specific chemicals generally found at superfund sites. They also provide an assessment of the safety of these chemicals. As part of my activities at ATSDR, I began a modernization of the ToxProfiles to use systematic review methods in their assessments; this effort was linked to a similar effort that I had helped to implement at the NIEHS/NTP.

Aside from my official duties in my various federal jobs, I also served on numerous national and international science advisory panels. Most notable, for my qualifications for this statement, are my serving as Chair from 2005 to 2010 of the Subcommittee on Toxics and Risk of the President's National Science and Technology Council, member and chair of EPA'S Science Advisory Panel from 1998 to 2003 (focused specifically on advising their pesticides program) and chair of the International Agency for Research on Cancer (IARC) advisory group that updated and improved its rules for reviewing scientific data to ensure that conclusions on the carcinogenicity of human exposures are the best possible (Preamble)[30]. As part of my work on science advisory panels, I have served on EPA's Science Advisory Board, as an advisor to the Australian Health Council on risk assessment methods, as an advisor to the Korean Food and Drug Administration on toxicological methods, and served on several World Health Organization (WHO) International Program on Chemical Safety scientific panels dealing with risk assessment. Besides the guidelines for evaluating cancer hazards used by the IARC, I have either chaired or served as a member of scientific panels developing guidance documents for other organizations including the EPA.

I have received numerous awards, most notably the Outstanding Practitioner Award from the International Society for Risk Analysis and the Paper of the Year Award (twice) from the Society of Toxicology Risk Assessment Specialty Section. I am a fellow of the American Statistical Association, the International Statistical Institute, the World Innovation Foundation and the Ramazinni Institute. I have published over 250 peer-reviewed scientific papers, book chapters and technical documents on topics in toxicology and risk assessment.

Finally, I have served on numerous national and international committees tasked with evaluating the risk and/or hazard of specific environmental chemicals, including glyphosate. For example, I have contributed to risk assessments for EPA, the Food and Drug Administration, the Centers for Disease Control and Prevention, the National Institutes of Health, the WHO and IARC.

Reliance List

During the course of my preparation for this report, I have reviewed the following materials:

a. All epidemiological data relating to the ability of glyphosate formulations to cause NHL in humans.

b. Scientific papers on the cellular origins of NHL
c. Peer-reviewed scientific data relating to the carcinogenicity, genotoxicity and oxidative stress caused by glyphosate
d. Technical reports relating to the carcinogenicity of glyphosate provided by the defendant to the lawyers for the plaintiff
e. The USEPA, the European Food Safety Authority (EFSA), the German Federal Institute for Risk Assessment, the European Chemical Agency, the IARC and the WHO/Food and Agriculture Organization Joint Meeting on Pesticide Residues reviews of the scientific literature relating to the potential for glyphosate to cause cancer.
f. Technical documents available from EFSA regarding animal carcinogenicity data on glyphosate prepared by organizations other than the defendant
g. Various other documents produced in the litigation

A complete list of my reliance materials is at the end of this report.

Methodology for Causality Evaluation

The evaluation of whether glyphosate and/or GBFs can cause NHL in humans requires the review and synthesis of scientific evidence from studies of human populations (epidemiology), animal cancer studies, and studies investigating the mechanisms through which chemicals cause cancer. Many different approaches[31, 32] are used to synthesize these three areas of science to answer the question "Does this chemical cause cancer in humans?" In any of these three science areas, the quality of the individual studies has to be assessed and summarized to make certain the studies included in the overall assessment are done appropriately. Once the quality of the individual studies has been assessed, a judgment needs to be made concerning the degree to which the studies support a finding of cancer in humans. To do this, the EPA, IARC, the European Chemical Agency (EChA), the US Report on Carcinogens, and many others use guidelines[30, 33-35] that rely upon aspects of the criteria for causality developed by **Hill (1965)**[36].

Hill listed nine (9) aspects of epidemiological studies and the related science that one should consider in assessing causality. The presence or absence of any of these aspects is neither sufficient nor necessary for drawing inferences of causality. Instead, the nine aspects serve as means to answer the question of whether other explanations are more credible than a causal inference. As noted by Hill:

> *"None of my nine viewpoints can bring indisputable evidence for or against the cause-and-effect hypothesis and none can be required as a sine qua non. What they can do, with greater or less strength, is to help us to make up our minds on the fundamental question — is there any other way of explaining the set of facts before us, is there any other answer equally, or more, likely than cause and effect?"*

The nine aspects cited by Hill include consistency of the observed association, strength

of the observed association, biological plausibility, biological gradient, temporal relationship of the observed association, specificity of the observed association, coherence, evidence from human experimentation and analogy. These are briefly described below.

An inference of causality is strengthened when several of the studies show a **consistent positive association** between cancer and the exposure. This addresses the key issue of replication of studies which is critical in most scientific debates. If studies are discordant, differences in study quality, potential confounding, potential bias and statistical power are considered to better understand that discordance.

An inference of causality is strengthened when the **strength of the observed association** in several studies are large and precise. These large, precise associations lessen the possibility that the observed associations are due to chance or bias. A small increase in risk of getting cancer does not preclude a causal inference since issues such as potency and exposure level may reduce the ability of a study to identify larger risks. Meta-analyses provide an objective evaluation of the strength of the observed association across several studies with modest risks to help clarify strength of the observed associations.

An inference of causality is strengthened when there is data supporting **biological plausibility** demonstrated through experimental evidence. Animal carcinogenicity studies, in which tumor incidence is evaluated in experimental animals exposed to pure glyphosate, play a major role in establishing biological plausibility. There are numerous types of mechanisms that can lead to cancer[37], most of which can be demonstrated through experimental studies in animals, human cells, animal cells, and/or other experimental systems. Occasionally, occupational, accidental or unintended exposures to humans allow researchers to evaluate mechanisms using direct human evidence.

An inference of causality is strengthened when there is a **biological gradient** showing a reasonable pattern of changing risk with changes in exposure (e.g. risk increases with increasing exposure or with longer exposure). In many epidemiological studies, this aspect cannot be examined due to limitations in the study design or due to a lack of clarity in the presentation of the results. When a study does address an exposure-response relationship, failure to find a relationship can be due to a small range of exposures, insufficient sample size or a changing exposure magnitude over time that has not been accounted for.

An inference of causality is strengthened when there is a **temporal relationship** in which the exposure comes before the cancer. This aspect is necessary to show causality; if it is not present, a causal inference is not plausible. Because the latency period for cancers can be long (years), evaluation of studies should consider whether the exposure occurred sufficiently long ago to be associated with cancer development.

An inference of causality is strengthened when the exposure is **specific** for a given cancer. This would mean that the disease endpoint being studied is only due to the cause being assessed. This issue is seldom applicable and, since NHL has other causes, specificity is not applicable to the determination of causality for glyphosate.

An inference of causality is strengthened when other lines of experimental evidence are **coherent** with a causal interpretation of the association seen in the epidemiological evidence. To evaluate coherence, information from animal carcinogenicity studies, mechanistic investigations and information on the metabolism of the chemical being studied would be considered.

An inference of causality is strengthened when there is **experimental evidence in humans** supporting a causal interpretation. Seldom is this type of information available when addressing the toxicity of chemicals. However, experiments in which an individual reduces or limits exposures and the risk of cancer is reduced would carry considerable weight in the evaluation (e.g. studies evaluating the cancer risks of people who stop cigarette smoking compared with continuing smoking have demonstrated reduced lung cancer risks). No such data are available for glyphosate.

Finally, an inference of causality is strengthened when there are other chemical agents with **analogous** structures showing similar effects in humans and/or animals and/or showing similar biological impacts in mechanistic studies. No such data are available for glyphosate.

The most logical approach to developing an inference of causality is to step through each of the aspects of causality developed **by Hill (1965)**[36] and apply them to the available data for glyphosate and for glyphosate formulations. This is done in the sections that follow.

Consistency of the Associations seen in Human Epidemiological Studies

Relevant Epidemiology Studies

In their meta-analysis, **Chang and Delzell (2016)**[38] performed a systematic literature search of all scientific literature up to June, 2015, to identify all epidemiological studies that were pertinent to evaluating an association between glyphosate and NHL. They identified 12 relevant epidemiology studies[39-50]. Their search agrees with all current reviews of glyphosate and I will use their findings from the literature up until 2015. To cover from June 2015 to the present (April 1, 2017), I used their searching algorithm and identified 117 additional published studies, none of which were new epidemiology studies. These same 12 studies will be considered for use in this evaluation. Other experts will be discussing the studies as well as their strengths and their weaknesses; I will focus on using the results of these studies in evaluating causality so I will only briefly describe each study.

Cantor et al. (1992)[39] did an in-person interview study comparing 622 white men, newly diagnosed with NHL, to 1245 population-based controls in Iowa and Minnesota. They originally identified 780 cases, of which 694 (89%) were interviewed. After pathology review, only 622 were found to have NHL, the remaining cases having leukemia or other diseases. Three different sources of controls were used, random digit dialing (76.7% response rate), Health Care Financing Administration rolls (79% response

rate) and deceased controls with eligible proxies (77% response rate). Both cases and controls were questioned regarding their use of agricultural products including Roundup® and any other glyphosate-based formulations. For deceased or incompetent controls (184) and cases (number not given), proxy interviews were done with a close relative. When cases in farmers were compared to cases in non-farmer controls, 26 cases (out of 266) and 49 controls (out of 547) had handled herbicides containing glyphosate yielding an odds ratio[1] (OR) of 1.1 (95% confidence interval 0.7-1.9). This analysis controlled for vital status, age, state, cigarette smoking status, family history of lymphopoietic cancer, high-risk occupations and high-risk exposures in a logistic analysis. The authors noted there was "minimal evidence for confounding of results for any single pesticide by exposure to pesticides belonging to other chemical families." Because the exposure is determined based on interviews in cases and controls, this study has the potential for recall bias[2]. However, the authors note that the bias could both increase or decrease the OR because of non-differential exposure misclassification[3] because of difficulties in accurate recall of past pesticide exposures for both controls and treated individuals. This study will not be included separately into the evaluation since it overlaps with **De Roos et al. (2003)**[43]

Two additional studies conducted by **Zahm et al. (1990)**[51] in Nebraska and **Hoar et al. (1986)**[52] in Kansas collected information on pesticide and herbicide use, but did not report specifically on the effects of glyphosate. **De Roos et al. (2003)**[43] pooled the data from these two studies with the data from **Cantor et al. (1992)**[39] to examine pesticide exposure to glyphosate in farming as risk factors for NHL. The three case-control studies[39, 51, 52] had slightly different designs. The design for the Minnesota study[39] is

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[1] The odds ratio (OR) is calculated as the proportion of exposed cases with disease to exposed controls divided by the proportion of non-exposed cases to non-exposed controls. For rare diseases, this value approximates the population risk ratio (PRR) which is the probability of having the disease in exposed individuals divided by the probability of having the disease in non-exposed individuals. If the PRR is 1, then there is no difference in the probability of having the disease regardless of your exposure. Values of PRR greater than 1 imply the risk is higher in the exposed population. Because the OR is an estimate of the PRR for rare diseases, it is usually accompanied by a 95% confidence interval that describes the probable range of the estimate. If the OR is greater than 1, then the exposure is associated with the disease. If the lower 95% confidence bound for the OR is greater than 1, this is typically used to say the association is statistically significant.

[2] Recall bias occurs when cases are more likely to say they are exposed to glyphosate than controls or when controls are more likely to say they are exposed to glyphosate than cases. The recall must be different for the cases than the controls for this to cause a bias; errors in recalling past exposures that happen for both cases and controls would not be recall bias.

[3] Non-differential exposure misclassification occurs when the probability of an error in determining whether an individual is exposed or not is the same for both cases and controls.

provided directly above. In Nebraska[51], the cases were identified through the Nebraska Lymphoma Study Group and area hospitals for 66 counties and included all white men and women diagnosed with NHL between July 1, 1983 and June 30, 1986. Controls were obtained by random-digit dialing, Medicare records or state mortality files depending upon age and vital status. All study participants were over age 21 and even though this study included a few women, they were excluded from the **De Roos et al. (2003)** analysis. The response rates for cases and controls were 91% and 87% respectively. In Kansas[52], cases were randomly sampled from a registry at the University of Kansas of white men, over age 21, diagnosed between 1979 and 1981. The response rates for cases and controls were 96% and 94% respectively. Controls were population-based matched on age and vital status. As for the Nebraska study, controls for live cases were obtained from Medicare records for cases 65+ and by random-digit dialing for cases <65 years; controls for deceased patients came from state mortality records. The resulting pooled case-control study had 870 cases and 2569 controls (for analyzing the relationship between glyphosate and NHL, there were only 650 cases and 1933 controls following exclusion of subjects with missing data). For any glyphosate exposure, there were 36 exposed cases and 61 exposed controls with an OR (95% confidence interval) of 2.1 (1.1-4.0) in a logistic regression analysis controlling for all other pesticides reported, age and study site. The authors also analyzed the data using a Bayesian hierarchical regression analysis yielding an OR (95% confidence interval) of 1.6 (0.9-2.8) controlling for the same parameters as the logistic regression. They also conducted an analysis of "potentially carcinogenic" pesticides which included glyphosate. When just one of these pesticides was used by subjects, the logistic regression OR was 1.6 (0.8-3.1), two to four pesticides yielded an OR of 2.7 (0.7 to 10.8) and when more than five were used, the OR was 25.9 (1.5-450.2) in the logistic regression analysis and 1.1 (0.8-1.7), 1.3 (0.7-2.3) and 2.0 (0.8-5.2) respectively for the Bayesian analysis. Removing glyphosate from the list of "potentially carcinogenic" pesticides yielded equivalent ORs of 1.2 for one pesticide, 1.2 for two to four pesticides and 1.1 for five or more pesticides. The authors note that the positive results seen in their study are not likely due to recall bias since there were few associations seen over the 47 pesticides they studied. Also, although some of the positive results could be due to chance, the use of the hierarchical regression analysis theoretically decreases the chance of false positive findings. In the Kansas study[52], suppliers for 110 subjects with farming experience were identified and provided information on the subjects' crops and pesticide purchases. In general, the suppliers reported less pesticide use than the subjects of the study with no consistent differences in agreement rates between cases and controls. The agreement between suppliers and subjects improved when pesticide use during the last 10 years was considered. This supports a reduced role of recall bias in these studies and a possible role of non-differential exposure misclassification. The reduced ORs when using the Bayesian analysis as compared to the logistic regression is not surprising because the authors used a non-informative prior rather than a less conservative prior. In addition, adjustment for 47 pesticides is also likely to reduce the significance of the observed ORs for pesticides that are associated with NHL as demonstrated by the analysis of "potentially carcinogenic" pesticides (this model is possibly over-parameterized since it

includes over 47 dependent variables for only 36 exposed cases; this can significantly reduce the ORs and increase the confidence bounds). This pooled case-control study is the strongest study with sufficient power (3.8% of subjects exposed) and will be included in the evaluation of causation.

Lee et al. (2004)[44] pooled data from **Zahm et al. (1990)**[51] and **Cantor et al. (1992)**[39] (previously described) to evaluate whether asthma acts as an effect modifier of the association between glyphosate exposure and NHL. Women were included in this analysis whereas **De Roos et al. (2003)**[43] excluded women. The final study published by Lee included 872 cases and 2336 controls of which 45 cases and 132 controls had been told by their doctors they had asthma. The OR of association between glyphosate and NHL in non-asthmatics was 1.4 (0.98-2.1) and 1.2 (0.4-3.3) in asthmatics when controlling for age, vital status and state (geographical location). This study completely overlaps with the study by **De Roos et al. (2003)**[43] with the exception of the inclusion of the few women in the study by **Zahm et al. (1990)**[51]. Since this study only looks at effect modification due to asthma, it does not contribute to the overall evaluation of causality and it will be excluded from further evaluations.

Nordstrom et al. (1998)[40] conducted a population-based case-control study of hairy cell leukemia (HCL); a subtype of B-cell NHL) in Sweden that included an evaluation of exposures to glyphosate. The study included 111 men with NHL reported to the Swedish Cancer Registry between 1987 and 1992 (with one patient from 1993 accidentally included). Controls (400 in total) were drawn from the National Population Registry matched for age and county with the cases. The response rates were 91% for cases (10 refused to participate out of the original 121) and 83% (84 controls refused to participate out of 484 selected). Almost all questionnaires were answered by the subject of the study (4 cases and 5 controls were answered by proxies). The study reported an OR for glyphosate exposure and HCL of 3.1 (0.8-12) controlling only for age. This study had very limited power for detecting an association because there were only four cases and five controls with glyphosate exposure (1.8% of the total study population). In addition, because they failed to adjust for other exposures, the potential for confounding in this study is greater than those presented previously. The authors noted that they attempted to minimize recall bias by only using living cases in the analysis. Also, even though matching was performed to identify the controls, this matching was not used in the final analysis. This study was later used in a pooled analysis of HCL and NHL[42] and will not be considered independently in the evaluation for causation but will be used in the context of the pooled analysis.

Hardell and Eriksson (1999)[41] conducted a population-based case-control study of all male patients older than 25 years diagnosed with NHL between 1987 and 1990 in the four most northern counties of Sweden. After excluding misdiagnosed cases, they included 442 cases of which 404 answered their questionnaire (most by proxy) for a response rate of 91%; 192 of these cases were deceased. For each living case, two male matched controls were chosen from the National Population Registry and matched on age and county. For each deceased case, two male controls were chosen from the National Registry for Causes of Death, matched for age and year of death. The response

rate for the controls was 84% (741 out of 884 identified). Study subjects were sent a detailed questionnaire and, in most cases, this was supplemented with a phone interview. A complete working history was obtained with questions regarding exposure to numerous chemicals to avoid a focus on pesticides and organic solvents, the focus of the study. Exposure was defined as at least one full day of exposure more than one year before diagnosis. For glyphosate exposure, the authors identified four cases and three controls with exposures and a univariate OR of 2.3 (0.4-13). A multivariate analysis of both glyphosate and phenoxy herbicides produced an OR of 5.8 (0.6-54). The study has limited power for detecting an effect because the exposure frequency is very low (0.6% exposed). This study was later used in a pooled analysis of HCL and NHL[42] and will not be considered independently in the evaluation for causation but will be used in the context of the pooled analysis.

Hardell et al. (2002)[42] conducted a pooled analysis of NHL and HCL by combining the studies of **Nordstrom et al. (1998)**[40] and **Hardell and Eriksson (1999)**[41]. This study fully overlaps with the previous two studies. The analysis controlling for age, study, county and vital status yielded an OR of 3.04 (1.08-8.52) based on eight exposed cases and eight exposed controls. A more extensive analysis additionally controlled for other pesticides and yielded a smaller OR of 1.85 (0.55-6.20). As for the study by **De Roos et al. (2003**), the analysis may be over-parameterized (more than eight dependent variables with only eight exposed cases) which could lead to a reduction in the ORs and larger confidence bounds. Even with the pooled data, **Hardell et al. (2002)** had limited power to detect an effect because the exposure frequency for cases and controls was very low (1% exposed). This study is a valid case-control study and will be used in the evaluation of causality.

In a later study, **Eriksson et al. (2008)**[46] conducted a population-based case-control study where cases were identified as NHL patients aged 18-74 years diagnosed in four major hospitals in Sweden from December 1, 1999 until April 30, 2002. In total, 995 cases were identified as matching the study parameters with 910 (91%) answering the questionnaire shortly after diagnosis. All cases were classified into subgroups with 810 B-cell, 53 T-cell, and 38 unspecified lymphomas. Controls (1,108) were randomly selected from the population registry and matched on health service, region, sex and age and interviewed in several periods during the conduct of the study; 1,016 controls responded to the questionnaire (92% response rate). Study subjects were sent a detailed questionnaire and, in many cases, a phone interview followed. Exposure was defined as at least one full day of exposure more than one year before diagnosis. The univariate analysis, adjusting for age, sex and year of diagnosis (cases) or enrollment (control) yielded an OR of 2.02 (1.10-3.71) based on 29 exposed cases and 18 exposed controls. When cases and controls were divided into those with ≤10 days per year exposure and those with >10 days per year exposure, the ORs were 1.69 (0.70-4.07) and 2.36 (1.04-5.37) respectively. When diagnoses were grouped into various subtypes of NHL, the results did not change dramatically except for small lymphocytic lymphoma and chronic lymphocytic lymphoma which showed an increased OR of 3.35 (1.42-7.89). A multivariate analysis of glyphosate controlling for other agents with statistically

increased odds ratios and/or odds ratios greater than 1.5 yielded an OR of 1.51 (0.77-2.94). In a similar analysis to the multivariate analysis, latency periods of one to ten years showed an OR of 1.11 (0.24-5.08) and >10 years had an OR of 2.26 (1.16-4.40). This study was much larger than the previous Swedish studies (2.3% exposed) and, although there may have been confounding from other pesticides, this was addressed in the multivariate analysis and the latency analysis. This study is a valid case-control study and will be used in the evaluation of causality.

McDuffie et al. (2001)[50] recruited incidence cases of NHL in men 19 years or older from six Canadian provinces with a first diagnosis between September 1, 1991 and December 31, 1994. Each provincial Cancer Registry or, in the case of Quebec, hospital, had a target number of cases and ended recruitment when the case number was reached. Controls were men 19 years or older selected at random from provincial health insurance records, computerized telephone listings or voter registration lists, depending upon the province. Cases and controls were sent questionnaires with surrogates ineligible to answer the questionnaires for deceased cases or controls. Each subject who reported 10 hours per year or more of pesticide exposure and a random sample of 15% who reported less exposure were interviewed by telephone to obtain details on pesticide use. A pilot study was conducted to obtain an improved version of the telephone interview questionnaire used by **Hoar et al. (1986)**[52] and **Zahm et al. (1990)**[51] that would provide accurate pesticide exposure assessment in the form of a screening questionnaire and a telephone interview questionnaire. This was followed by a validation study (27 farmers) where the final questionnaires used to screen and include potential cases and controls were administered and the answers regarding pesticide usage showed excellent concordance with purchases through their local agrochemical supplier. The screening questionnaire was returned by 517 cases of NHL (67.1% response rate) and 1506 controls (48% response rate). Following analysis of the screening questionnaire, the telephone interview was administered to 179 cases and 456 controls to obtain more detailed exposure information. The OR for glyphosate exposure and NHL was 1.26 (0.87-1.80) stratified by age group and province of residence and the OR was 1.20 (0.83-1.74) when the analysis also controlled for significant medical variables (51 exposed cases and 133 exposed controls). An exposure-response evaluation was performed where the OR for exposure between zero to two days per year was 1.0 (0.63-1.57) and for greater than two days per year was 2.12 (1.20-3.73) with the latter group having 23 exposed cases and 36 exposed controls. This study had excellent sample size and power (8.1% of subjects exposed), but a low response rate to the screening questionnaire. Also, by adjusting for significant medical variables, this study ruled out many confounders but did not adjust for other pesticide exposures. The effort to validate the recall of pesticide usage for farmers supports a lack of recall bias in the study. This study is a valid case-control study and will be used in the evaluation of causality.

Hohenadel et al. (2011)[48] re-analyzed the data of **McDuffie et al. (2001)**[50] to specifically investigate the impact of exposure to multiple pesticides on NHL. Four cases of NHL were excluded from this evaluation following a pathology review. They reported associations with the use of glyphosate with and without malathion but not with

glyphosate overall. The OR for glyphosate (ever used) without malathion (ever used) was 0.92 (0.54-1.55) and the OR for glyphosate (ever used) with malathion (ever used) was 2.1 (1.31-3.37). **Chang and Delzell (2016)**[38] combined the ORs from the glyphosate only analysis with the glyphosate and malathion analyses using random-effects meta-analysis to get a combined OR for glyphosate of 1.4 (0.62-3.15). This study was specifically targeted to interactions of various pesticides and does not substantively contribute to an evaluation of glyphosate. Since it is a refined analysis of **McDuffie et al. (2001)**[50], it will be included in the evaluation of causation only in the context of the combined analysis provided by **Chang and Delzell (2016).**

Orsi et al. (2009)[47] conducted a hospital-based case-control study of men and women diagnosed with lymphoid neoplasms in five hospitals in France between 2000 and 2004 who were aged 20-75 years (the abstract gives the age range as 18-75 years). All diagnoses were cytologically or histologically confirmed. The evaluation only included men and questionnaires/interviews were completed by 491 cases (95.7% response rate) which included 244 cases with NHL. Controls were patients in the same hospital (mostly orthopedic or rheumatological patients) with no prior history of lymphoid neoplasms and excluding patients admitted to the hospital for cancer or a disease directly related to occupation, smoking or alcohol abuse. The controls were matched to cases by hospital and age. Of the 501 candidate controls, 456 participated (91% response). Exposure was evaluated differently for subjects who had non-occupational exposures from those who had occupational exposures. For both, the subjects had to fill out a questionnaire/interview on occupations and home gardening pesticide exposures. For those who had worked professionally as farmers or gardeners for at least 6 months, a specific agricultural occupational questionnaire/interview was administered and exposure was determined on the basis of this extra data. The OR for occupational use of glyphosate and NHL was 1.0 (0.5-2.2) with 12 exposed cases and 24 exposed controls stratified by age and center category. A further analysis was done by individual subtypes of NHL with an OR of 1.0 (0.3-2.7) for diffuse large cell lymphoma, 1.4 (0.4-5.2) for follicular lymphoma, 0.4 (0.1-1.8) for chronic lymphocytic leukemia (CLL) and 1.8 (0.3-9.3) for HCL. No separate analysis of non-occupational use of glyphosate was provided, nor does it seem specific data on glyphosate usage was ascertained for subjects who were not professional farmers or gardeners. This could lead to non-differential misclassification of exposure which could reduce the ORs of the study. Barring this, the sample size was sufficient to detect an effect (5.3% with occupational exposure) and this study will be included in the evaluation of causality.

Cocco et al. (2013)[49] evaluated data from a multi-center case-control study of lymphoid neoplasms in six European countries from 1998 to 2004. Cases included only adult patients diagnosed with lymphoma during the study period drawn from participating centers. Controls were either selected by sampling from the general population on sex, age group, and residence area (Germany, Italy), or from hospital controls matched to the patient excluding patients with cancer, infectious diseases, and immunodeficiency diseases (Czech Republic, France, Ireland, Spain). The study included 2348 lymphoma cases (88% participation) and 2462 controls (81% response rate in hospital-based controls and 52% in population-based controls). Exposures were derived using an

occupational exposure matrix developed by industrial hygienists and occupational experts from the research centers. Only 35 individuals (cases and controls not broken out) in the study were exposed to carbamates (glyphosate was grouped with the carbamates). No results were provided for NHL and the only OR provided for glyphosate was for B-cell lymphoma where the OR was 3.1 (0.6-17.1) based on four exposed cases and two exposed controls. No information was provided on the total number of cases for each type of lymphoma evaluated. This study has very limited power to evaluate an association between NHL and glyphosate and provides only information on B-cell lymphomas with very few exposed cases and controls. As has been done by most researchers evaluating these data, this study will receive very little weight in the evaluation of causality.

De Roos et al. (2005)[45] reported results on the association of glyphosate and cancer incidence from the Agricultural Health Study (AHS), a prospective cohort study in Iowa and North Carolina, which included 57,311 private and commercial applicators who were licensed to apply restricted-use pesticides at the time of enrollment. Recruitment occurred between 1993 and 1997 and cohort members were matched to cancer registry files to identify cases and the National Death Index (1999) to ascertain vital status. Incident cancers were identified from the date on enrollment until 31 December, 2001, with the average follow-up time being 6.7 years. Comprehensive use data was obtained by self-administered questionnaire for 22 pesticides, ever/never use for 28 additional pesticides, and general information on work practices. Applicators were given a second self-administered questionnaire on occupational exposures and lifestyle factors. They used three exposure metrics in their analyses: a) ever personally mixed or applied pesticides containing glyphosate; b) cumulative exposure days of use of glyphosate (years of use times days per year); and c) intensity weighted cumulative exposure days (years of use times days per year times intensity of use). Persons whose first primary tumor occurred before the time of enrollment (1074) were excluded from the analysis as were those who were lost to follow-up (298), did not provide age information (7) or information on glyphosate use (1678) leaving 54,315 subjects for inclusion. There were 92 cohort members with a diagnosis of NHL during the study period of which 77.2% had ever used glyphosate resulting in a rate ratio[4] (RR) of 1.2 (0.7-1.9) when controlling for age and an RR of 1.1 (0.7-1.9) when controlling for age, lifestyle factors, demographics and five other pesticides for which cumulative-exposure-day variables were most highly associated with glyphosate cumulative-exposure-days (2,4-D, alachlor, atrazine, metalochlor, and trifluralin) or, for chemicals with only ever/never exposure information that were most highly associated with glyphosate ever/never use (benomyl, maneb, paraquat, carbaryl and diazinon). When cumulative exposure days in exposed individuals are divided into tertiles and RRs examined using the lowest exposed tertile as

[4] The rate ratio (RR) is estimated as the incidence in the exposed population divided by the incidence in the unexposed population. Incidence is calculated as the number of events in a fixed period of time divided by the person years at risk. Unlike the OR, the RR does not require the assumption of a rare disease to serve as a good estimate of the population risk ratio (PRR).

the reference group, the RRs drop with values of 0.7 (0.4-1.4) and 0.9 (0.5-1.6) for tertiles 2 and 3 respectively controlling for demographic and lifestyle factors and other pesticides (30,699 subjects). When intensity-weighted exposure days are examined again using exposed tertile 1 as the reference group, the RRs drop with values of 0.6 (0.3-1.1) and 0.8 (0.5-1.4) for tertiles 2 and 3 intensity-weighted exposure days respectively controlling for demographic and lifestyle factors and other pesticides (30,699 subjects). Analyses are not shown for the evaluation of the exposed tertiles against never exposed because the authors felt that never exposed and exposed subjects differed in terms of socio-economic factors and other exposures like smoking[45].

This is a typical cohort study, but has some limitations in terms of its interpretation. The majority (75.5%) of subjects in the cohort reported having ever personally mixed or applied products containing glyphosate and was composed primarily of male, middle-aged, private applicators. For glyphosate, reliability of the answers by subjects on the use of glyphosate between the first and second questionnaire were evaluated in the AHS[53]: 82% agreement for whether they had ever mixed or applied glyphosate, 53% agreement on years mixed or applied, and 62% agreement on days per year mixed or applied and 62% agreement on decade first applied. They saw no differences in over versus under reporting between the two questionnaires suggesting this could lead to non-differential exposure bias and reduce the RRs in this study. Another weakness, noted by the authors, is that the small number of incident cases during follow-up period hindered precise effect estimates. Also, the high frequency of exposure to many pesticides (e.g. 73.8% were exposed to 2,4-D) means subjects unexposed to glyphosate were likely to be exposed to other agents that may also induce NHL, reducing the RRs. Also, as noted by the EPA's FIFRA Science Advisory Panel (SAP)[54] in their review of the EPA's issue paper on the carcinogenicity of glyphosate and as noted in a critique[55] of the European Food Safety Agency's risk assessment for glyphosate, the follow-up time in this cohort study may not be long enough to produce a sufficient sample size for evaluation of the association between NHL and glyphosate. Like other studies, this study has few exposed cases and controls, but the authors adjust their analysis for many other pesticides which could reduce ORs and increase confidence bounds limiting the ability of the study to show positive results. This study could also suffer from a survival bias because pesticide applicators were recruited as case participants after their exposure had begun and those with a cancer prior to enrollment were excluded.

This study will be included in the evaluation of causality.

Consistency of Associations

Hill (1965)[36] defines consistency as the answer "yes" to the question "Has it repeatedly been observed by different persons, in different places, circumstances and times?" For these studies, the answer is indeed yes.

If the population relative risk (PRR) for an association of glyphosate with NHL were equal to 1 (no effect), then one would expect very few statistically significant results in multiple studies and that about half of the studies would have ORs or RRs below one

and half above one. As noted by both the **IARC Monograph 112 (2015)**[56] and by **Chang and Delzell (2016)**[38], when comparing studies, the most reasonable comparison is to use the most-fully-adjusted risk estimates. I will mostly limit my comments to these most-fully-adjusted risk estimates.

Consistency of the associations across several epidemiology studies is not simply a matter of seeing how many were statistically significant and how many were not but must also address the consistency of the direction of the responses. Figure 1 shows a forest plot of all ORs and RRs from the epidemiology studies discussed previously. Each horizontal line in the forest plot shows the mean estimate of the OR/RR as a black square and the 95% confidence interval around this estimate as whiskers extending left and right from the black square.

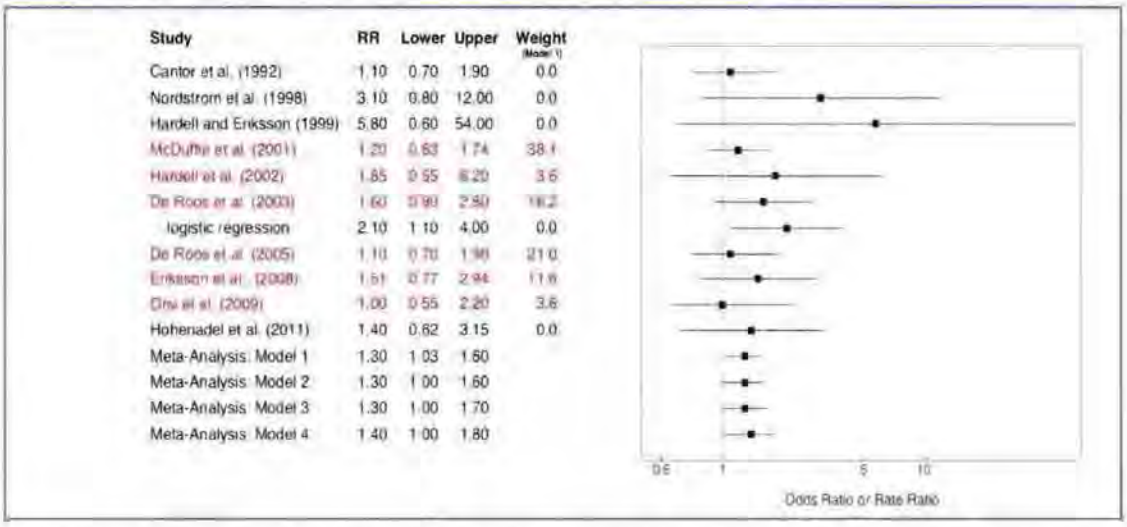
The first obvious conclusion to be drawn from Figure 1 is that all of the mean OR/RR estimates (black squares) are consistently ≥1. This implies that all of the studies are pointing in the same direction toward a positive effect. In their meta-analyses, **Schinasi and Leon (2014)**[57], **IARC (2015)**[56] and **Chang and Delzell (2016)**[38] all identified 6 papers (highlighted in red in Figure 1) as being the most reliable for evaluation of the ability for glyphosate to induce NHL in people: **McDuffie et al. (2001)**[50], **Hardell et al. (2002)**[42], **De Roos et al. (2003)**[43] and **(2005)**[45], **Eriksson et al. (2008)**[46] and **Orsi et al. (2009)**[47]. I will refer to these papers as the six core epidemiology studies. As noted above, if the true underlying risk ratio was 1 (no effect), you would expect about half of the findings to be below 1 and half to be equal to 1 or greater. Using only the results from the 6 core studies, you can see that all are ≥1; the probability of this happening is $(0.5)^6$ or 0.016, strongly suggesting the studies do not agree with an underlying PRR=1 and that they consistently support a positive effect.

A second way in which consistency can be evaluated is to combine the individual studies using meta-analysis to obtain a combined analysis using both the ORs and the RR (CRR) and test for heterogeneity in the studies. The meta-analysis done by **Chang and Delzell (2016)** includes the same analysis as that done by the **IARC (2015)** and is an improvement over **Schinasi and Leon (2014),** so I will focus my comments on using the **Chang and Delzell (2016)** meta-analysis. **Chang and Delzell (2016)** did four separate meta-analyses on the glyphosate epidemiology studies using two different methods (random-effects and fixed-effects models). In their first analysis (model 1)[5], they combined the most-fully-adjusted risk estimates from the six core studies to yield a CRR of 1.27 (1.01-1.59) for both random-effects and fixed-effects models supporting an association between NHL and glyphosate exposure in these studies. In a second analysis (model 2), they replace the results of the Bayesian analysis in **De Roos et al. (2003)** with the results of the logistic regression analysis and get the same CRR of 1.30 (1.03-1.64) for both random-effects and fixed-effects models. In a third analysis (model 3), they replace from model 1 the **McDuffie et al. (2001)** results in with a combined meta-

[5] **Chang and Delzell (2016)** provided only one significant digit to the right of the decimal point in their confidence bounds; the EPA SAP (2017) re-calculated models 1-4 of **Chang and Delzell (2016)** to provide two significant digits – these are presented here.

analytic result they derived from analyses by **Hohenadel et al. (2011)** (this study reanalyzed the same data as **McDuffie et al. (2001)**, splitting results between asthmatics and non-asthmatics) resulting in a CRR of 1.32 (1.00-1.73) for both random-effects and fixed-effects models. Finally, in a fourth analysis (model 4), they use model 3 but replaced the Bayesian analysis in **De Roos et al. (2003)** with the logistic regression analysis yielding a CRR of 1.37 (1.04-1.82) for both random-effects and fixed-effects models. In essence, none of the different meta-analyses rejected the notion of a combined, statistically significant positive effect.

Figure 1: Odds Ratios and Rate Ratios from the most-fully-adjusted risk estimates from selected epidemiology studies and from the meta-analyses of **Chang and Delzell (2016)**[38]. "RR" refers to the OR or RR from the study, "Lower" refers to the 95% lower bound, "Upper" to the 95% upper bound and "Weight" refers to the weight applied to that specific study in Model 1 of the meta-analysis (Table 3 in Chang and Delzell). For **De Roos et al. (2003)**, the first row is for the Bayesian model analysis and the second row, labelled "logistic regression" is from the logistic model analysis.



As stated above, another way to evaluate consistency in the epidemiological data would be to evaluate the heterogeneity in the studies. Heterogeneity may be due to differences in participants, outcomes, exposure metrics, methods for questioning study subjects, sex of the subjects, etc. **Chang and Delzell (2016)** formally tested for heterogeneity of the responses from the six core studies using Cochran's Q statistic and the I^2 statistic[58]. For models 1 to 4, the p-values from Cochran's Q test are 0.84, 0.59, 0.85, and 0.63 respectively (typically you reject the concept of homogenous studies in favor of heterogeneous studies if $p<0.10$). The I^2 statistic for all four models are 0.0% (values for I^2 can range from 0-100% with concern for heterogeneity above 50%). The fact that the fixed-effects models and random-effects models gave the same results also supports a lack of heterogeneity in the data. There is no indication of heterogeneity in these six core studies. Lack of heterogeneity supports the interpretation of the meta-analyses as showing a positive association and strong consistency of the findings across the six core studies.

Chang and Delzell (2016) also evaluated the association between subtypes of NHL and glyphosate exposure where possible. For B-cell lymphomas, they combined the results of **Eriksson et al. (2008)**[46] with those of **Cocco et al. (2013)**[49] and saw a CRR (random-effects and fixed-effects) of 2.0 (1.1-3.6) with an I^2 of 0 and a Cochran's Q test p-value of 0.58. For diffuse large B-cell lymphomas, they combined the results of **Eriksson et al. (2008)**[46] with those of **Orsi et al. (2009)**[47] and saw a CRR (random-effects and fixed-effects) of 1.1 (0.5-2.3) with an I^2 of 0 and a Cochran's Q test p-value of 0.79. For combined chronic lymphocytic leukemia and small lymphocytic lymphoma, they combined the results of **Eriksson et al. (2008)**[46] with those of **Orsi et al. (2009)**[47] and saw a CRR using the random-effects model of 1.3 (0.2-10) and for the fixed effects model 1.9 (0.9-4.0) with an I^2 of 83.7% and a Cochran's Q test p-value of 0.01. For follicular lymphomas, they combined the results of **Eriksson et al. (2008)**[46] with those of **Orsi et al. (2009)**[47] and saw a CRR (random-effects and fixed-effects) of 1.7 (0.7-3.9) with an I^2 of 0 and a Cochran's Q test p-value of 0.73. And finally, for HCL, they combined the results of **Nordstrom et al. (1998)**[40] with those of **Orsi et al. (2009)**[47] and saw a CRR (random-effects and fixed-effects) of 2.5 (0.9-7.3) with an I^2 of 0 and a Cochran's Q test p-value of 0.63. These subtype analyses are based upon small numbers of cases and only two studies making them unreliable, when considered individually, to address the question of consistency in the data. However, when they are combined with the results for the meta-analyses of the core studies of NHL, these studies add support to the conclusion that these data are consistent.

Chang and Delzell (2016) also performed a sensitivity analysis by only doing meta-analyses on studies with similar characteristics. Using only the five case-control studies, the CRR was 1.3 (1.0-1.7). Breaking them into the type of control used, there were four studies using population controls with a CRR of 1.4 (1.0-1.8). There were four studies with males only with a CRR of 1.3 (1.0-1.7) and two studies with males and females with a CRR of 1.2 (0.8-1.8). Three studies were done in North America with a CRR of 1.2 (1.0-1.6), three in Europe with a CRR of 1.3 (0.8-2.1); two of the three studies were in Sweden with a CRR of 1.6 (0.9-2.8). All of the resulting meta CRRs were the same for the fixed-effects model and the random-effects model. This sensitivity analysis shows that the results do not differ significantly from the main CRR for the six core studies combined adding support to the findings being consistent across the different studies.

In case-control studies, selection bias arises when the reasons cases and controls choose to participate in the study could lead to systematic biases that might result in a positive or negative finding independent of the exposure being studied. For example, if cases with exposure are more likely to participate than controls with exposure, the result would be higher OR values; however, this difference has to be differential and not simply a difference in participation rates. It is possible that in a few of these studies, the method by which controls were selected could contribute to selection bias that might lead to increased ORs. However, given the diverse types of cases and controls used in the five core case-control studies, this is unlikely to explain the consistent findings seen from these studies. It is also possible that the lack of complete data on cases versus controls could result in selection bias if the reasons for not completing the questionnaire/interview are different between cases and controls and relates to

exposure. There is no indication of this type of selection bias in these reports, and this is unlikely to explain the consistency seen in these data.

Exposure misclassification can lead to increases or decreases in the OR or RR values seen in both case-control and cohort studies. For example, in case-control studies, if cases are more likely to say they were exposed to glyphosate than controls, this would inflate the OR values; this is one type of recall bias. This type of bias is less likely in cohort studies. In all six of the core studies, this issue was discussed by the authors. In every case, they concluded there was bound to be some exposure misclassification, but that it was most likely non-differential, meaning that the misclassification was random; this would likely reduce the OR/RRs seen in the studies rather than increase them.

Confounding occurs when there is an exposure or some other factor that is tightly associated with both glyphosate exposure and NHL diagnosis that, if controlled for, could explain the results. The most likely source of confounding in these studies would be exposures to other pesticides. Four[42, 43, 45, 46] of the six core studies controlled for exposure to other pesticides and saw basically the same findings as the other two studies. Another concern for confounding would be if the cases had immune deficiencies that could be linked to NHL; in all of the case-control studies, such cases were excluded. Finally, other agricultural exposures (e.g. animals, other chemicals, infectious agents) could be correlated with glyphosate exposure and may be linked to NHL; none of the studies controlled for these factors. However, not all exposed cases were farmers; if confounding via other agricultural exposures is occurring, it is not possible to determine the magnitude or direction of such an effect from these data.

In conclusion, we have six core epidemiology studies done on two different continents by four different research groups using different designs, questionnaires and study populations that are highly consistent with no obvious bias or confounding that would explain the results. **There is a consistency of associations across the six core studies.**

Strength of the Association seen in Human Epidemiological Studies

To explain strength of association, **Hill (1965)** gives the classic example of John Snow and the cholera epidemic of 1855 where the risk ratio of dying if you drank water from the Southwark and Vauxhall Company (polluted by sewage) compared to drinking from the Lambeth Company water (sewage free) was 14. Yet, for the six core studies, the OR/RR ranges from 1.0 to 1.85 for the most-fully-adjusted risk estimates and to 2.1 if you include the fully adjusted risk estimate from De Roos et al. (2003)[45] using logistic regression. These are moderate OR/RR estimates making it conceivable they are individually due to either chance or bias. Thus, with the exception of the logistic regression analysis in **De Roos et al. (2003)**[45], none of the core studies demonstrate large, precise risks as envisioned by **Hill (2016)**[36]. However, **Hill (1965)** was not expressing himself in statistical terms where the significance of an association is dependent upon the precision of the observations. If the statistical variation around an OR/RR estimate is large relative to the estimate itself, the estimate is not very precise

and generally would not be statistically significant. The result from the study by **Hardell and Eriksson (1999)** shown in Figure 1 is an example of an estimate with very large statistical variation. On the other hand, a very small (in value), precise OR or RR estimate could be statistically significant and prove important in deciding causation. The meta-analyses shown in Figure 1 all demonstrate estimates of OR/RR that are significantly different from 1 rejecting the concept that the overall association is due to chance. The statistically significant estimate of the OR/RR for B-cell lymphomas in the meta-analysis support this finding as well.

In summary, we have six core epidemiology studies that all show approximately the same, modest increase in OR/RR that, when combined, demonstrate a significant strength of association. **There is a strong association across the six core studies**

Biological Plausibility

The range of data one can use to determine biological plausibility is quite diverse and can be exceptionally complicated. For simplicity, it can be divided into the types of assays that can be used in this evaluation: animal cancer bioassays, toxicokinetic studies, studies from accidental exposures in humans, and studies of specific biological mechanisms in animals or cells derived from humans or animals. Animal cancer bioassays are intended to test whether glyphosate can cause cancers in mammals, thus supporting the concept that the chemical could cause cancer in humans. Toxicokinetic studies provide insight into the degree to which glyphosate is absorbed by humans, distributed to various organs in the body, what happens to the chemical once it is in the body (metabolism), and, finally, how it is eliminated from the body. Studies from accidental exposures in humans can provide some information on the effects of glyphosate through changes in the chemistry and cellular structure of human blood. Studies of biological mechanisms are generally addressing what effects the chemical may have on human and animal cells under controlled, laboratory conditions. Some of the studies in this section were done with technical grade (virtually pure) glyphosate and some with the glyphosate formulations that humans encounter in occupational and environmental settings. I will summarize the literature in each of these areas and offer an opinion to their support of biological plausibility of NHL in humans.

Animal Cancer Bioassays

Typical animal cancer bioassays will expose animals (generally rats or mice) to a chemical for a substantial proportion of the animal's life (generally 2 years) then kill the animal and examine its organs and tissues for tumors. There are guidelines on how to conduct and analyze these studies. Typically, chemical registrants conduct cancer bioassays for pesticide approval pursuant to guidelines developed under the guidance of the Organization for Economic Cooperation and Development (OECD[59]). Other groups[30, 33, 34] provide guidance on how to analyze these studies based upon methodology papers from the published literature. These studies are conducted in a way that controls for everything in the animal's environment (e.g., food type, water quality, how often the animals are handled) leaving only the exposure to explain

differences in tumor formation between control and exposed animals. Even then, non-cancer endpoints can also be modified by the chemical and these may have an impact on tumor rates in the animals (e.g., survival, death from some other toxic effect of the chemical); these must be accounted for when reaching conclusions from the study.

Studies generally use four groups of animals, one group receiving no exposure (control) and the remaining three groups are test animals, with each group receiving different dose exposures to the chemical[60]. Doses generally above human experience are used in animal carcinogenicity studies because only relatively small numbers of animals are being used to evaluate risk for a large human population and because even the best known human carcinogens do not cause cancer in large fractions (say 20%) of the human population. The basic underlying premise of this design consideration is that, as the dose increases, so does the risk of getting a tumor. By exposing animals to the highest dose possible, you increase the ability of the study to identify a risk if one is present. However, one must be careful not to use a dose that is so high it will cause cancers by processes that would never work at lower doses. To avoid this, studies are designed around a maximum tolerated dose (MTD) or limit dose. This dose is generally determined based upon a subchronic study (90 days) in the same animals and is usually the maximum dose that can be tolerated by the animals without any signs of significant toxicity in the exposed animals (e.g., weight loss, tissue damage). The OECD and EPA provide guidelines[33, 59] on how to choose this top dose. These guidelines are in general agreement with the scientific literature[60].

The guidelines also address the methods by which the data should be analyzed. For example, the EPA guidelines[61] state that:

"A trend test such as the Cochran-Armitage test (Snedecor and Cochran, 1967) asks whether the results in all dose groups together increase as dose increases. A pairwise comparison test such as the Fisher exact test (Fisher, 1950) asks whether an incidence in one dose group is increased over that of the control group. By convention, for both tests a statistically significant comparison is one for which p is less than 0.05 that the increased incidence is due to chance. Significance in either kind of test is sufficient to reject the hypothesis that chance accounts for the result."

In fact, most guidelines and peer-reviewed publications come to the same conclusion[30, 59, 60, 62] on what tests to use, as did EPA's FIFRA Scientific Advisory Panel (SAP) in their review of the EPA's issue paper of the carcinogenicity of glyphosate[54]. The US National Toxicology Program (NTP) uses both a trend test[3, 4, 63] and Fisher's exact test for analyzing carcinogenicity data. Unless otherwise noted in this document, all p-values presented in this section on animal cancer studies were recalculated on my computer and are the exact one-sided p-values for the Fisher test (p_{Fisher}) and/or the Cochran-Armitage linear trend test (p_{Trend}) where appropriate. Exact one-sided p-values for the trend test were calculated using a permutation algorithm with 100,000 permutations[6].

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[6] The p-value resulting from a permutation test of this type is subject to statistical error. In this case, if the exact p-value is 0.05, the test with 100,000 permutations should be no further than 0.0004 from the true value; about 1%.

To avoid doing large numbers of tests and over-analyzing the data, my comments will generally rely upon the use of the trend test with the results from Fisher's exact test serving as a descriptive discussion of the findings. This is in agreement with SAP comments[54] and is generally accepted in the evaluation of animal cancer studies.

Even with the high doses used in these studies, it is sometimes necessary to use "historical controls" to evaluate a given response. Historical controls are generally the historical collection of tumor responses from untreated control groups from studies in the same laboratory within two to three years of the study being evaluated[30, 34, 59, 64, 65]. Evaluation of the data using the historical controls should be done rigorously to correctly evaluate the responses seen in a given study. Where a valid historical control dataset was available, I used the mean tumor response in the controls to calculate the probability of observing the trend seen in the study or a more significant trend if the true probability of response is the historical control average; this is labeled p_{Hist}. In all cases, the guidelines and literature support the use of the control in the current study as the most appropriate control group to use unless there is a specific need to address historical responses. Many guidelines[30, 33, 34, 66] suggest historical controls be used for evaluating rare tumors and findings in assays that appear to be unusual. It is explicitly noted that significant increases in tumors over what is seen in the concurrent control should not be rejected simply because the tumors are in the range of the historical controls[30]. Nor is it recommended to reject significant increases in tumor responses because the control response is on the low end of the historical range. Animals are randomly assigned to control and exposure groups and any low response in controls is likely to also reflect similar response patterns in treated animals. This is in agreement with SAP comments[54] on the EPA issue paper on glyphosate[61] and with all guidelines for analyzing animal carcinogenicity data.

There are 13 animal carcinogenicity studies in rats[67-79] and eight in mice[80-87]. Only two studies[70, 76] appear in the peer-reviewed literature; the remaining studies are partially available through several sources. For three of the rat studies[69, 73, 77] and two mouse studies[82, 85], technical reports from the performing laboratory are available from documents provided by the registrant. For the remaining unpublished studies, data was obtained from the EPA review of glyphosate[61], the European Food Safety Authority review of glyphosate[88, 89] and supplemental material from a review of the carcinogenicity of glyphosate by a panel of scientists on behalf of Monsanto[90].

Many additional endpoints, other than cancer incidence and related toxicities, were evaluated in these studies; I will only provide comments on the tumor incidence data and related data where relevant to the cancer findings.

It is unusual to have multiple carcinogenicity studies in the same experimental animal model arising from different laboratories. Methods for the combined analysis of multiple animal cancer bioassays are not available in the scientific literature. However, pooled analyses, as conducted in epidemiology[91, 92] are applicable for combining animal carcinogenicity studies. The basic concept is to pool all data from the same sex/species/strain into one study and analyze it appropriately. The basic steps are: 1) select the studies to be pooled; 2) merge the data for analysis; 3) estimate study specific

effects; 4) estimate pooled effects; 5) explain the differences between the pooled effects and the individual study effects; 6) do a sensitivity analysis if possible. These steps will be used to analyze pooled data from animal carcinogenicity studies where pooling is done by sex, species, strain and duration of exposure to limit heterogeneity across pooled studies. In their recommendations to the EPA regarding EPA's issue paper on the carcinogenicity of glyphosate[54], the FIFRA Science Advisory panel strongly supported the use of a pooled analysis to address the question of consistency citing my comments to the EPA[93].

Rat Studies

Reyna and Gordon (1974)[75] exposed Albino rats (probably Sprague-Dawley) to ammonium salt of glyphosate (13.85% purity) in a two-year chronic feeding study. Only EPA[61] reported on this study and provided no details other than to report there were approximately 70 animals per group and there was insufficient reporting on the histopathology findings. Insufficient detail is available on this study.

This study is inadequate for use in deciding on causality.

Burnett et al. (1979)[69] exposed male and female albino rats to an aqueous monosodium salt solution of glyphosate by oral intubation (purity not given). There were 90 animals per group and doses were 0, 3, 10 and 30 mg/kg/day for 24 months. EPA[61] reported that no histopathological alterations were observed; no additional information was available on this study. This study had severely reduced sensitivity to observe any cancer findings because the highest dose used in this study is very low compared to the MTDs in the other rat studies. This study does not contribute to the evaluation of cancer causation in laboratory animals and will be excluded from any further discussion.

Lankas et al. (1981)[73] exposed groups of 50 male and 50 female Sprague-Dawley rats to glyphosate (98.7% purity) in feed (see Table 1 for doses) for 26 months. This study is not in concordance with OECD guidelines (they were not available at the time of this study), but as noted by EFSA[88], it was in general accordance with the 1981 OECD guidelines. Information on this study was available from EPA[61], EFSA[88], Greim et al.[90], the original study report from Bio/dynamics Inc.[94] and memos from Monsanto to EPA provided by Monsanto.

There were no survival differences in this study and there was no indication that the highest dose used exceeded the maximum-tolerated dose.

Table 1 shows the statistically significant trend in testicular interstitial cell tumors that was observed (p_{Trend}=0.011). Historical controls were provided in the study report for five studies with response rates of 4/116, 5/75, 4/113, 6/113 and 5/118 for a mean response of 4.5% (24/535). Comparing this historical control mean to the observed response yields p_{Hist}=0.006, showing that this result is significant, even when comparing it to the historical control dataset. **Lankas et al. (1981)** argued that the tumor rates at sacrifice were not statistically significant from control suggesting this finding is not related to glyphosate. However, by reducing the numbers of animals to only those at

terminal sacrifice, the power to find an effect was significantly reduced. Also, if the tumor increases the animal's chances of dying, then some animals with tumors will die early, which could bias results only seen at terminal sacrifice. This type of analysis is simply never done; it appears to have been developed for this case to dismiss the effects seen in the study. **Lankas et al. (1981)** also suggested the control response was low compared to the historical rates, but the concurrent control is always the best control group to use unless it is clearly flawed[33, 34, 59]; in this case, there was no apparent problem with the controls because the probability of seeing 0/50 if the true background response is 4.5% is about 10% and this control group is not significantly different than the historical controls. **EFSA**[88] noted rates for interstitial cell hyperplasia (a potential precursor for the interstitial cell tumors) and saw no dose-response trend (Table 1). However, these very low rates would suggest that the tumors arising in the 10 animals that did get interstitial cell tumors are independent of a mechanism involving interstitial cell hyperplasia. The tumor response for interstitial cell tumors was not monotonic (tumor rates increasing as dose increases), but was still within statistical variation. The EPA SAP agrees, concluding that "requiring visual confirmation of a monotonic trend in scatter plots of data ... is known to be a poor way of assessing trend"[54].

An increase in Thyroid C-cell carcinomas (Table 1) was observed in female rats (p_{Trend}=0.003) but combining adenomas and carcinomas was not significant (p_{Trend}=0.096). Independent pathologists brought in by Monsanto argued these tumors were not treatment related. The authors also mentioned that the incidence of lymphocytic hyperplasia in the thymus and lymph nodes were slightly elevated above controls, but no tumor counts or statistical evaluations were provided. The authors provided historical control data for both carcinomas and carcinomas combined with adenomas from nine control groups with mean responses of 4/453=0.9% for carcinomas and 46/453=10.2% for the combined tumors. The significance of both results was unchanged using the historical control data.

This study also had a statistically significant increase in pancreatic islet cell tumors in the lowest dose (p_{Fisher}=0.028) in males (Table 1), but not any of the other doses; the trend test was not significant (p_{Trend}=0.32).

The highest dose used in this study in Sprague-Dawley rats is far below the MTD. Even though EFSA[88] noted that this study was in general accordance with the 1981 OECD guidelines, they dismissed it for not meeting current guidelines due to the low-doses used. EPA[61] also excluded this study from consideration. However, the study saw an increase in testicular tumors in males and Thyroid C-cell carcinomas in females that should be carefully evaluated in determining causality. Also, this is the study with the longest exposure (24 months) and provides unique information to the overall evaluation.

Table 1: Tumors of interest in male and female Sprague-Dawley rats the 26-month feeding study of **Lankas (1981)**[73]

Tumor	Sex	Doses (mg/kg/day)				p-values
	Male	0	3.05	10.30	31.49	
	Female	0	3.37	11.22	34.02	
Testicular interstitial cell tumors	Male	0/50	3/50	1/50	6/50**	P_{Trend}=0.011 P_{Hist}=0.006
Interstitial cell hyperplasia	Male	1/50	1/50	1/50	0/50	NS
Thyroid C-cell Carcinomas	Female	1/47	0/49	2/50	6/47	P_{Trend}=0.003 P_{Hist}=<0.001
Thyroid C-cell Adenomas and Carcinomas	Female	6/47	3/49	8/50	9/47	P_{Trend}=0.096 P_{Hist}=0.072
Pancreas Islet Cell Tumors	Male	0/50	5/50*	2/50	3/50	P_{Trend}=0.315

*- p_{Fisher}<0.05, **- p_{Fisher}<0.01

In conclusion, this study shows positive result for testes interstitial cell tumors and hepatocellular adenomas in male Sprague-Dawley rats and a positive response for thyroid c-cell carcinomas in female Sprague-Dawley rats and will be included in the overall evaluation of causation.

Stout and Ruecker (1990)[77] exposed groups of 50 male and 50 female Sprague-Dawley rats to glyphosate (98.7% purity) in feed (see Table 2 for doses) for 24 months. This study was done under OECD guidelines.

There were no survival differences in this study and there was no indication that the highest dose used exceeded the maximum-tolerated dose.

Pancreatic islet cell tumors were increased in all dose groups relative to the controls in male rats and statistically significant for the lowest (p_{Fisher}=0.015) and highest (p_{Fisher}=0.032) dose groups (Table 2). However, these rates include the 10 animals that were sacrificed at one year. Due to the short duration of exposure, the rats terminated at one year were likely not at risk of developing this tumor; it is very unusual to include these animals in the final tumor counts (EPA[61] also excluded these animals). In the pathology tables for this study, there were no tumors in any of the 10 animals at the interim sacrifice. Removing these 10 animals does not alter the p-values for trend or Fisher's exact test. Historical control data for this tumor in this laboratory was reported as 23/432 or 5.3%[95] and a trend comparison against this control rate was not significant (p_{hist}=0.15). The lack of a trend is driven by the up and down nature of the response. Assuming the historical rate of 5.3% is correct, the chances of seeing eight or more tumors in 47 animals is 0.003. Similarly, for the mid- and high-doses, this probability is 0.124 and 0.014, respectively. Females did not show an increase in this tumor. The authors provided a table with the combined results for pancreatic islet-cell adenomas and carcinomas from this study with the tumor counts from the **Lankas et al. (1981)**[73] study arguing the results do not show a dose-related increase; the trend test of the pooled results are p_{Trend}=0.08 and p_{Hist}=0.020 (with historical control mean response of

5.3%) indicating a slight trend. Animals studied for 26 months versus 24 months can have very different responses to the same chemical and very different control incidence.

In male rats, there was a statistically significant trend (p_{Trend}=0.019) after removal of interim-sacrificed animals for hepatocellular adenomas but a marginal[7] increase for adenomas and carcinomas combined (p_{Trend}= 0.06, Table 2) and not in females. Live carcinomas are generally also provided in a separate analysis, but these data were not provided by the authors (the data would suggest the hepatocellular carcinomas would have a negative trend).

There was also a marginal increase in thyroid C-cell adenomas in the female rats (p_{Trend}=0.057) and adenomas and carcinomas combined (p_{Trend}=0.06) regardless of whether interim sacrificed animals are included (Table 2). In males, the trend for adenomas was p_{Trend}=0.094 and for adenomas and carcinomas was p_{Trend} =0.103. Adenomas were seen in male rats at the interim sacrifice demonstrating that male rats at the interim sacrifice were at risk for this tumor. If these animals are added back into the analysis, the trend test in males has p_{Trend}=0.07 for adenomas and p_{Trend}=0.079 for adenomas and carcinomas combined.

Several other tumors demonstrating significant findings in other studies of Sprague-Dawley rats are included in Table 2 and do not show significant effects.

In conclusion, the finding of an increased incidence of pancreatic islet-cell tumors in this study cannot easily be ruled out as a chance finding. Findings of significant increases in liver adenomas in male rats with no increases in carcinomas could be due to chance. The findings of marginally significant increases in thyroid c-cell tumors in males and females should be compared with other studies. This study will be included in the overall evaluation of causation.

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[7] In statistics, it is common to refer to p-values in the range of 0.10>p-value>0.05 as marginal when the target p-value is ≤0.05; this is done to avoid missing trends in data reflected by almost significant findings

Table 2: Tumors of interest in male and female Sprague-Dawley rats from the 24-month feeding study of Stout and Ruecker (1990)[77]

Tumor	Sex	Doses (mg/kg/day)				p-values
	Male	0	89	362	940	
	Female	0	113	457	1183	
Pancreas Islet Cell Tumors (with interim sacrifice)	Male	1/58	8/57*	5/60	7/59*	P_{Trend}=0.15 P_{Hist}=0.14
Pancreas Islet Cell Tumors (without interim sacrifice)	Male	1/48	8/47*	5/50	7/49*	P_{Trend}=0.15 P_{Hist}=0.15
Hepatocellular adenomas (without interim sacrifice)	Male	3/50	2/50	3/50	8/50	P_{Trend}=0.019,
Hepatocellular Adenomas and Carcinomas (without interim sacrifice)	Male	6/50	4/50	4/50	10/50	P_{Trend}=0.060
Thyroid C-Cell Adenomas (with interim sacrifice)	Female	2/60	2/60	6/60	6/60	P_{Trend}=0.056
Thyroid C-Cell Adenomas (without interim sacrifice)	Female	2/50	2/50	6/50	6/50	P_{Trend}=0.057
Thyroid C-Cell Adenomas and Carcinomas (with interim sacrifice)	Female	2/60	2/60	7/60	6/60	P_{Trend}=0.060
Thyroid C-Cell Adenomas and Carcinomas (without interim sacrifice)	Female	2/50	2/50	7/50	6/50	P_{Trend}=0.059
Thyroid C-Cell Adenomas (with interim sacrifice)	Male	2/60	4/60	8/60*	7/60	P_{Trend}=0.070
Thyroid C-Cell Adenomas (without interim sacrifice)	Male	0/50	4/50	8/50*	5/50*	P_{Trend}=0.094
Thyroid C-Cell Adenomas and Carcinomas (with interim sacrifice)	Male	2/60	6/60	8/60*	8/60*	P_{Trend}=0.079
Thyroid C-Cell Adenomas and Carcinomas (without interim sacrifice)	Male	0/50	6/50*	8/50*	6/50*	P_{Trend}=0.103
Testis Interstitial Cell Tumors	Male	2/50	0/50	3/50	2/50	P_{Trend}=0.300
Kidney Adenomas	Males	0/50	2/50	0/50	0/50	P_{Trend}=0.814
Thyroid Follicular Adenoma/Carcinoma	Males	2/50	1/48	3/48	3/50	P_{Trend}=0.230

*- $p_{Fisher}<0.05$, **- $p_{Fisher}<0.01$

Atkinson et al. (1993)[67] conducted a combined chronic toxicity/carcinogenicity study of glyphosate (98.9% pure). They used 50 Sprague-Dawley rats in each group for both sexes with dietary exposures given in Table 3. An additional 35 rats/sex/dose were included for interim sacrifices.

There were no survival differences in this study and there was no indication that the highest dose used exceeded the maximum-tolerated dose.

Table 3: Tumors of interest in male and female Sprague-Dawley rats from the 24-month feeding study of Atkinson et al. (1993)[67]

Tumor	Sex	Doses (mg/kg/day)					p-values
	Male	0	11	112	320	1147	
	Female	0	12	109	347	1134	
Thyroid Follicular Adenomas and Carcinomas	Male	0/50	0/21	0/17	2/21	2/49	P_{Trend}=0.036
Thyroid Follicular Adenomas and Carcinomas (adding terminal sacrifice animals to denominator)	Male	0/50	0/50	0/50	2/50	2/49	P_{Trend}=0.038
Thyroid C-cell Adenomas and Carcinomas	Female	8/50	1/27	1/29	1/29	7/49	P_{Trend}=0.081
Thyroid C-cell Adenomas and Carcinomas	Male	9/50	1/21	1/17	2/21	9/49	P_{Trend}=0.177
Testes Interstitial Cell Tumors	Male	3/50	1/25	0/19	0/21	2/50	P_{Trend}=0.354
Kidney Adenomas	Males	1/50	0/50	0/50	0/50	0/50	p_{Trend}=1
Hepatocellular Adenomas	Males	2/50	1/50	1/50	2/50	3/50	P_{Trend}=0.159

*- p_{Fisher}<0.05, **- p_{Fisher}<0.01

The authors reported no significant effects, as do **EPA**[61] and **EFSA**[88]. The study did not do detailed histopathological examination on all animals in all groups for every tumor type, but did examine all control and high dose animals, all animals that died before study termination and animals showing macroscopic tumors at study termination; liver, kidney and lungs were examined for all animals. This severely weakens the study for addressing dose-response trends. However, in reviewing the pathology tables provided in **Greim et al. (2015)**[90], thyroid follicular adenomas and carcinomas were found to be statistically significant (p_{Trend}=0.037) by the trend test. If the three middle exposure groups had seen no other tumors and the denominators were the entire 50 animals on study, the significance of the trend analysis would not change.

Without examination of the animals free of gross tumors at terminal sacrifice, the findings from this study will be given less weight in the overall evaluation of causation.

Brammer (2001)[68] conducted a two-year carcinogenicity study in Wistar rats in which groups of 52 animals were exposed to glyphosate (97.6% pure) at doses provided in Table 4 . An additional 12 animals were sacrificed at one-year.

A significant positive trend in survival was noted by the EPA (p=0.03), however this trend was not accomplished using a Kaplan-Meir test[96] (the appropriate test), but simply a test relating to the percent surviving to terminal sacrifice. There was no indication that the highest dose used exceeded the maximum-tolerated dose.

EPA[61], but not **EFSA**[88], noted there was a statistically significant trend of combined hepatocellular adenomas in male rats with the highest dose also being statistically significant from the control. Trend analysis gives p_{Trend}=0.009 and the Fisher's exact test comparison of high dose to control is p_{Fisher}=0.028. EPA dismissed this finding as potentially due to a slight difference in the number of animals at the terminal sacrifice in this study versus controls. However, no formal statistical evaluation of survival is provided and it cannot be assumed from these numbers that survival was significantly impacted in these animals. **Greim et al. (2015)**[90] used slightly different numbers for this tumor because three animals (one in the control group, one in the low-dose group and one in the mid-dose group) in the interim sacrifice group died before their sacrifice time and, from the pathology tables provided in their paper, these could not be separated from others. These numbers have been included in Table 4, but it does not change the significance of the findings. **Greim et al. (2015)**[90] dismissed these findings, partly because of the same survival argument used by the EPA and partly because they had a historical control dataset where the range of historical response was from 0-11.5%; they did not provide the mean response or the individual tumor responses for these historical controls. As mentioned earlier, dismissing results because they are in the range of the historical controls is an unacceptable method for using historical controls to evaluate a study, and in this case, there is no reason to question the concurrent controls.

Table 4: Tumors of interest in male and female Wistar rats from the 24-month feeding study of Brammer (2001)[68]

Tumor	Sex	Doses (mg/kg/day)				p-values
	Male	0	121	361	1214	
	Female	0	145	437	1498	
Hepatocellular Adenoma	Male	0/52	2/52	0/52	5/52*	P_{Trend}=0.009
Hepatocellular Adenoma (from Greim et al., 2015[90])	Male	0/53	2/53	0/53	5/52*	P_{Trend}=0.009 P_{Hist}=0.006
Mammary Gland Adenomas and Adenocarcinomas	Female	3/51	2/51	0/51	2/51	P_{Trend}=0.574

*- p_{Fisher}<0.05, **- p_{Fisher}<0.01

I obtained historical control data from 16 control groups in Wistar rats from Charles River Laboratories for the years 2003 to 2011[97]. Although these are outside of the optimal time range for the animals used in the **Brammer (2001)** study, they can serve as an illustration of why using a range can be misleading. There were 52 liver adenomas seen in 1217 control animals for a mean response of 4.27% with a range of 0% to 17.5% (individual study findings of 6/100, 0/60, 1/60,1/50,1/80, 14/112, 1/65, 0/60, 21/120, 0/50, 1/50, 2/60, 0/50, 1/100, 1/150, 2/50; 13 studies with ≤2% response). Assuming

the underlying probability of having a tumor in controls is 4.27%, p_{Hist}=0.006 (Table 4). Thus, even though the responses seen in **Brammer (2001)** are in the range of the historical controls, the trend is highly significant when historical controls are used appropriately. **Greim et al. (2015)** also mentioned findings of increased toxicity at the high dose for which they provided numbers for only hepatocyte fat vacuolation and hepatitis; none of these findings were statistically significant by any test.

In conclusion, this study shows a positive result for hepatocellular adenomas in male Wistar rats and will be included in the overall evaluation of causation.

Pavkov and Wyand (1987)[74] exposed Sprague-Dawley rats to glyphosate trimesium salt (sulfosate, 56.2% pure) in feed for two years. Eighty animals/sex were tested in the control, low-dose and mid-dose groups, and 90/sex were tested in the high dose group. Doses of 0, 4.2, 21.2 and 41.8 mg/kg/day were used in males and 0, 5.4, 27, and 55.7 mg/kg/day in females. This study showed no significant findings according to EPA[61]. No details were given beyond that simple statement and no others reported on this study. The doses in this study are far below the MTD so this study would have reduced sensitivity to detect an effect if one existed. This study also used a different chemical than the other Sprague-Dawley rat studies and is not comparable on that basis.

This study is not acceptable for use in the evaluation of causality due to the lack of details about the study.

Suresh, (1996)[78] exposed Wistar rats to glyphosate (96.8% pure) in feed for two years. Fifty animals/sex were tested in four exposure groups shown in Table 5.

There were no survival differences in this study and there was no indication that the highest dose used exceeded the maximum-tolerated dose.

EPA[61] concluded there were no tumors increased due to glyphosate exposure in this study and **EFSA**[88] concluded that, "[n]one of the significant microscopic changes, increased and decreased incidences (in liver, spleen, lymph nodes, adrenals, thymus, gonads, uterus, mammary gland) observed have shown dose relationship, hence appeared to be incidental and not related to the treatment with the test compound." (page 491). **Greim et al. (2015)**[90] provided data on hepatocellular adenomas and carcinomas in both sexes but none of these showed significant trends or pairwise tests (Table 5). However, there was another study [insert author name for reference] with a strong significant trend in hepatocellular adenomas in Wistar rats[68] so these are also included in Table 5 for comparison. No other tumors were mentioned by any other group and an examination of the grouped pathology tables provided by **Greim et al. (2015)** show an increase in mammary gland adenomas at the mid-dose (p_{Fisher}=0.017) but no significant trend. However, there was another study [insert author name for reference] with a strong significant trend in mammary gland adenomas and adenocarcinomas combined in Wistar rats[79] so these are also included in Table 5 for comparison. Like the **Atkinson et al. (1993)**[67], **Suresh (1996)** did not do full pathology on all of the animals in the interim exposure groups making interpretation for mammary gland adenomas problematic.

This study will be included in the overall evaluation of causation.

Table 5: Tumors of interest in male and female Wistar rats from the 24-month feeding study of Suresh(1996)[78]

Tumor	Sex	Doses (mg/kg/day)				p-values
	Male	0	6.3	59.4	595.2	
	Female	0	8.6	88.5	886	
Mammary Gland Adenoma and Carcinoma	Female	5/40	3/28	8/33*	2/48	P_{Trend}=0.875
Hepatocellular Adenoma	Male	24/50	22/50	10/50	21/50	P_{Trend}=0.371

*- p_{Fisher}<0.05, **- p_{Fisher}<0.01

Enemoto (1997)[71] exposed Sprague-Dawley rats to glyphosate (95.7% pure) in feed for two years. Fifty animals/sex were tested in four exposure groups (see Table 6). In addition, 10 animals per exposure group were exposed for 1 year and another 10 for 18 months at which point they were sacrificed and examined. These interim sacrifice animals (1 year and 18 months) are included in the analysis if tumors were seen in these groups.

There were no survival differences in this study and there was no indication that the highest dose exceeded the maximum-tolerated dose.

Table 6: Tumors of interest in male and female Sprague-Dawley rats from the 24-month feeding study of Enemoto (1997)[71]

Tumor	Sex	Doses (mg/kg/day)				p-values
	Male	0	104	354	1127	
	Female	0	115	393	1247	
Mammary Gland Adenoma	Female	23/50	27/50	24/50	30/50	P_{Trend}=0.194
Kidney Adenoma	Male	0/50	0/50	0/50	4/50	P_{Trend}=0.004
Thyroid C-cell Adenomas/Carcinomas	Female	4/60	7/60	8/60	4/60	P_{Trend}=0.682
Thyroid C-cell Adenomas/Carcinomas	Male	8/70	10/70	6/70	7/70	P_{Trend}=0.685
Thyroid Follicular-cell Adenomas/Carcinomas	Male	4/70	2/70	1/70	0/70	P_{Trend}=0.989
Testes Interstitial Cell Tumors	Male	3/49	2/50	0/50	2/50	P_{Trend}=0.588
Hepatocellular Adenomas	Male	1/60	0/60	2/60	1/60	P_{Trend}=0.371

*- p_{Fisher}<0.05, **- p_{Fisher}<0.01

EPA and EFSA both found no significant changes in tumors in any group. **Greim et al. (2015)** again provide tables for a number of tumors, none of which show significant effects except for the incidence of kidney adenomas in male rats (p_{Trend}=0.004, Table 6). Examining the pathology tables provided in **Greim et al. (2015)** reveals no additional tumors showing an increase in tumor incidence with dose. A different study[73] in

Sprague-Dawley rats demonstrated a strong significant trend in mammary gland adenomas, thyroid C-cell carcinomas and testicular interstitial cell tumors so these are also included in Table 6 for comparison.

This study showed a significant increase in kidney adenomas and will be included in the overall evaluation of causation.

Wood et al. (2009)[79] exposed Wistar rats to glyphosate (94.7% to 97.6% pure) in feed for two years. Fifty-one animals/sex were tested in four exposure groups at doses shown in Table 7. **EFSA**[88] found no dose-related tumor increases while **EPA**[61] noted an increase in mammary gland adenomas and adenocarcinomas combined with p_{Trend}=0.062 for adenomas, p_{Trend}=0.046 for adenocarcinomas and p_{Trend}=0.008 for the combined tumors (Table 7). EPA concluded there was no progression from adenoma to adenocarcinoma and argued the increase was not glyphosate related. This conclusion is contradicted by the fact that 6 animals in control and the lower dose groups got carcinomas with no adenomas in any of the animals in these groups. It seems likely that, in this case, mammary gland adenocarcinomas can arise without the presence of any adenomas. **Greim et al (2015)**[90] also noted an increase in skin keratoacanthoma in males (p_{Trend}=0.033). Review of the pathology tables identified no other tumors with increased tumor rates as a function of dose. There was another study with a strong significant trend in hepatocellular adenomas in Wistar rats[68] so this tumor is also included in Table 7 for comparison.

This study showed an increase in mammary tumors in females and skin keratoacanthomas in males and will be used in the evaluation of causality.

Table 7: Tumors of interest in male and female Wistar rats from the 24-month feeding study of Wood et al. (2009)[79]

Tumor	Sex	Doses (mg/kg/day)				p-values
	Male	0	85.5	285.2	1077.4	
	Female	0	104.5	348.6	1381.9	
Mammary Gland Adenomas	Female	0/51	0/51	0/51	2/51	P_{Trend}=0.064
Mammary Gland Adenocarcinomas	Female	2/51	3/51	1/51	6/51	P_{Trend}=0.046
Mammary Gland Adenomas and Adenocarcinomas	Female	2/51	3/51	1/51	8/51*	P_{Trend}=0.008
Skin Keratocanthoma	Male	2/51	3/51	0/51	6/51	P_{Trend}=0.034
Hepatocellular Adenoma	Male	0/51	2/51	1/51	1/51	P_{Trend}=0.416

*- p_{Fisher}<0.05, **- p_{Fisher}<0.01

Excel (1997)[72] exposed Sprague-Dawley rats to glyphosate (purity not given) in feed for two years. Fifty-one animals/sex were tested in four exposure groups at doses of 0, 150, 780 and 1290 mg/kg/day in males and 0, 210, 1060 and 1740 mg/kg/day in females. **EPA**[61], **EFSA**[88] and **Greim et al. (2015)**[90] had concerns with the quality of this study,

the characterization of the chemical being used and with tumor rates in this strain of animals being too low. The Supplemental Material from **Greim et al. (2015)** on this study shows no significant increase in any tumor and virtually all animals having no tumors in controls and treated animals.

This study is inadequate for use in deciding on causality for the same reasons given by the **EPA**, **EFSA** and **Greim et al. (2015)**.

Chruscielska, K. (2000)[70] exposed Wistar rats to glyphosate as a 13.8% solution (purity not given) in drinking water for two years. According to **Greim et al. (2015)**[90], this appears to be the glyphosate formulation Perzocyd. Eighty-five animals/sex were tested in four exposure groups. The authors listed the doses as control, 300 mg/L, 900 mg/L and 2700 mg/L in drinking water. **Greim et al. (2015)**[90] estimated the intake of glyphosate to be 0, 1.9, 5.7 and 17 mg/kg/day for females and 0, 2.2, 6.5, and 19 mg/kg/day in males. There was a slight increase in malignant adenomas of the pituitary gland and an opposite decrease in pituitary adenomas suggesting no effect or potentially a promotional effect in which adenomas are promoted to carcinomas by glyphosate. No other increased tumor responses were reported in the manuscript. Because of the low exposures, this study is an inadequate challenge to the animals (the highest dose is far below the MTD). The reporting of this study is very limited and it the overall quality of the work cannot be evaluated.

This study is inadequate for use in deciding on causality.

Seralini, G. E., et al. (2014)[76] exposed Sprague-Dawley rats to the glyphosate formulation Roundup in drinking water for two years as part of a broader experiment on Roundup-Ready Corn. Ten animals/sex were tested in four exposure groups at doses of 0, 0.00005, 400 and 22500 mg/L in females. The authors reported an increase in the incidence of mammary gland tumors (mainly fibroadenomas and adenocarcinomas) in female rats with incidences of 5/10 for control and 9/10, 10/10, 9/10 (p_{Fisher}=0.016) in the low-, mid- and high-doses groups respectively. It is difficult to assess the quality of this study due to limited reporting on the histopathological descriptions of the tumors and the very small sample size.

This study will not be used in the evaluation of causality.

Summary - Rats

Table 8 summarizes the significance for all tumors of interest in rats.

Brammer (2001)[68] saw a significant increase in hepatocellular adenomas in male Wistar rats with increasing dose (p_{Trend}=0.009, Table 4). The other two acceptable studies in Wistar rats (**Wood et al. (2009)**[79] and **Suresh (1996)**[78] did not see significant increases (Tables 5 and 7). On the basis of statistical significance, these studies are inconsistent. To reject these findings based upon only 1/3 being positive is the same as rejecting a coin as being fair if, in three flips of the coin, the result is one head and two tails; it simply is not possible and there is a better way to address these findings. Given different doses and different sample sizes, we need to formally test for consistency in these studies. **Suresh (1996)** saw 48% response for hepatocellular adenomas in controls

whereas the other two studies saw no tumors in the control animals. Thus, although all three studies are in Wistar rats, **Suresh (1996)** has a significantly different control response from the other two. **Suresh (1996)** did not give a substrain for the Wistar rats used, but **Brammer (2001)** and **Wood et al. (2009)** used different substrains. All three studies used different diets and were conducted in different facilities. Thus, there is no obvious explanation for the dramatically different rates in **Suresh (1996).** It is known that the same strain of rats from different laboratories can have markedly different control tumor responses. Because they have similar control response, **Brammer (2001)** and **Wood et al. (2009)** can be pooled into a single study to ask the question "Does the significant trend for **Brammer (2001)** disappear when it is pooled with the negative study of **Wood et al. (2009)**?" The analysis of the pooled studies yields p_{Trend}=0.014 supporting the conclusion that glyphosate causes hepatocellular adenomas in Wistar rats with similar background responses.

Table 8: Summary of significance tests for 5 tumors from 7 studies in Rats

Study	Strain	Neoplasm						
		Hepatocellular Adenomas (males)	Mammary Gland Tumors (females)	Thyroid C-Cell Tumors (females)	Thyroid C-Cell Tumors (males)	Thyroid Follicular Cell Tumors (males)	Testis Interstitial Cell Tumors (male)	Kidney Adenomas (males)
Brammer (2001)[68]	Wistar	+++[1]	-					
Wood (2009)[79]		-	+++					
Suresh (1996)[78]		-	-					
Pooled Wistar Rats[2]		++	++					
Lankas (1981)[73]	Sprague Dawley	NA[3]		+	NA[3]	NA[3]	++	NA[3]
Enemoto (1997)[71]		-		-	-	-	-	+++
Atkinson et al. (1993)[67]		-		+	-	++	-	-
Stout and Ruecker (1990)		++		-	+	-	-	-
Pooled Sprague-Dawley Rats		++		-	+	-	-	++

[1]entries are p_{Trend}/p_{Hist} with values: – p>0.1, + 0.1≥p>0.05, ++ 0.05≥p>0.01, +++ p≤0.01; [2]pooling results from **Stout and Ruecker (1990)**, **Brammer (2001)** and **Wood (2009)** only; [3]NA=not available

Wood et al. (2009)[79] saw a significant increase in mammary gland adenomas and adenocarcinomas (p_{Trend}=0.008, Table 7) in females that was not seen in the other two studies (Tables 4 and 6). The background rates in these studies differ only slightly and a pooled analysis of all three studies yields p_{Trend}=0.303, suggesting that combining the data eliminates the dose-response trend seen in **Wood et al. (2009)**. However, if the Wistar rats used in **Suresh (1996)** differed in their response for hepatocellular adenomas, they may differ for this tumor as well. Combining only **Wood et al. (2009)** with **Brammer (2001)** results in p_{Trend}=0.038. Given the mixed results from the pooling for this tumor I conclude there is only limited support for the notion that glyphosate can cause mammary gland adenomas and adenocarcinomas in Wistar rats.

In Sprague-Dawley rats, there were four studies that were acceptable for inclusion in the evaluation of causality with one[73] yielding strong positive responses for thyroid C-cell carcinomas in females and testicular interstitial tumors and hepatocellular adenomas in males and another[71] yielding a strong result for kidney adenomas in males. **Lankas (1981)**[73] saw a significant increase in thyroid C-cell carcinomas in female rats exposed to glyphosate (p_{Trend}=0.003, Table 1) and a marginal increase in C-cell adenomas and carcinomas combined (p_{Trend}=0.096, p_{hist}=0.072, Table 1; two of the other three studies also saw marginal results for this thyroid C-cell adenomas and carcinomas in females (Tables 2 and 3). A pooled analysis using all four studies yields p_{Trend}=0.287. This pooled analysis does not support the results seen in **Lankas (1981)**. However, the **Lankas (1981)** study was for 26 months and the other three were for 24 months; the C-cell carcinomas could be a result of the longer exposure period even though the dose is substantially lower in this study compared to the other two. From these data, I conclude that the evidence is weak that glyphosate causes thyroid C-cell tumors in female Sprague-Dawley rats.

Thyroid C-cell adenomas and carcinomas combined, in males, show marginally significant dose-response trends in **Stout and Ruecker (1990**, Table 2) but not in the remaining two studies for which there is data; data is unavailable for this tumor for **Lankas (1981)**. Pooling all three studies yields a marginally significant trend of p_{Trend}=0.053. From these data, I conclude that the evidence is weak that glyphosate causes thyroid C-cell tumors in male Sprague-Dawley rats.

Thyroid follicular-cell adenomas and carcinomas combined, in males, show a significant dose-response trend in **Atkinson et al. (1993**, Table 3) but not in the remaining two studies for which there is data; data is unavailable for this tumor for **Lankas (1981)**. Pooling all three studies yields no significant trend with p_{Trend}=0.387. From these data, I conclude that there is no evidence that glyphosate causes thyroid follicular-cell tumors in male Sprague-Dawley rats.

Hepatocellular adenomas, in males, show a significant dose-response trend in **Stout and Ruecker (1990**, Table 2) but not in the remaining two studies for which there is data; data is unavailable for this tumor for **Lankas (1981)**. Pooling all three studies yields a significant trend with p_{Trend}=0.024. From these data, I conclude that there evidence that glyphosate causes thyroid follicular-cell tumors in male Sprague-Dawley rats.

Another significant trend seen in Sprague-Dawley rats is the finding of testes interstitial cell tumors from **Lankas (1981)**[73] (P_{Trend}=0.011, Table 1); the other three studies were negative for this tumor (Tables 2, 3 and 6). Combining the other three studies with that of **Lankas (1981)** for testes interstitial tumors results in a p-value for trend that is clearly non-significant (p_{Trend}=0.516). However, as noted above, the **Lankas (1981)** study was for 26 months and the other two were for 24 months; the tumors could be a result of the longer exposure period even though the dose is substantially lower in this study compared to **Stout and Ruecker (1990)**, **Atkinson et al.(1993)** and **Enemoto (1997)**.

The final tumor in Sprague-Dawley rats showing a strong significant trend is kidney adenomas in males from the study by **Enemoto (1997)**[71] (P_{Trend}=0.004, Table 6). The kidney tumor data is not available for the study by **Lankas (1981)**[73] and is not significant in the studies by **Atkinson et al. (1993)**[98] (Table 3) and **Stout and Ruecker (1990)**[77] (Table 2). Pooling the **Enemoto (1997)** study with that of **Stout and Ruecker (1990)** and **Atkinson et al. (1993)** yields p_{Trend}=0.031; thus, the association between glyphosate and kidney adenomas in male Sprague-Dawley rats is supported by these data, even with the difficulty associated with interpreting the results in the low- and mid-doses in the **Atkinson et al. (1993)** study. There is evidence to support an increase in kidney tumors in male Sprague-Dawley rats exposed to glyphosate.

In summary, there is evidence that glyphosate causes hepatocellular adenomas in male Wistar rats, mammary gland adenomas and adenocarcinomas in female Wistar rats and kidney adenomas and hepatocellular adenomas in male Sprague-Dawley rats. There is weak evidence glyphosate causes thyroid C-cell adenomas and carcinomas in male Sprague-Dawley rats.

Mouse Studies

Reyna and Gordon (1974)[85] exposed Swiss White mice to glyphosate (>97% purity) in feed for 16 months in males and 18 months in females. Fifty animals/group/sex were tested in three exposure groups; control, 17 mg/kg and 50 mg/kg. Only 10 animals per group were examined for histopathological changes.

There was no impact on survival of administration of glyphosate and no indication that the high dose exceeded the MTD.

No significant increases were seen in any tumor from this study. However, given the small sample size for histopathological evaluation and the low doses used for this study, this study is inadequate.

This study will not be used in the evaluation of causality.

Knezevich and Hogan, (1983)[82] exposed CD-1 mice to glyphosate (99.8% pure) in feed for two years. Fifty animals/group/sex were tested in four exposure groups (see Table 9).

There were no survival differences in this study and there was no indication that the highest dose used exceeded the MTD.

Table 9: Tumors of interest in male and female CD-1 mice from the 24-month feeding study of **Knezevich and Hogan (1983)**[82]

Tumor	Sex	Doses (mg/kg/day)				p-values
	Male	0	157	814	4841	
	Female	0	190	955	5874	
Kidney Adenoma[1] (original pathology)	Male	0/49	0/49	1/50	3/50	P_{Trend}=0.019 P_{Hist}=0.005
Kidney Adenoma (EPA pathology)	Male	1/49	0/49	0/50	1/50	P_{Trend}=0.436 P_{Hist}=0.121
Kidney Carcinoma[2] (EPA pathology)u	Male	0/49	0/49	1/50	2/50	P_{Trend}=0.063 P_{Hist}=0.002
Kidney Adenoma and Carcinoma Combined[3] (EPA pathology)	Male	1/49	0/49	1/50	3/50	P_{Trend}=0.065 P_{Hist}=0.011
Malignant Lymphoma[4]	Male	2/49	5/49	4/50	2/50	P_{Trend}=0.738 P_{Hist}=0.767
Hemangiosarcoma[5]	Male	0/50	0/49	1/50	0/50	P_{Trend}=0.499 P_{Hist}=0.591
Bilateral Chronic Interstitial Nephritis	Male	5/49	1/49	7/50	11/50	P_{Trend}=0.009
Hemangiosarcoma[6]	Female	Data not available				
Lung Adenocarcinoma[7]	Male	4/48	3/50	2/50	1/50	P_{Trend}=0.907 P_{Hist}=0.899

*- p_{Fisher}<0.05, **- p_{Fisher}<0.01, [1]historical rate=0.27%, [2]historical rate=0.15%, [3]historical rate=0.44%, [4]historical rate=6.2%, [5]historical rate=2.5%, [6]Historical Control Rate=1.7%, [7]Historical rate=9.2%

EPA[99] found a significant increase in kidney tubular cell adenomas in male mice based upon the original pathology done from the study and this analysis is shown in Table 9 (p_{Trend}=0.019). Kidney tubular cell adenomas are very rare tumors in CD-1 mice so it is important to compare these results with the historical controls. No historical controls were available from the laboratory that conducted **Knezevich and Hogan, (1983)**so IARC, EPA and EFSA all used historical control databases from published studies in the literature[100-102]. These studies have virtually identical rates for the important tumors seen in CD-1 mice; I will use the study by **Giknis and Clifford (2000)**[101] since it best covers the range of studies we have for CD-1 mice. For studies of approximately two years, the mean historical tumor response in controls is 0.27%. Applying this control response rate to the kidney adenomas yields p_{Hist}=0.005, strengthening the significance of the evaluation against the concurrent control. EPA originally used a similar analysis and reached the same conclusions. However, in 1985, the registrant had a group of pathologists review the kidney slides. Using additional kidney sections from this study,

the pathologists identified an additional adenoma in the control animals and changed the classification for three adenomas to carcinomas (Table 9). With these changes, the adenomas no longer have a significant trend (P_{Trend}=0.436, P_{Hist}=0.121) but carcinomas have a marginally significant trend against concurrent controls and a clearly significant trend using historical controls (p_{Trend}=0.063, p_{Hist}=0.002, historical control rate of 0.15%). These historical control rates may not apply to this analysis because the reevaluation of the kidney tumors considered additional sections and no information is available on how additional sections affect historical control rates in this strain of mice; differences have been seen in other settings[103]. The incidence of combined carcinomas and adenomas has the same marginal significance against the concurrent control and significance against the historical controls (p_{Trend}=0.065, p_{Hist}=0.011, historical control rate of 0.44%). Other CD-1 mice studies have seen increases in malignant lymphomas and hemangiosarcomas. Evaluations of those tumors for this study yields results that are not significant; for malignant lymphoma, p_{Trend}=0.74, p_{Hist}=0.77, with the historical control rate equal 6.2% and for hemangiosarcomas p_{Trend}=0.500.59, p_{Hist}=0.77, with the historical control rate equal to 2.5%. No other tumors were found in this study.

The **EPA**[61] has produced many different arguments to dismiss the findings of renal tumors from this study. One argument is that the pathology working group requested by the EPA in 1986 concluded these lesions were not glyphosate related because *"1) renal tubular cell tumors are spontaneous lesions for which there is a paucity of historical control data for this mouse stock; 2) there was no statistical significance in a pairwise comparison of treated groups with the concurrent controls and there was no evidence of a statistically significant linear trend; 3) multiple renal tumors were not found in any animal; and 4) compound-related nephrotoxic lesions, including pre-neoplastic changes, were not present in male mice in this study."* Reason number one no longer exists as there are two very good historical control databases for CD-1 mice[100, 101]. The second reason, while technically correct, is not supportable since the Agency's own guidelines for evaluating carcinogenicity studies state that *"Significance in either kind of test* [trend or pair-wise] *is sufficient to reject the hypothesis that chance accounts for the result."* The third reason is also weak since one would not expect (nor require) multiple tumors to appear when dealing with a rare tumor. For the fourth point, EPA provides data on the rate of bilateral chronic interstitial nephritis in the study which it considers to show no statistically significant results although the trend test is highly significant (p_{Trend}=0.009, Table 9). EPA then states, without reference, that *"chronic interstitial nephritis is not considered to be a precursor lesion for tubular neoplasms"*. I could find no published research to either support or refute this statement. However, chronic interstitial nephritis is an inflammation of the interstitial tissue surrounding the glomeruli and tubules in the kidney. Inflammation is well known to play an important role in kidney cancer[104] and many other cancers so this argument also fails to support rejection of these findings.

In summary, this study shows a positive result for kidney tumors in male CD-1 mice and will be included in the overall evaluation of causation.

Atkinson, et al., (1993)[80] exposed CD-1 mice to glyphosate (>97% purity) in feed for two years. Fifty animals/group/sex were tested in four exposure groups (see Table 10).

There was no impact on survival of administration of glyphosate and no indication that the high dose exceeded the MTD.

Table 10: Tumors of interest in male and female CD-1 mice from the 24-month feeding study of **Atkinson et al. (1993)**[80]

Tumor	Sex	Doses (mg/kg/day)				p-values
	Male	0	98	297	988	
	Female	0	102	298	1000	
Kidney Adenoma and Carcinoma Combined[1]	Male	2/50	2/50	0/50	0/50	P_{Trend}=0.980 P_{Hist}=1
Malignant Lymphoma[2]	Male	4/50	2/50	1/50	6/50	P_{Trend}=0.092 P_{Hist}=0.085
Hemangiosarcoma[3]	Male	0/50	0/50	0/50	4/50	P_{Trend}=0.004 P_{Hist}=0.001
Hemangiosarcoma[4]	Female	0/50	2/50	0/50	1/50	P_{Trend}=0.439 P_{Hist}=0.376
Lung Adenocarcinoma[5]	Male	10/50	7/50	8/50	9/50	P_{Trend}=0.455 P_{Hist}=0.4491

*- p_{Fisher}<0.05, **- p_{Fisher}<0.01, [1]historical rate=0.44%, [2]historical rate=6.2%, [3]historical rate=2.5%, [4]Historical Control Rate=1.7%, [5]Historical rate=9.2%

Hemangiosarcomas were the only tumors showing a significant trend in this study (P_{Trend}=0.004, P_{Hist}=0.001, Table 10). Also shown in Table 10 are the results for malignant lymphomas and kidney tumors; there is a marginal trend for malignant lymphomas (P_{Trend}=0.092, P_{Hist}=0.085) and no trend for kidney tumors.

The **EPA**[61] concluded the findings in this study were not treatment related based upon the tumors appearing only in the high dose group, a lack of statistical significance between the response in this group and control response and that these tumors are commonly observed in mice as both spontaneous and treatment related effects. There is no scientific support for excluding positive findings in the highest dose group, a view also held by the **SAP**[54]. I have already commented on how EPA's guidelines treat trend tests and Fisher's Exact test results, although in this case, the value of the comparison of the highest exposure group to controls, p_{Fisher}=0.059, is marginally significant. The argument regarding the frequency of this tumor in controls is addressed directly by the evaluation against the historical control rates; if these rates were high enough to exclude this finding, P_{Hist} would have be above 0.05 instead of 0.001. The mean historical control incidence of hemangiosarcomas in controls from two-year cancer bioassays in CD-1 mice is 2.5% and the response seen in the high-dose group is 8.9%. The **SAP**[54] stated very clearly that the practice, being used by the EPA, of negating a positive finding because of historical control data was not acceptable[54]. (page 63). The EPA Cancer Guidelines[33] state this very clearly "*...statistically significant increases in*

tumors should not be discounted simply because incidence rates in the treated groups are within the range of historical controls or because incidence rates in the concurrent controls are somewhat lower than average."

In summary, this study shows a positive result for hemangiosarcomas in male CD-1 mice and will be included in the overall evaluation of causation.

Wood et al., (2009)[87] exposed CD-1 mice to glyphosate (95.7% pure) in feed for 80 weeks. Fifty-one animals/groups/sex were tested in four exposure groups (see Table 11).

There was no effect on survival and no information suggesting the study exceeded the MTD.

No increase in kidney tumors or hemangiosarcomas were seen in this study. There was a monotonic increase in lung adenocarcinomas (p_{Trend}=0.038, p_{Hist}=0.031) in females and a monotonic increase in malignant lymphomas (p_{Trend}=0.008, p_{Hist}=0.007) in males. The historical control incidence for this study is different from the earlier studies because this study is only for 80 weeks instead of 104 weeks (two years); the historical control rate for malignant lymphomas in CD-1 mice after 80 weeks is 2.6% instead of 6.2%, the historical control rate at two years[101].

For lung adenocarcinomas, the **EPA**[61] again argued a lack of significance for pairwise comparisons (in violation of its guidelines) and that there was no evidence of progression from adenomas to carcinomas. Even though there was no increase in lung adenomas as a function of exposure, it is possible to have an increase in lung adenocarcinomas without an associated increase in adenomas[105]. For malignant lymphomas, EPA notes that there was a statistically significant response and that the high dose was significantly different from control (p_{Fisher}=0.028), but then uses an argument based upon the number of analyses done in this study to adjust the Fisher Exact test p-value to 0.082 (an adjustment for multiple comparisons is indeed warranted in evaluating the outcomes of these animal cancer studies, this will be addressed later in my report in the evaluation of all of the studies combined).

The **EPA**[61] uses historical control data[102, 106] to exclude the malignant lymphomas and cite a mean response of 4.5% and a range of 1.5% to 21.7%. **Son and Gopinath (2004)**[106] saw 21 animals out of 1453 examined prior to 80 weeks with lung adenocarcinomas (1.4%). **Giknis and Clifford (2005)**[102] saw a mean rate of 4.5% with a range of 0% to 21.7% in 52 studies which included mostly 78 week controls (26 studies) and 104 week controls (21 studies). Including only studies of 80 weeks or less, the rate in **Giknis and Clifford (2005)** is 37/1372=2.7% with a range of 0% to 14%. **Giknis and Clifford (2000)**[101] (the reference I have been citing) did a similar evaluation, using mostly the same data as their 2005 paper and saw an average tumor incidence before 80 weeks of 2.6% with a range of 0% to 14%. Based upon its flawed interpretation of the **Giknis and Clifford (2005)** historical controls, EPA argues that the incidence of concurrent controls in the study was low (it was 0%) and rejected the positive finding. In fact, of the 26 animals in the 18-month control groups evaluated by **Giknis and Clifford (2005),** eight (31%) had response of 0% and eight (31%) had only one tumor.

The evaluation used by the EPA is incorrect. In addition, as noted earlier, the use of historical control data to negate a positive finding is not supported by **EPA's guidelines**[33, 54] or its **SAP**[54].

There was an increase in the number of animals with multiple malignant tumors (P_{Trend}=0.050)

In summary, this study shows a positive result for malignant lymphomas and lung adenocarcinomas in male CD-1 mice and will be included in the overall evaluation of causation.

Table 11: Tumors of interest in male and female CD-1 mice from the 18-month feeding study of **Wood et al. (2009)**[87]

Tumor	Sex	Doses (mg/kg/day)				p-values
	Male	0	71.4	234.2	810	
	Female	0	97.9	299.5	1081.2	
Kidney Adenoma[1]	Male	0/51	0/51	0/51	0/51	P_{Trend}=1
Malignant Lymphoma[2]	Male	0/51	1/51	2/51	5/51*	P_{Trend}=0.008 P_{Hist}=0.007
Hemangiosarcoma	Male	0/51	0/51	0/51	0/51	P_{Trend}=1
Lung Adenocarcinoma[3]	Male	5/51	5/51	7/51	11/51	p_{Trend}=0.038 P_{Hist}=0.031
Hemangiosarcoma[4]	Female	1/51	1/51	1/51	1/51	p_{Trend}=0.509 P_{Hist}=0.820
Animals with Malignant Neoplasms	Male	14/51	20/51	17/51	20/51	P_{Trend}=0.249
Animals with Malignant Neoplasms	Female	23/51	15/51	17/51	18/51	P_{Trend}=0.598
Animals with multiple malignant tumors	Male	1/51	2/51	3/51	5/51	P_{Trend}=0.050

*- p_{Fisher}<0.05, **- p_{Fisher}<0.01, [1]historical rate=0.44%, [2]historical rate=2.6%, [3]Historical rate=2.5%, [4]Historical Control Rate=0.14%

Sugimoto (1997)[86] exposed CD-1 mice to glyphosate (94.61-95.67% pure) in feed for two years. Fifty animals/group/sex were tested in four exposure groups (see Table 12).

There were no effects of treatment on survival and no indication the highest dose had exceeded the MTD.

Kidney adenomas (p_{Trend}=0.063, p_{Hist}=0.005), malignant lymphomas (p_{Trend}=0.019, p_{Hist}=0.017) and hemangiosarcomas (p_{Trend}=0.063, p_{Hist}=0.004) in male mice and hemangiosarcomas (p_{Trend}=0.002, p_{Hist}<0.001) in female mice all showed increased tumor incidence with increasing dose. The evaluation of lung adenocarcinomas in males showed no significant dose-related trend (p_{Trend}=0.156, p_{Hist}=0.140). This study also had an increase in animals with any malignancy in males (p_{Trend}=0.004) but not in

females (p_{Trend}=0.372). Note that no hemangiosarcomas were seen in the 26 control groups evaluated by **Giknis and Clifford (2000)** so the development of an estimate of the historical control response is difficult (if the historical control rate is 0, then any observed response other than 0 has a p-value of 0). The fact that this tumor was never seen in the historical controls should strongly support any positive finding as being significant. However, to still allow for a test using historical control data, I used the historical control estimate of the mean response that would result in a 5% chance of seeing no tumors in 1149 animals. This estimated historical control response value was 0.0026. This value was used in the analysis for hemangiosarcomas in male CD-1 mice exposed for 18 months (p_{Hist} <0.001).

Table 12: Tumors of interest in male and female CD-1 mice from the 18-month feeding study of **Sugimoto (1997)**[86]

Tumor	Sex	Doses (mg/kg/day)				p-values
	Male	0	165	838.1	4348	
	Female	0	153.2	786.8	4116	
Kidney Adenoma[1]	Male	0/50	0/50	0/50	2/50	P_{Trend}=0.063 P_{Hist}=0.005
Malignant Lymphoma[2]	Male	2/50	2/50	0/50	6/50	P_{Trend}=0.019 P_{Hist}=0.017
Hemangiosarcoma[3]	Male	0/50	0/50	0/50	2/50	P_{Trend}=0.063 P_{Hist}=0.004
Hemangiosarcoma[4]	Female	0/50	0/50	2/50	5/50*	P_{Trend}=0.002 P_{Hist}<0.001
Lung Adenocarcinoma[5]	Male	1/50	1/50	6/50	4/50	P_{Trend}=0.156 P_{Hist}=0.140
Number of animals with Malignant Neoplasms	Male	5/50	5/50	11/50	16/50**	P_{Trend}=0.004
Number of animals with Malignant Neoplasms	Female	9/50	13/50	16/50	13/50	P_{Trend}=0.372

*- p_{Fisher}<0.05, **- p_{Fisher}<0.01, [1]historical rate=0.44%, [2]historical rate=2.6%, [3]historical rate=0/1424 (0.26% - 95% confidence limit), [4]Historical Control Rate=0.14%, [5]Historical rate=2.5%

EPA[61] only addressed the hemangiosarcomas in the female mice and did not note any other significant effects. For the females, EPA argued that the high dose was approximately four times higher than the current recommended high dose from the **OECD guidelines**[107]. This study was correctly designed under the previous guidelines (the limit was <5% in feed) and there is no indication that this dose exceeded the MTD. The EPA also argued that when the p-value for Fisher's Exact test was adjusted for multiple comparisons, the new p-value for the high-dose group for hemangiosarcomas was 0.055. However, that adjustment does not account for the extreme rarity of this tumor (2 cases in 1149 female mice in the historical control database with 25 of the 26 control groups examined having zero tumors and the remaining study having just two)

and any adjustment against such a low background rate would reduce its adjusted p-value considerably.

For the hemangiosarcomas in males, none of the 26 historical control groups examined by **Giknis and Clifford (2000)** had hemangiosarcomas, making this a very rare tumor in males prior to 80 weeks on study. The malignant lymphomas in males are statistically significant against both the controls and the historical controls. Finally, there is clearly an overall increase of malignancies in the males.

In summary, this study shows a positive result for kidney adenomas, malignant lymphomas and hemangiosarcomas in male CD-1 mice, hemangiosarcomas in female CD-1 mice and an overall increase in malignancies as a function of exposure in male CD-1 mice. This study will be included in the overall evaluation of causation.

Kumar (2001)[83] exposed Swiss Albino mice to glyphosate (>95% purity) in feed for two years. Fifty animals/group/sex were tested in four exposure groups (see Table 13).

The survival was decreased in the highest exposure group but this was not statistically significant and there was no other data indicating the MTD was exceeded for this study.

Kidney adenomas (p_{Trend}=0.062), malignant lymphomas (p_{Trend}=0.102, p_{Hist} =0.070) in male mice demonstrated marginal statistical significance and hemangiosarcomas (p_{Trend}=0.502) in male mice all demonstrated no statistical significance. In this study, not all animals in the low- and mid- dose groups were evaluated for kidney tumors, so a second analysis was done based on only the animals examined in these two groups (p_{Trend}=0.062). No historical control data was available for hemangiosarcomas and kidney adenomas in Swiss Albino mice. For the malignant lymphomas, EFSA provided a historical control data set showing a mean response of 46/250=0.184 (18.4%) with a range of 6% to 30%. Using this historical control data, the trend is only marginally significant (p_{Hist}=0.07). There is a statistically significant increase in the highest exposure group p_{Fisher}=0.038. I have some concern that the responses at two of the doses are outside of the historical control range and the third dose is at the upper limit of the historical control range. However, this is a small historical control dataset for a tumor with a relatively high background tumor rate, thus placing too much emphasis on this historical control population is not warranted.

In summary, this study shows support for an increase for malignant lymphomas as a function of exposure in male Swiss Albino mice. This study will be included in the overall evaluation of causation.

Table 13: Tumors of interest in male and female Swiss Albino mice from the 18-month feeding study of **Kumar (2001)**[83]

Tumor	Sex	Doses (mg/kg/day)				p-values
	Male	0	14.5	149.7	1453	
	Female	0	15	151.2	1466.8	
Kidney Adenoma	Male	0/50	0/50	1/50	2/50	P_{Trend}=0.062
Kidney Adenoma (only tissues examined microscopically)	Male	0/50	0/26	1/22	2/50	P_{Trend}=0.062
Malignant Lymphoma[1]	Male	10/50	15/50	16/50	19/50*	P_{Trend}=0.102 P_{Hist}=0.070
Hemangiosarcoma	Male	0/50	0/50	2/50	0/50	P_{Trend}=0.502

*- p_{Fisher}<0.05, **- p_{Fisher}<0.01, [1]Historical control rate=0.184 (46/250 mice)

Pavkov and Turner (1987)[84] exposed CD-1 mice to glyphosate trimesium salt (56.2%) and 1% propylene glycol (wet weight vehicle) in feed for two years. Eighty animals/sex/group were tested in control, low- and mid-dose groups and 90 animals/sex were tested at the high dose. Exposure levels were 0, 11.7, 118 and 991 mg/kg/day in males and 0, 16, 159 and 1341 mg/kg/day in females. **EPA**[61] lists this study as completely negative for any cancer findings. No details on this study are provided by the EPA nor is it listed in the **Greim et al. (2015)**[90] manuscript. There was limited information on this study in a Data Evaluation Report from EPA (accession number 4021 40-06) that discussed findings from this study. EPA noted that body weight and food consumption were reduced in the highest exposure group, but the actual amounts of these reductions were not available. They also noted that the authors failed to make it clear that the tumors reported in the study had been histopathologically validated. Data was presented for tumors in the livers and lungs of male mice and the lungs of female mice. No other data is provided.

This study is not acceptable for inclusion in the evaluation of causation due to the lack of information on the tumor incidence in tissues other than liver and lung.

George et al. (2010)[81] exposed groups of 20 male Swiss Albino mice to a glyphosate formulation (Roundup Original, 36g/L glyphosate) at a dose of 25 mg/kg (glyphosate equivalent dose) topically three times per week, topically once followed one week later by 12-o-tetradecanoylphorbol-13-acetate (TPA) three times per week, topically three times per week for three weeks followed one week later by TPA three times per week, or a single topical application of 7,12-dimethyl-benz[a]anthracene (DMBA) followed one week later by topical application of glyphosate three times per week for a total period of 32 weeks. Appropriate untreated, DMBA-treated, and TPA-treated controls were included. The group exposed to DMBA followed by glyphosate demonstrated a significant increase ($p<0.05$) in the number of animals with tumors (40% of the treated animals versus no tumors in the controls) indicating glyphosate has a promotional effect

on carcinogenesis in the two-stage model in skin. This study addresses the question of whether glyphosate is more likely to cause skin tumors through initiation (starting the cancer process) or promotion (moving the process along after it starts). This study supports the overall concept that glyphosate can have an impact on tumor incidence.

EPA[61] discounted this study because it included only 20 animals per group, tested only males and did not conduct a histopathological analysis. It is hard to understand how EPA could reject a positive finding using 20 mice; typically one would ignore a negative study that had too few animals as not having sufficient statistical power to see an effect but never reject positive findings for this reason. Also, 20 animals per group is common for skin-painting initiation-promotion studies like the one presented here. Doing a study in only males is not a reason to ignore the positive findings in a study. Finally, in initiation-promotion studies of mouse skin, histopathological evaluation would be done if one were interested in separating papillomas from carcinomas. It is highly unlikely that the lesions seen in 40% of the DMBA/glyphosate treated mice were not papillomas or carcinomas.

Some members of the EPA SAP noted[54] that the rodent data were consistent with glyphosate acting as a tumor promoter but, because "*[t]here has been no direct test of this hypothesis (such as in a standard initiation-promotion bioassay)...,*" this "*conclusion was speculative.*" (page #). Because the EPA dismissed this study without any discussion, the SAP did not recognize there was an initiation-promotion supporting a promotional effect of glyphosate.

This study is included in the evaluation of causality as support for a promotional effect of glyphosate on some tumors.

Joint Analysis

In their evaluation of the mouse studies, **EPA**[61] and **EFSA**[88] chose to challenge the results in each study separately, dismiss the studies as showing no effect, and never compared results across the various studies. In response to the evaluation done by the **IARC**[30], **EFSA**[89] extracted the original data and did trend tests on kidney tumors, malignant lymphomas and hemangiosarcomas in male mice in five of the mouse studies, the same five studies I believe are acceptable for a causation analysis. Rather than formally evaluate these cancer responses for consistency by pooling the data where appropriate, EPA and EFSA simply produced a table with the responses for each dose group in each study and concluded (subjectively) they were inconsistent. In addition, EPA and EFSA argued that doses above 1000 mg/kg/day (there are only two of these) were outside the range of what would be tested today under OECD guidelines and should be excluded. I will now address both points.

In CD-1 mice, there are four useful animal carcinogenicity studies and one study in Swiss Albino mice. As with the rats, consistency across studies can be addressed in two ways. The first is by simply looking at the overall findings to evaluate where they agree or disagree in terms of statistical significance. Table 14 summarizes the positive and negative findings for all five cancers in which at least one study in CD-1 mice showed a significant trend. It is clear that not every tumor shows a positive trend with glyphosate

exposure in every study. For hemangiosarcomas in males, there are clear positive findings in the studies by **Sugimoto (1997)** and **Atkinson et al. (1993)** and non-significant responses in **Wood et al. (2009**) and **Knezevich and Hogan (1983)**. In females, hemangiosarcomas are only present in the study by **Sugimoto (1997)**. Malignant lymphomas in males are clearly positive in two studies[86, 87] and marginally positive in a third[80] but negative in the fourth[82]. Both of the strong positive studies exposed animals for 18 months. Kidney tumors in males are positive in two studies[82, 86] and negative in the remaining two[80, 87]. Lung adenocarcinomas in males are only positive in the study by **Wood et al. (2009)**. **Sugimoto (1997)** had four clearly positive associations between tumors and glyphosate while the others had two or less.

Table 14: Summary of significance tests for 5 tumors from 4 studies in CD-1 Mice

Study	Months on Study	Neoplasm				
		Hemangio-sarcoma (male)	Hemangi-sarcoma (female)	Malignant Lymphoma (male)	Kidney Tumor (male)	Lung Adeno-carcinoma (male)
Sugimoto 1997[86]	18	+/+++[1]	+++/+++	++/++	+/+++	-/-
Wood 2009[87]	18	-/-	-/-	+++/+++	-/-	++/++
Sugimoto & Wood Pooled		++/+++	+++/+++	+++/+++	++/+++	-/-
Atkinson 1993[80]	24	+++/+++	-/-	+/+	-/-	-/-
Knezevich 1983[82]	24	-/-	NA	-/-	+/++	-/-
Atkinson & Knezevich Pooled		-/-	NA	-/-	++/+	-/-
All CD-1 Studies Pooled		++/++	+++/+++	+/+	+++/+++	-/-

[1]entries are p_{Trend}/p_{Hist} with values: – p>0.1, + 0.1≥p>0.05, ++ 0.05≥p>0.01, +++ p≤0.01

As seen for the rat studies, this simple evaluation of the positive versus negative findings fails to resolve the issue of which findings are driving the overall responses in these data. To do this, I will again pool the studies. Table 14 summarizes the pooled analyses.

For kidney tumors in males, pooling the two 18-month studies yields significant increases in incidence (p_{Trend}=0.016, p_{Hist}=0.003) as does pooling of the two year studies (p_{Trend}=0.033, p_{Hist}=0.054). Pooling all four studies results in (p_{Trend}=0.006, p_{Hist}=0.007), thus the positive trend remains. **Knezevich and Hogan (1983)** saw a 4% response for kidney carcinomas in their highest exposure group. The largest response seen for kidney carcinomas in controls in 48 studies by **Giknis and Clifford (2000**) and in 52

studies by **Giknis and Clifford (2005)** was 2% and in the control groups from 11 two-year cancer studies, **Chandra and Frith (1992)**[100] saw only one animal out of 725 with a kidney carcinoma. In 46 control datasets, **Giknis and Clifford (2000)** saw 39 control groups with no adenomas, five with one adenoma and two with two adenomas; both 24-month studies saw two adenomas in the highest exposure group, a very rare finding. To better illustrate, there are 16 groups of animals in the four studies. For any one group, there is a 2/44 or 4.3% chance of getting a response 4% or larger. The chances of randomly getting 3 or more such responses in 16 groups is 2.9% and the chances of two of these being in any two of the four highest exposure groups is 0.01. In summary, the strong finding in two of the four studies, the positive finding when all four studies are pooled and the very low probability that this is due to chance when compared to historical controls support the conclusion that glyphosate causes kidney tumors in male mice.

For malignant lymphomas in males, pooling the two 18-month studies, **Sugimoto (1997)** and **Wood et al. (2009**), results in a significant trend (p_{Trend}=0.006, p_{Hist}=0.006). Pooling the two 24-month studies, **Knezevich and Hogan (1983)** and **Atkinson et al. (1993)**, yields (p_{Trend}=0.640, p_{Hist}=0.649). The main differences between these two findings is in the control response; the pooled control response at 24 months is 6/99 (6%) versus 2/101 at 18 months (2%). This is expected since, in the absence of any exposure, tumor rates increase as a function of age[5]. **Giknis and Clifford (2000)** show a control response at 18 months of 4% and a control response at 24 months of 6% (matching the value for the pooled studies). Pooling all four studies results in (p_{Trend}=0.086, p_{Hist}=0.080). However, the responses seen for malignant lymphomas in controls by **Giknis and Clifford (2000)** show only one historical control group in twenty-six 18-month groups with 10% or higher response. The responses at the high doses (10% and 12%) in the two 18-month studies are very unlikely to have arisen by chance. There are eight groups of animals in the two studies. For any one group, there is a 1/26 or 3.8% chance of getting a response of at least 10% based on the 26 control groups from **Giknis and Clifford (2000)**. The chances of getting two or more such responses in eight groups is 0.035 and the chances of these being in three of the four highest exposure groups is 0.004. For the 24-month studies, the higher background rate makes it difficult to identify a small change in incidence, thus the findings in the 24-month studies and the 18-month studies are not inconsistent. In summary, the very strong findings in the 18-month studies, the very strong positive findings when the two 18-month studies are pooled, the low probability that the responses seen in the 18-month studies are due to chance, and the increase in malignant lymphomas in the 18-month study in Swiss Albino mice[83] support the conclusion that glyphosate causes malignant lymphoma in male mice.

For hemangiosarcomas in males, pooling the two 18-month studies results in a significant trend (p_{Trend}=0.015, p_{Hist}=0.002). Pooling the two 24-month studies yields (p_{Trend}=0.486, p_{Hist}=0.429). The main difference between these two findings is the 0/50 response in animals exposed at 4841 mg/kg/day in the study by **Knezevich and Hogan (1983)**. Removing this one exposure group in the pooled 24-month analysis yields (p_{Trend}<0.001, p_{Hist}<0.001). Pooling all four studies results in (p_{Trend}=0.046, p_{Hist}=0.043). No hemangiomas were seen in controls groups from twenty-six 18-month studies by

Giknis and Clifford (2000) so the two hemangiosarcomas seen in the high dose group in the study by **Sugimoto (1997)** are biologically very significant. For the 24-month historical controls, only two out of 20 control groups had a response greater than 8%. In summary, the very strong findings in the 18-month studies, the positive finding when all four studies are pooled and the low probability that the responses seen in the 18-month studies are due to chance support the conclusion that glyphosate causes hemangiosarcomas in male CD-1 mice.

For hemangiosarcomas in females, pooling the two 18-month studies results in a significant trend (p_{Trend}=0.002, p_{Hist}<0.001). Data was available for only one of the two-year studies and this study had (p_{Trend}=0.439, p_{Hist}=0.376). Pooling all three studies results in (p_{Trend}=0.001, p_{Hist}=0.002). In 18-month historical controls, **Giknis and Clifford (2000)** saw only one study in 26 studies with any response (2/49 or 4%). In the **Sugimoto (1997)** study, the second highest dose had a response of 2/50 and the highest dose had a response of 5/50; this cannot be due to chance. In summary, the very strong findings in one 18-month study, the positive finding when all three studies are pooled and the low probability that the responses seen in the **Sugimoto (1997)** study are due to chance, support the conclusion that glyphosate causes hemangiosarcomas in female CD-1 mice.

For lung adenocarcinomas in male CD-1 mice, pooling the two 18-month studies results shows no significant trend (p_{Trend}=0.155, p_{Hist}0.126). Pooling the two 24 month studies yields (p_{Trend}=0.989, p_{Hist}=0.993). Pooling all four studies results in (p_{Trend}=0.731, p_{Hist}=0.744). In summary, the moderate findings in one 24 month study, and the negative finding when any studies are pooled suggest that the linkage between glyphosate and lung adenocarcinomas in male CD-1 mice is due to chance.

The one study in Swiss Albino mice[83] was effectively negative for all endpoints except malignant lymphomas where a significant tumor response was seen in the highest dose group and the historical control evaluation produced a marginally significant finding (p_{Hist}=0.07). Considering the findings for malignant lymphoma in CD-1 mice, glyphosate appears to also cause malignant lymphomas in male Swiss Albino mice from the study of **Kumar (2001)**.

To summarize the findings in mice, glyphosate causes hemangiosarcomas, kidney tumors and malignant lymphomas in male CD-1 mice and hemangiosarcomas in female CD-1 mice after 18 months of exposure, kidney tumors in male CD-1 mice after 24 months exposure and probably malignant lymphomas in male Swiss albino mice. When 18-month and 24-month studies are pooled, there is a significant increase in hemangiosarcomas in male and female mice and kidney tumors in male mice.

Discussion and Summary Animal Carcinogenicity Studies

As noted earlier, there has been a suggestion that using doses substantially larger than 1000 mg/kg/day exceeds the current limit dose set by the OECD. The only place in the **OECD guidance**[66] that addresses a dose of 1000 mg/kg/day is in paragraph 23 which reads:

> *"For the chronic toxicity phase of the study, a full study using three dose levels may not be considered necessary, if it can be anticipated that a test at one dose level, equivalent to at least 1000 mg/kg body weight/day, is unlikely to produce adverse effects. This should be based on information from preliminary studies and a consideration that toxicity would not be expected, based upon data from structurally related substances. A limit of 1000 mg/kg body weight/day may apply except when human exposure indicates the need for a higher dose level to be used."*

This language does not preclude the use of a dose exceeding 1000 mg/kg/day nor does it advocate ignoring such doses when evaluating the results of an animal carcinogenicity study. In fact, the reasons for excluding a dose in an animal carcinogenicity study are clearly outlined in paragraph 90 within **OECD guidance**[59] and reads:

> *"If the main objective of the study is to identify a cancer hazard, there is broad acceptance that the top dose should ideally provide some signs of toxicity such as slight depression of body weight gain (not more than 10%), without causing e.g., tissue necrosis or metabolic saturation and without substantially altering normal life span due to effects other than tumours. Excessive toxicity at the top dose level (or any other dose level) may compromise the usefulness of the study and/or quality of data generated. Criteria that have evolved for the selection of an adequate top dose level include: (in particular) toxicokinetics; saturation of absorption; results of previous repeated dose toxicity studies; the MOA and the MTD."*

While one study has a slight decrease in body-weight gain, there are no indications in any other studies of an exceedance in dose that would support ignoring the findings from any exposure group.

EPA[33] uses a slightly different criteria to determine which dose to include or exclude based on an earlier OECD document. These are spelled out in EPA's guideline document for carcinogenicity risk assessment[33]

> *"Other signs of treatment-related toxicity associated with an excessive high dose may include (a) significant reduction of body weight gain (e.g., greater than 10%), (b) significant increases in abnormal behavioral and clinical signs, (c) significant changes in hematology or clinical chemistry, (d) saturation of absorption and detoxification mechanisms, or (e) marked changes in organ weight, morphology, and histopathology. It should be noted that practical upper limits have been established to avoid the use of excessively high doses in long-term carcinogenicity studies of environmental chemicals (e.g., 5% of the test substance in the feed for dietary studies or 1 g/kg body weight for oral gavage studies [OECD, 1981])."* As before, this applies to only one study presented in this review.

Both of these guidelines make good scientific sense. In the 12 acceptable rodent carcinogenicity studies included in this evaluation, no study had sufficient toxicity at the highest dose to justify removing the highest dose from the analysis. Hence, the analyses presented here did not drop the doses >1000 mg/kg/day. This is also supported by one

member of the **EPA's SAP**[54].

Twenty chronic rodent carcinogenicity studies have been done using glyphosate as the test compound. Eight of these studies are unacceptable for use in an evaluation of causality leaving seven studies in rats and five studies in mice. Because of the large number of evaluations done in an individual animal carcinogenicity study, there is concern that the false-positive rates could be exaggerated. For example, if 20 evaluations are done and a finding is deemed significant if $p_{Trend}<0.05$, then you would expect that 20*0.05=1 evaluation would be positive simply due to chance.

The EPA asked the SAP to comment on its evaluation of glyphosate[61] at a meeting in Washington, DC in December 2016[54]. Many comments were received from outside experts at this meeting; one such set of comments came from Dr. J. K. **Haseman (2016)**[108]. **Haseman (2016)** directly addressed the false-positive error rate and concluded that the results seen in these studies were due to chance. He did this by deciding how many evaluations were likely for each study (broken into sex-by-species groups) and then aggregating the findings. He concluded that the effective number of analyses were 10.5 in male mice, 15 for female mice, 21.5 for male rats, and 25.5 for female rats. **Haseman (2016)** made two assumptions in his analysis that are not valid. The first was that all of the possible trend tests had been done on all of the sites he considered reasonable for such an evaluation. He identified eight positive findings. However, EPA had not evaluated all of the sites nor had they considered doing a formal analysis using historical control data. EPA identified eight sex/species groups that had at most one positive tumor finding using the trend test with $p_{Trend}\leq0.05$. In Tables 1-14 above, I have identified 18 tumors with $p_{Trend}\leq0.05$ or $p_{Hist}\leq0.05$ and 11 with $p_{Trend}\leq0.01$ or $p_{Hist}\leq0.01$ (Table 15). Secondly, Dr. Haseman assumed one could aggregate all the studies into one large analysis of Type-1 error. However, inference in these studies is always made by sex/species/strain (e.g. glyphosate causes hemangiosarcomas in male CD-1 mice; not glyphosate causes cancer in rodents), and the analysis should have been done by grouping each separately. Table 15 shows these analyses as well as the aggregated analysis for all of the acceptable studies.

With the exception of male Sprague-Dawley rats, the observed number of tumors are at or near the expected number for the different sex/strain groups in rats (Table 15). For male Sprague-Dawley rats, 0.8 cases with $p_{Trend}\leq0.01$ or $p_{Hist}\leq0.01$ are expected and two were observed (p=0.21). In female CD-1 mice and Swiss Albino mice, the expected and observed numbers are approximately equal. However, in male CD-1 mice, there were 2.1 tumors expected for $p_{Trend}\leq0.05$ or $p_{Hist}\leq0.05$ and eight were observed ($p<0.001$) and there were 0.4 expected for $p_{Trend}\leq0.01$ or $p_{Hist}\leq0.01$ and five were observed ($p<0.001$). This clearly could not have occurred by chance alone. Even if one incorrectly groups all sexes and species together, there are 4.6 expected responses for $p_{Trend}\leq0.01$ or $p_{Hist}\leq0.01$ and 11 observed (p=0.007). Thus, chance does not explain the positive results seen in these studies.

Table 15: Observed versus expected tumor sites with significant trends in the 12 acceptable rodent carcinogenicity studies using glyphosate.

Species	Strain	Sex	Total Sites[1]	Exp. <0.05	Obs. <0.05	Tumors[2] p<0.05	Exp. <0.01	Obs. <0.01	Tumors p<0.01
Rat (7 studies)	Sprague-Dawley (4 studies)	M	86	4.3	4	TICT, TFAC, KA, HA	0.9	2	TICT, KA
		F	102	5.1	1	TCCC	1.0	1	TCCC
	Wistar (3 studies)	M	64.5	3.2	2	HA, SK	0.6	1	HA
		F	76.5	3.8	2	MC, MAC	0.8	1	MAC
Mouse (5 studies)	CD-1 (4 studies)	M	42	2.1	8	KA, KC, KAC, H(2)[3], ML(2), LAC	0.4	5	KA,KC, H(2), ML
		F	60	3	1	H	0.6	1	H
	Albino (1 study)	M	10.5	0.5	0		0.1	0	
		F	15	0.8	0		0.2	0	
Rats (7 studies)	All (7 studies)	M	150.5	7.5	6	TICT, KA, HA(2), TFAC, SK	1.5	3	TICT, KA, HA
		F	178.5	8.9	3	TCCC, MC, MAC	1.8	2	TCCC, MAC
		Both	329	16.5	9	TICT, KA, HA(2), TFAC, SK, TCCC, MC, MAC	3.3	5	TICT, KA, HA, TCCC, MAC
Mice (5 studies)	All (5 studies)	M	52.5	2.6	8	KA, KC, KAC, H(2), ML(2), LAC	0.5	5	KA,KC, H(2), ML
		F	75	3.8	1	H	0.7	1	H
		Both	127.5	6.4	9	KA, KC, KAC, H(3)[3], ML(2), LAC	1.3	6	KA,KC, H(3), ML
All (12 studies)	All (12 studies)	M	203	10.1	14	TICT, KA(2), HA(2), TFAC, SK, KC, KAC, H(2), ML(2), LAC	2.0	8	TICT, HA, KA(2),KC, H(2), ML
		F	253.5	12.7	4	TCCC, MC, MAC, H	2.5	3	TCCC, MAC, H
		Both	456.5	22.8	18	TICT, KA(2), HA(2), TFAC, SK, KC, KAC, H(3), ML(2), LAC, TCCC, MC, MAC	4.6	11	TICT, HA, KA(2), KC, H(3), ML, TCCC, MAC

[1]Number of sites examined is based upon suggestions by Dr. J. Haseman in his written testimony to the EPA; male mice – 10.5 sites; female mice – 15 sites; male rats – 21.5 sites; female rats – 25.5 sites

[2]Tumor abbreviations are: KA – kidney adenoma; KC – kidney carcinoma; KAC – kidney adenoma or carcinoma; H – hemangiosarcoma; HA – hepatocellular adenoma; LAC – lung adenoma or adenocarcinoma; ML – malignant lymphoma; MC – mammary gland carcinoma; MAC – mammary gland adenoma or carcinoma; TCCC – thyroid C-cell carcinoma; TFAC – thyroid follicular cell adenoma or carcinoma; TICT – testes interstitial cell tumor; SK – skin keratocanthoma

[3](x): x studies with this result

Conclusion for Animal Carcinogenicity Studies

There are several general issues that pertain to all animal carcinogenicity studies. There is considerable genetic variability across animal strains both over time and space. It is difficult to compare experiments done in different laboratories even when using the same strain of animal. This is obvious when you examine the rates for hepatocellular adenomas in Wistar rats across the three studies using this strain. Thus, each study should be considered separately with regard to the findings in that study before being compared across studies.

The use of a p-value of 0.05 as the cut off for increasing tumor incidence does not account for trends in the data across multiple studies. Three studies with marginal responses of 6-8% in a given tumor could, when pooled for analysis, lead to highly significant findings. This issue is well-recognized in epidemiology but not usually

considered in toxicology because of a lack of replicate studies. This case is fairly unique because of the larger number of studies available for analysis and requires a more rigorous evaluation of the data such as the pooled analysis presented in this report.

Pooling of the data for the evaluation of replicate studies makes sense as it addresses the question "Does the data as a whole support a finding of increased cancer incidence in these studies?" Some toxicologists may argue that the studies are not replicates and hence cannot be pooled. But if they are not replicates, then they cannot be compared to see if there is consistency across the studies. This is because there may be some subtle change from one study to another that leads to a positive finding in one study but a negative finding in other studies. Thus, either the studies are not good replicates so you cannot compare across studies and you cannot pool them, or they are good replicates so you can compare across studies and you can pool them. There is no argument that would support a comparison across studies that is appropriate when pooling is inappropriate.

There were seven rat studies and five mouse studies that were of sufficient quality and with sufficient details available for inclusion in this evaluation.

Glyphosate has been demonstrated to cause cancer in two strains of rats and one strain of mice. Glyphosate causes hepatocellular adenomas in male Wistar rats and male Sprague-Dawley rats, mammary gland adenomas and adenocarcinomas in female Wistar rats and kidney adenomas in male Sprague-Dawley rats. Glyphosate causes hemangiosarcomas, kidney tumors and malignant lymphomas in male CD-1 mice and hemangiosarcomas in female CD-1 mice and possibly causes malignant lymphomas in male Swiss albino mice. Thus, glyphosate causes cancer in mammals.

Mechanisms Relating to Carcinogenicity

Many human carcinogens act via a variety of mechanisms causing various biological changes, taking cells through multiple stages from functioning normally to becoming invasive with little or no growth control (carcinogenic). **Hanahan and Weinberg (2011)**[109] identified morphological changes in cells as they progress though this multistage process and correlated these with genetic alterations to develop what they refer to as the "hallmarks of cancer." These hallmarks deal with the entire process of carcinogenesis and not necessarily with the reasons that cells begin this process or the early stages in the process where normal protective systems within the cells remove potentially cancerous cells from the body. While tumors that arise from a chemical insult to the cell may be distinct from other tumors by mutational analysis, they all exhibit the hallmarks as described by **Hanahan and Weinberg (2011)**.

Systematic review of all data on the mechanisms by which a chemical causes cancer is complicated by the absence of widely accepted methods for evaluating mechanistic data to arrive at an objective conclusion on human hazards associated with carcinogenesis. Such systematic methods exist in other contexts[110], but are only now being accepted as a means of evaluating literature in toxicological evaluations[111-114].

In this portion of the report, I am focusing on the mechanisms that can cause cancer. **Smith et al. (2015)**[37] discussed the use of systematic review methods in identifying and using key information from the literature to characterize the mechanisms by which a chemical causes cancer. They identified 10 "Key Characteristics of Cancer" useful in facilitating a systematic and uniform approach to evaluating mechanistic data relevant to carcinogens. These 10 characteristics are presented in Table 16 (copied from Table 1 of **Smith et al. (2015)**[37]). While there is limited evidence on glyphosate for most of the key characteristics, genotoxicity (characteristic two) and oxidative stress (characteristic five) have sufficient evidence to warrant a full review.

Table 16: Key characteristics of carcinogens, **Smith et al. (2016)**[37]

Characteristic	Examples of relevant evidence
1. Is electrophilic or can be metabolically activated	Parent compound or metabolite with an electrophilic structure (e.g., epoxide, quinone), formation of DNA and protein adducts
2. Is genotoxic	DNA damage (DNA strand breaks, DNA–protein cross-links, unscheduled DNA synthesis), intercalation, gene mutations, cytogenetic changes (e.g., chromosome aberrations, micronuclei)
3. Alters DNA repair or causes genomic instability	Alterations of DNA replication or repair (e.g., topoisomerase II, base-excision or double-strand break repair)
4. Induces epigenetic alterations	DNA methylation, histone modification, microRNA expression
5. Induces oxidative stress	Oxygen radicals, oxidative stress, oxidative damage to macromolecules (e.g., DNA, lipids)
6. Induces chronic inflammation	Elevated white blood cells, myeloperoxidase activity, altered cytokine and/or chemokine production
7. Is immunosuppressive	Decreased immunosurveillance, immune system dysfunction
8. Modulates receptor-mediated effects	Receptor in/activation (e.g., ER, PPAR, AhR) or modulation of endogenous ligands (including hormones)
9. Causes immortalization	Inhibition of senescence, cell transformation
10. Alters cell proliferation, cell death or nutrient supply	Increased proliferation, decreased apoptosis, changes in growth factors, energetics and signaling pathways related to cellular replication or cell cycle control, angiogenesis

Abbreviations: AhR, aryl hydrocarbon receptor; ER, estrogen receptor; PPAR, peroxisome proliferator–activated receptor. Any of the 10 characteristics in this table could interact with any other (e.g., oxidative stress, DNA damage, and chronic inflammation), which when combined provides stronger evidence for a cancer mechanism than would oxidative stress alone.

Genotoxicity

Genotoxicity refers to the ability of an agent (chemical or otherwise) to damage the genetic material within a cell, thus increasing the risks for a mutation. Genotoxic substances interact with the genetic material, including DNA sequence and structure, to damage cells. DNA damage can occur in several different ways, including single- and double-strand breaks, cross-links between DNA bases and proteins, formation of micronuclei and chemical additions to the DNA.

Just because a chemical can damage DNA does not mean it will cause mutations. So, while all chemicals that cause mutations are genotoxic, all genotoxic chemicals are not necessarily mutagens. Does that mean that the genotoxicity of a chemical can be ignored if all assays used for identifying mutations in cells following exposure to a chemical are negative? The answer to that question is no and is tied to the limitations in tests for mutagenicity (the ability of a chemical to cause mutations in a cell). It is unusual to see an evaluation of the sequence of the entire genome before exposure with the same sequence after exposure to determine if the genome has been altered (mutation). There are assays that can evaluate a critical set of genes that have previously been associated with cancer outcomes (e.g. cancer oncogenes), but these are seldom applied. In general, mutagenicity tests are limited in the numbers of genes they actually screen and the manner in which these screens work.

Because screening for mutagenicity is limited in scope, any genetic damage caused by chemicals should raise concerns because of the possibility of a mutation arising from that genetic damage. In what follows, I will systematically review the scientific findings available for evaluating the genotoxic potential of glyphosate. This will be divided into six separate sources of data based on the biological source of that data: (1) data from exposed humans, (2) data from exposed human cells in a laboratory setting, (3) data from exposed mammals (non-human), (4) data from exposed cells of mammals (non-human) in the laboratory, (5) data from non-mammalian animals and others, and (5) data from cells from non-mammalian animals and others. These six areas are based upon the priorities one would apply to the data in terms of impacts. Seeing genotoxicity in humans is more important than seeing genotoxicity in other mammals, which is more important than seeing genotoxicity in non-mammalian systems. In addition, seeing genotoxicity in whole, living organisms (*in vivo*) carries greater weight than seeing responses in cells in the laboratory (*in vitro*). Basically, the closer the findings are to real, living human beings, the more weight they should be given.

The data being included in this review come from the peer-reviewed scientific literature, the summaries of reports in regulatory documents that are proprietary and for which I have limited access to the original work, and reports from industry that are proprietary to which I have been given greater access. All of these studies are included in the overall evaluation of causation.

Genotoxicity in Humans *in-vivo*

Three studies have evaluated the potential genotoxicity of glyphosate formulations in exposed humans. **Paz-y-Miño et al. (2007)**[115] analyzed the blood of 24 exposed

individuals (living within 3 kilometers of spraying) and 21 unexposed individuals (living 80 kilometers away from the spraying area) for DNA damage using the comet assay. All study subjects were from Ecuador and none of the controls or exposed individuals smoked, drank alcohol, took non-prescription drugs or had been exposed to pesticides during the course of their normal daily lives. Exposed and control individuals did some cultivating and harvesting but without pesticides or herbicides. Exposed individuals were analyzed within two months of spraying for the eradication of plants associated with illegal narcotics. An average of 200 cells per person were ranked between 0-400 depending on the amount of DNA in the comet's tail in order to calculate the mean amount of DNA damage. There was a significant difference between the mean total migration level of exposed individuals to controls ($p<0.001$). Data was given for each individual classified into five groups based upon the amount of DNA in the comet's tail. There was clearly a shift in the distribution of DNA in cells with the controls never seeing scores in the top two categories while all but three exposed had some scores in the top two categories. In essence, some of the DNA had been fragmented by the exposure.

In a second study by the same group, **Paz-y-Miño et al. (2011)**[116] evaluated the karyotypes (the chromosome count of the individuals and any alterations to the chromosomes as seen under a microscope) of 92 people living in 10 communities in northern Ecuador. Controls were from areas without spraying and both controls and exposed subjects had no history of exposure to smoking or other genotoxic compounds. This study saw no changes between controls and exposed subjects for 182 karyotypes evaluated.

Bolognesi et al. (2009)[117] studied women of reproductive age and their spouses in five areas of Colombia, four of which are subject to spraying for either narcotics control or sugar cane growing. There were 60 subjects from the Santa Marta area (organic coffee is grown without the use of pesticides), 52 from Boyaca (manual spraying for illicit drugs), 58 from Putumayo (aerial spraying for illicit drugs using a glyphosate formulation), 63 from Nariño (same exposure as Putumayo) and 28 from Valle del Cauca (aerial spraying of Roundup 747 (74.7% glyphosate) without additional adjuvant for sugar cane maturation). All subjects were interviewed with a standardized questionnaire designed to obtain information about current health status, health history, lifestyle and potential exposure to possible confounding factors (smoking, use of medicinal products, severe infections or viral diseases during the last six months, recent vaccinations, presence of known indoor/outdoor pollutants, exposure to diagnostic x-rays, and previous radio- or chemotherapy). In Santa Marta, blood samples were taken once, during the initial interview. In Boyaca, blood samples were taken at the initial interview and 1 month later. In Nariño, Putumayo and Valle del Cauca, blood samples were taken at the initial interview, within five days after spraying and 4 months later. In lymphocytes, binucleated cells with micronuclei (BNMN) were lowest in Santa Marta and similar in the four exposed regions prior to exposure. Statistically significant increases in BMNM in Nariño, Putumayo and Valle del Cauca were seen between first and second sampling. The mean BNMN in Nariño and Putumayo was greater in respondents who self-reported direct contact with sprayed fields, but differences were not statistically significant. Multiple linear regression demonstrated statistically

significant increases in BMNM in all four exposed regions post exposure when compared to pre-exposure and controlling for all other variables ($p<0.001$). The largest total change in mean BMNM values pre-exposure compared to immediate post exposure occurred in Valle del Cauca where spraying is done using Roundup with no additional adjuvant.

Kier (2015)[118] identified 16 additional studies of pesticide use that included some exposure to glyphosate. Eleven of the 16 studies demonstrated some degree of genotoxicity in the human populations studied but did not adequately attribute the exposure primarily to glyphosate so they are not included in this review.

In summary, two of the three studies in which genotoxicity endpoints were evaluated in humans in areas with exposure to glyphosate spraying showed statistically increased changes in DNA damage in blood. In the strongest study, in three areas where chromosomal damage (micronuclei) was examined in individuals pre- and post-spraying (<5 days) showed statistically significant increases. In one other area where post-exposure damage was measured one month after exposure, there was little change.

Genotoxicity in Human Cells (*in vitro*)

Studies have explored the *in vitro* genotoxicity of glyphosate using a variety of different cell types (lymphocytes, fibroblasts, and immortalized cells from cancers of the larynx, mouth, blood and liver) using several different assays for markers of genotoxicity with or without metabolic activation.

Mladinic et al. (2009)[119] induced DNA strand breaks (comet assay) from exposure to glyphosate (purity not given) in lymphocytes from three healthy human donors (questionnaire used to exclude genotoxic exposures) at concentrations of 3.5, 92.8 and 580 µg/ml with S9 activation and saw effects at only the highest doses for cells without S9 activation.

Alvarez-Moya et al. (2014)[120] conducted a similar study using lymphocytes from human volunteers (questionnaire used to exclude genotoxic exposures) and exposure to glyphosate (96% purity) at concentrations of 0.12, 1.2, 12 and 120 µg/ml. A significant increase in DNA strand breaks (comet assay) was seen for all exposure groups with a clear dose-response relationship without metabolic activation (metabolic activation was not tested).

Using human HEP-2 cells, **Manas et al. (2009)**[121] induced DNA damage (comet assay) by glyphosate (96% pure) at all concentrations ranging from 676 µg/ml to 1270 µg/ml (no S9 activation tested). Cell viability at the highest concentration was below 80% and values at the other concentrations were not given.

Monroy et al. (2005)[122] induced significant DNA damage (comet assay) in fibroblast GM 38 cells at concentrations of glyphosate (technical grade, purity not given) ranging from 676 µg/ml to 1000 µg/ml with a clear dose-response pattern. Over this same concentration range, they also saw concentration-dependent decreases in cell viability at all doses making the comet assay results difficult to interpret. In a similar analysis in the same paper, using fibrosarcoma HT1080 cells, they also saw concentration-

dependent DNA damage and loss of cell viability. Activation by S9 was not used in either experiment.

Lueken et al. (2004)[123] induced DNA damage (comet assay) in fibroblasts GM 5757 at a concentration of glyphosate (98.4% purity) of 12,680 µg/ml in combination with exposure to 40 or 50 mM H_2O_2. Activation by S9 was not used in this experiment. According to the authors, cell viability at this exposure level was above 80%.

Koller et al. (2012)[124] significantly induced DNA damage (comet assay) in human TR146 cells (buccal carcinoma cells) from exposure to glyphosate (>95% purity) in a dose-dependent fashion at concentrations of 20 and 40 µg/ml. Above 40 µg/ml, there was a significant increase in tail intensity relative to controls, but the actual amount increased did not change as the dose increased (plateau). Using Roundup (Ultra Max) the authors saw virtually the same level of DNA damage at 20 and 40 µg/ml, but the concentration response continued to increase above that exposure. These experiments did not use S9 activation. They also used the CBMN assay in the same system to evaluate the total number of micronuclei in binucleated cells (MNI), the number of binucleated cells with micronuclei (BN-MNI), the number of nuclear buds (NB) and the number of nucleoplasmic bridges (NPB) caused by glyphosate and Roundup exposure. Two endpoints (NB, NPB) had significant increases at concentrations of 10, 15 and 20 µg/ml and two (MNi, BN-MNi) were significantly elevated for concentrations of 15 and 20 µg/ml. Equivalent Roundup exposures resulted in significant increases in all four measures of DNA damage at 10, 15 and 20 µg/ml. The results for the Roundup were greater than for glyphosate alone.

Gasnier at al. (2009)[125] exposed cells from the hepatoma cell line HepG2 to glyphosate (purity not given) and four glyphosate formulations. Only one glyphosate formulation was tested for DNA damage (comet assay) and they saw significant effects at equivalent concentrations of 0.05 µ/ml to 4 µg/ml of glyphosate (p-values not given). No p-values are provided and presentation of the results does not provide a clear means to compare these results with other studies. This study will not be used in the evaluation.

Manas et al. (2009)[121] obtained human blood samples from three healthy, non-smoking women and three healthy men with no history of pesticide exposure. Lymphocytes were cultured with glyphosate (96% purity) at concentrations of 34, 203, and 1015 µg/ml with no statistically significant changes in chromatid breaks, chromosome breaks, chromatid gaps, chromosome gaps, dicentrics, acentric fragments, or endoreduplication.

Mladinic et al. (2009)[126] used blood from three non-smoking, healthy volunteers to evaluate the formation of micronuclei, nuclear buds and nucleoplasmic bridges as a function of exposure to glyphosate (98% purity). Significant changes in micronuclei were seen following exposure to glyphosate at 92.8 and 580 µg/ml in S9 activated cells, but not those without metabolic activation. Changes in nuclear buds were seen at 580 µg/ml for both S9 activated and non-activated cells while significant changes in nucleoplasmic bridges were seen only at 580 µg/ml in S9 activated cells. This study contained a positive control (ethyl methanesulfonate at 200 µg/ml) which was also

negative in all assays, many times showing effects below that seen for glyphosate.

Bolognesi et al. (1997)[127] obtained blood from two healthy female donors and exposed it to glyphosate (99.9% purity) or a Roundup formulation (30.4% glyphosate). At concentrations of 1000, 3000 and 6000 µg/ml of glyphosate and at 100 and 330 µg/ml of glyphosate formulation, significant changes in sister chromatid exchanges (SCEs) were seen. At 330 µg/ml, a non-significant increase in SCEs was seen for glyphosate alone that was approximately 20% below that seen for an equivalent glyphosate exposure from the Roundup formulation. This study did not consider S9 activation.

Lioi et al. (1998)[121, 128] obtained blood from three healthy donors and exposed it to glyphosate (>98% purity). At concentrations of 1.4, 2.9, and 8.7 µg/ml of glyphosate, significant changes in sister chromatid exchanges (SCEs) and chromosomal aberrations were seen. This study did not consider S9 activation.

Vigfusson and Vyse (1980)[129] exposed cultured human lymphocytes from two people to Roundup (% glyphosate unknown) at concentrations of 250, 2500 and 25000 µg/ml. Results for the highest concentration were not provided due to lack of cell growth in culture. SCEs were shown to be significantly increased for the remaining two concentrations in one donor and only for the lowest concentration in the other. While the relative SCE counts seen in this paper are similar to those from **Bolognesi et al. (1997),** the absolute counts in the controls are roughly three times higher in this study. This study did not consider S9 activation.

Genotoxicity in Non-Human Mammals (*in vivo*)

Bolognesi et al. (1997)[127] exposed groups of three Swiss CD-1 male mice by Intraperitoneal (IP) injection with a single dose of glyphosate (99.9% purity, 300 mg/kg) or Roundup (900 mg/kg, equivalent to 270 mg/kg glyphosate). Animals were sacrificed at four and 24 hours after injection and livers and kidney were removed to obtain crude nuclei from the adhering tissues. Both tissues demonstrated significant increases in DNA single-strand breaks ($p<0.05$) at four hours for both glyphosate and Roundup with no discernable difference between the responses. At 24 hours, the presence of strand breaks was reduced and no longer statistically significant from controls.

Peluso et al. (1998)[130] exposed groups of six (controls, lowest doses of glyphosate-salt and Roundup) or three Swiss CD-1 mice (males and females, specific numbers not specified, liver and kidney tissues combined for analysis) to the isopropylammonium salt of glyphosate or Roundup (30.4% isopropylammonium salt of glyphosate) for 24 hours. DNA adducts (^{32}P-DNA post labeling) were not evident in mice exposed to the glyphosate-salt alone in either liver or kidney, but were present in liver and kidney at all tested doses of Roundup showing a dose-response pattern.

Rank et al. (1993)[131] exposed male and female NMRI mice (three to five per sex) to glyphosate isopropylamine salt (purity not specified) and Roundup (480 g glyphosate isopropylamine salt per liter) by intraperitoneal injection. After 24 or 48 hours (only 24 hours for Roundup), polychromatic erythrocytes from bone marrow were extracted and micronuclei counted from a sample of 1000 cells. No significant increases were seen for

any concentration in glyphosate-exposed animals (100, 150 and 200 mg/kg) or Roundup-exposed animals (133 and 200 mg/kg glyphosate equivalent dose). The positive controls, while not statistically significant, showed an increase in micronuclei.

Bolognesi et al (1997)[127] exposed groups of three, four or six male Swiss CD-1 mice to glyphosate (99.9% purity) and Roundup (30.4% glyphosate) by intraperitoneal injection in two equal doses given 24 hours apart. After six or 24 hours following the last exposure, polychromatic erythrocytes from bone marrow were extracted and micronuclei counted from a sample of 1000 cells. Mice given two doses of 150 mg/kg of glyphosate showed a non-significant increase in micronuclei at 6 hours and a significant increase at 24 hours. In contrast, mice given two doses of 225 mg/kg glyphosate equivalent of Roundup showed a significant increase in micronuclei at both six and 24 hours. The relative differences in mean absolute increase (subtract mean response in controls) in micronucleii between glyphosate and Roundup at 24 hours was 3.6 whereas the relative difference in glyphosate equivalent dose was 1.5 indicating a greater effect of the glyphosate formulation.

Manas et al. (2009)[121] exposed groups of male and female Balb C mice (group size not given, tissues combined for analysis) to glyphosate (96% purity) by intraperitoneal injection in two equal doses given 24 hours apart. Twenty-four hours post exposure, polychromatic erythrocytes from bone marrow were extracted and micronuclei counted from a sample of 1000 cells. No significant increases were seen at doses of 50 mg/kg and 100 mg/kg in glyphosate-exposed animals but a significant increase was seen at 400 mg/kg. The positive controls showed a statistically significant increase in micronuclei (roughly three times the control rate).

Dimitrov et al. (2006)[132] exposed groups of eight male C57BL mice (tissues combined for analysis) to Roundup (41% glyphosate) via gavage at a dose of 1080 mg/kg. At 6, 24, 72, 96, or 120 hours post exposure, polychromatic erythrocytes from bone marrow were extracted and micronuclei counted from a sample of 4000 cells (500 per animal). No significant increases were seen. They also looked for chromosomal damage in these animals and saw no significant increases. The positive controls showed a statistically significant increase in micronuclei.

Prasad et al. (2009)[133] exposed groups of 15 male Swiss CD-1 mice to Roundup (30.4% glyphosate) by IP injection at doses of 25 and 50 mg/kg. At 24, 48 or 72 hours post exposure, polychromatic erythrocytes from bone marrow were extracted and micronuclei counted from a sample of 2000 cells per animal, five animals per sacrifice. Micronucleii counts were significantly increased ($p<0.05$) at all doses at all times relative to controls. In addition, the number of cells with chromosomal aberrations was significantly increased for all doses at all times. The control rate of micronuclei was similar to that of **Bolognesi et al. (1997),** but about 50% greater response for a dose that was approximately 10 times smaller.

Grisolia et al. (2002)[134] exposed groups of Swiss mice (sex and sample size not given) to Roundup (480 g glyphosate isopropylamine salt per liter) by IP injection at doses of 50, 100 and 200 mg/kg Roundup in two doses separated by 24 hours. At 24 hours post

exposure, polychromatic erythrocytes from bone marrow were extracted and micronuclei counted from a sample of 2000 cells per animal. Micronuclei counts were not increased at any dose. This exposure appears to be the same formulation of Roundup used in the study by **Rank et al. (1993)** which was also negative.

Coutinho do Nascimento and Grisolia (2000)[135] exposed groups of six male mice (strain not given) to Roundup (% glyphosate not given) by IP injection at doses of 50, 100 and 200 mg/kg in two doses separated by 24 hours. At 24 hours post exposure, polychromatic erythrocytes from bone marrow were extracted and micronuclei counted from a sample of 1000 cells per animal. A significant increase in micronuclei were seen at a dose of 85 mg/kg. No increase was seen at 42 or 170 mg/kg.

Cavusoglu et al. (2011)[136] exposed groups of six Swiss albino mice by IP injection with a single dose of glyphosate formulation (RoundupUltra Max, 450 g/l glyphosate, 50 mg/kg glyphosate equivalent dose). Animals were sacrificed at three days after injection. Micronuclei in normochromatic erythrocytes were counted from a sample of 1000 cells per animal. There was a significant increase in micronuclei in erythrocytes ($p<0.05$). *G. bilboa* eliminated these effects.

Chan and Mahler (1992)[137] exposed groups of 10 male and female $B6C3F_1$ mice to glyphosate (98.6% purity) in feed at doses of 0, 507, 1065, 2273, 4776, and 10780 mg/kg in males and 0, 753, 1411, 2707, 5846, and 11977 mg/kg in females for 13 weeks. At sacrifice, polychromatic erythrocytes from peripheral blood were extracted and micronuclei counted from a sample of 10,000 cells. No significant increases were seen at any of the tested doses.

Li and Long (1988)[138] exposed groups of 18 male and female Sprague-Dawley rats to glyphosate (98% purity) by IP injection at a dose of 1000 mg/kg. At 6, 12 and 24 hours post treatment, 6 animals of each sex were sacrificed and polychromatic erythrocytes from bone marrow were extracted and micronuclei counted from a sample of 50 cells per animal. The percentage of cells with chromosomal aberrations was not increased at any time point following exposure.

Genotoxicity in Non-Human Mammalian Cells (*in vitro*)

Li and Long (1988)[138] incubated Chinese hamster ovary cells (CHO-K1BH4) with glyphosate (98% purity) for three hours at concentrations of 5, 10, 50 and 100 mg/ml. Cells were then plated using 200 cells per sample in triplicate and incubated for 8-12 days. Colonies were then counted and results expressed as mutant frequency. No positive results were seen in any experimental group with or without S9 activation. It is not clear why there is such a large difference in the incubation times in the various groups in this experiment, nor is it clear which groups incubated longer. In a second study in the same publication, non-induced primary rat hepatocytes (Fischer 344) were incubated with seven concentrations of glyphosate (12.5 ng/ml to 125 µg/ml) for 18-20 hours. No significant increases were seen for net grains per nucleus at any exposure concentration. There was a four-fold increase in the lowest exposure groups relative to controls and then every other treated group was below the control response. This is a

very unusual finding and could be due to the way in which the data is adjusted for net grains in cytoplasm. The authors calculated net grains per nucleus by subtracting the highest cytoplasmic count from the nuclear count; if cytoplasmic count is increased by glyphosate this could bias the findings making any increase in nuclear count disappear. No data is provided to resolve this issue.

Roustan et al. (2014)[139] incubated Chinese hamster ovary cells (CHO-K1) with glyphosate (purity not provided) for three hours at concentrations of 2, 5, 10, 15, 17.5, 20, and 22.5 mg/ml. Cells were then plated using 200 cells per sample in triplicate and incubated for 24 hours. For each exposure concentration, 2000 bi-nucleated cells were examined for micronuclei. No positive results were seen in any experimental group without S9 activation but the four highest exposure groups were significant with a clear concentration-response pattern when S9 activation was present.

Lioi et al. (1998)[128] exposed lymphocytes from three unrelated healthy cows to glyphosate (>98% purity) for 72 hours to concentrations of 3, 14.4 and 28.7 µg/ml without S9 activation. Chromosomal aberrations scored from 150 cells were significantly increased ($P<0.05$) for all exposure concentrations of glyphosate with a clear concentration-response pattern. Similarly, SCEs per cell were increased at all concentrations ($p<0.05$) but no concentration response pattern was evident.

Sivikova and Dianovsky (2006)[140] exposed lymphocytes from two healthy young bovine bulls to glyphosate formulation (62% glyphosate) for 2, 24 and 48 hours using concentrations of 4.7, 9.5, 23.6, 47.3, 94.6 and 190 µg/ml without S9 activation. Chromosomal aberrations scored from 100 cells were not significantly increased ($P<0.05$) without S9 activation for any 24-hour exposure concentration of glyphosate (2- and 48-hours exposures were not done). SCEs per cell were increased at all 24-hour exposure concentrations ($p<0.05$) except the lowest concentration. At 48-hours, significant increases of SCEs per cell were seen at concentrations at or above 47.3 µg/ml (2-hour exposures were not done). Finally, after two hours of exposure with S9 activation, significant effects were seen at 5 and 10 µg/ml but not at 15 µg/ml (24- and 48-hour exposures were not done for S9 activation).

Holeckova (2006)[141] exposed lymphocytes from two healthy young bovine bulls to glyphosate formulation (62% glyphosate) for 24 hours to concentrations ranging from 28 to 1120 µmol/L without S9 activation. A significant increase in polyploidy was observed at 56 µmol/L, all other comparisons were without significance. However, this one finding cannot be easily dismissed because all exposure groups above this concentration had too few cells for evaluation. This study did not consider S9 activation.

Genotoxicity in Non-Human Systems (*in vivo* and *in vitro*)

Four studies[120, 142-144] in fish have seen positive results for genotoxicity (DNA strand breaks, different assays) following exposure to glyphosate. In addition, one study[145] in oyster sperm and embryos exposed to glyphosate saw no increase in DNA damage (comet assay) and one study[146] in two strains of Drosophila melanogaster showed an increase in mutations (wing spot test) at the higher doses of exposure.

Fourteen studies[134, 142, 144, 147-157] in multiple fish species evaluated the relationship between various glyphosate formulations and genotoxicity with all studies showing positive results for various endpoints (DNA strand breaks, micronucleus formation, and chromosomal aberrations). Two of the studies[147, 149] were negative for micronucleus formation after exposure to glyphosate formulations and one of these[147] was also negative for chromosomal aberrations but both were positive in other markers of genotoxicity. Two studies[158, 159] demonstrated genotoxicity (DNA strand breaks, micronuclei) in caiman from *in-vivo* exposure to a glyphosate formulation. Three studies[160-162] demonstrated genotoxicity (DNA strand breaks, micronucleus formation) in frogs or tadpoles from exposure to glyphosate formulations. One study[145] in oyster sperm and embryos, one study[163] in clams and one study[164] in mussels exposed to a glyphosate formulation saw no increase in DNA damage (comet assay). One study[165] in snails saw increased DNA damage (comet assay) following exposure to a glyphosate formulation. Two studies[166, 167] in worms saw mixed results for DNA damage (comet assay) with one of these studies[166] showing a positive result for micronucleus formation. One study[168] in Drosophila melanogaster showed an increase in sex-linked recessive lethal mutations.

In the published literature, five studies evaluated the impact of glyphosate in *in vitro* systems. Two of these studies[169, 170] looked at genotoxicity of glyphosate in combination with UVB radiation and saw significant increases in DNA strand breaks (FADU assay) in bacteria without metabolic activation. One study[171] in eukaryote fish saw a significant increase in DNA strand breaks (comet assay) without S9 activation. Another study[138] showed no increase in reverse mutations in two strains of bacteria with and without S9 activation.

Williams et al. (2000)[172] summarized the literature regarding the use of reverse mutation assays in *S. typhimurium* (Ames Test). Four studies using glyphosate and five studies of glyphosate formulations were all negative. They cited one study[131] of a glyphosate formulation that was positive with S9 activation and negative without S9 activation. However, this study was positive with S9 activation in TA100 cells, negative with S9 activation in TA98 cells, negative without S9 activation for TA100 cells and positive without activation for TA98 cells. They also summarized two studies of glyphosate in *e. coli* that were negative with and without activation.

Two additional studies[138, 173] of glyphosate using reverse mutation assays are available from the scientific literature, both of which are negative.

Regulatory Studies

EFSA[88] cited 14 reverse mutation assays in *S. typhimurium* (Ames Test), most of which were tested in strains TA 98, 100, 1535, 1537 (Table B.6.4-1). All 14 studies are listed as negative by EFSA. Actual data is provided for only one of the 14 studies and this study is clearly negative. **EPA**[61] cited 27 reverse mutation assays in *S. typhimurium* (Ames Test), most of which were tested in strains TA 98, 100, 1535, 1537 (EPA Table 5.1). All 27 studies are listed as negative. No data is provided for any of the studies. **Kier and**

Kirkland (2013)[174] cited results from 18 bacterial reverse mutation assays of glyphosate and 16 of glyphosate formulations. Tabulated results and background information were provided for all 34 studies. Six studies of glyphosate alone demonstrated positive findings in one or more groups.

EFSA[88] cites three studies of gene mutations in mammalian cells, all of which are listed as negative (EFSA Table B.6.4-5), two use the mouse lymphoma assay, and one uses the Chinese hamster ovary cell/hypoxanthine-guanine phosphoribosyl transferase (CHO/HGPRT) mutation assay. **EPA**[61] cites four studies, three of which appear to be the same as those cited by EFSA (EPA Table 5.2) and the fourth is another mouse lymphoma assay. All four are listed as negative. **Kier and Kirkland (2013)**[174] cite two of the mouse lymphoma studies and provide tabulated data. Neither study shows any indication of a statistically significant increase in mutation frequency at the thymidine kinase locus of L5178 mouse lymphoma tk(+/-) cells.

EFSA[88] cites one *in vitro* study of DNA damage and repair in mammalian cells which is listed as negative (EFSA Table B.6.4-6). This study is of unscheduled DNA synthesis (UDS assay) in primary rat lymphocytes. They also list five studies of chromosome aberrations (EFSA Table B.6.4-8), which are characterized as negative. Two studies are in human lymphocytes and two are in Chinese hamster lung (CHL) cells. Data for one of the studies in CHL is provided in tabular form and is clearly negative. **EPA**[61] cites eight *in vitro* studies of chromosome aberrations in mammalian cells (EPA Table 5.3); two of these studies match studies in the EFSA report. Four of the studies are from the literature[121, 128, 140, 175] and are reviewed above. Surprisingly, EPA refers to the study by **Manas et al. (2009)**[121] as negative although it was clearly positive in the comet assay., Additionally, EPA refers to the study by **Sivikova and Dainovsky (2006)**[140] as negative even though they saw clear effects of glyphosate on SCEs. Basically, all four of the literature studies cited by EPA are positive yet EPA lists only two of the four as positive. The remaining four studies are noted as negative; however, no data is supplied for these studies. **Kier and Kirkland (2013)**[174] cites eight literature studies (all reviewed above) and three regulatory studies with glyphosate exposure. The three regulatory studies are listed as negative, and the data are available as a table in the supplement material to **Kier and Kirkland (2013)**; these studies are negative at all tested concentrations in CHL cells; one matches the study data provided by EFSA[88].

EFSA[88] cites nine micronucleus assays, three in Swiss Albino mice, two in NMRI mice, two in CD-1 mice, one in Sprague-Dawley rats, and one in CD rats (EFSA Table B.6.4-12). They list one study in Swiss Albino mice as weakly positive in males, one study in CD-1 mice as positive at the highest dose (data for this study is provided) and all other studies as negative. They discard one study with low doses in male Swiss mice, but the tables provided for this study show a clearly significant result at the highest dose used (30 mg/kg) and clear dose-response. They provide data for two of the negative studies which indicate these studies were indeed negative. **EPA**[61] (EPA Table 5.5) cites 20 micronucleus assays, four are available in the scientific literature and three are reviewed above (the fourth reference[176] was unavailable to me at the time of preparation of this report). The remaining 16 studies include six studies in Swiss Albino mice, four studies

in CD-1 mice, three studies in NMRI mice, two studies in Sprague-Dawley rats and one study in Wistar rats. Since EFSA does not provide names associated with their micronucleus studies, I cannot determine if any of the studies cited by the EPA are the same as those cited by EFSA. EPA lists two of the literature studies as positive and two as negative (matching my reviews for the three studies I have access to) and all but one of the regulatory studies as negative (the one positive study was in Swiss-Albino mice). **Kier and Kirkland (2013)**[174] cite 12 regulatory micronucleus assays of glyphosate and provide data tables for all 12. All 12 of these studies are cited by EPA. **Kier and Kirkland (2013)** list 11 studies as negative and one as inconclusive. However, four of the studies show positive effects in at least one sex-by-treatment group. One of these four studies they list as inconclusive and the remaining three studies are determined to be negative because the response is within the range of the historical controls. As was discussed for the animal carcinogenicity studies, the correct group to use is the concurrent control. **Kier and Kirkland (2013)**[174] also cite 12 regulatory studies and three literature studies where animals are exposed to a glyphosate formulation. Two of the literature studies are reviewed above and the remaining study[176] was unavailable. Data for the 12 regulatory studies are all provided in tables by **Kier and Kirkland (2013)** and show two positive studies in CD-1 mice and negative studies for the remaining 10.

Summary for Genotoxicity

This is a complicated area from which to draw a conclusion due to the diversity of the studies available (there are multiple species, multiple strains within a species, multiple cell types from multiple species, differing lengths of exposure, differing times of evaluation after exposure, differing exposures, numerous markers of genotoxicity, and finally both glyphosate and multiple different glyphosate formulations). There are three studies that evaluate the genotoxicity of glyphosate in humans directly, 36 experiments in eight strains of mice, three studies in rats, nine studies in human lymphocytes and four studies in other human cells, 12 studies in non-human mammalian cell lines (two using mouse cells, five using hamster cells, two using rat cells and three using cells from cows), a large number of studies in a wide variety of non-mammalian species, and a plethora of studies, mostly identical, in bacteria.

Some conclusions are straightforward"; glyphosate does not appear to cause reverse mutations for histidine synthesis in *Salmonella typhimurium*, regardless of whether these reverse mutations are due to frameshift mutations or point mutations. I am cautious in this determination because there were several studies with positive results, but no clear pattern is evident. There is ample evidence supporting the conclusion that glyphosate formulations and glyphosate can cause genotoxicity in non-mammalian animal species. This clearly indicates that both glyphosate and the formulations are able to cause injury to DNA. So while findings of genotoxicity in these species do not speak directly to the hazard potential in humans, they do support a cause for concern.

The more important studies are those that have been done using mammalian systems, human cells and direct human contact. Table 16 summarizes these studies in a simple framework that allows all of the experimental data to be seen in one glance. This table does not address the subtlety needed to interpret any one study, but simply

demonstrates when a study produced positive versus negative results.

Clearly, for *in vitro* evaluations in human cells, the majority of the studies have produced positive results. There was only one regulatory study evaluating glyphosate genotoxicity in human lymphocytes from healthy volunteers and that study was negative. The study was not significantly different from the other six studies in this category, five of which produced positive results. The majority of these studies used either the comet assay (a simple way for measuring any type of DNA strand break) or methods that counted specific types of strand breaks in the cells (e.g. SCEs, micronuclei, nuclear buds and nucleoplasmic bridges). From these assays, we can conclude there is DNA damage. For glyphosate formulations, there are only three studies in humans *in vivo*, two of which were positive.

The magnitude of the concentrations used in these studies could potentially lead to false positives if the glyphosate is causing cytotoxicity in the cells. All six studies using the comet assay were positive with no study showing a negative response below 10 µg/ml and mixed results below that with positive results at 0.12 and 3.5 µg/ml and negative results at 2.91 and 10 µg/ml. In general, the comet assays provide strong support for genotoxicity.

The four studies that directly addressed specific types of strand breaks in cells following exposure to glyphosate showed markedly different responses across the various concentrations used. **Manas et al. (2009)** saw no changes in chromatid breaks, chromosome breaks, chromatid gaps, chromosome gaps, dicentrics, acentric fragments or endoreduplication over the range of concentrations 3.4-1015 µg/ml. In contrast, **Lioi et al. (1998)** saw changes in SCEs over concentrations ranging from 1.4 to 8.7 µg/ml. Both studies were done in lymphocytes from volunteers. **Mladinic et al. (2009)** saw significant changes in micronuclei above 92.8 µg/ml and **Bolognesi et al. (1997)** saw positive changes in SCEs above 1000 µg/ml but not at 330 µg/ml. While changes have been seen in three of the four studies, the actual concentrations in which the changes are seen is not consistent across studies. I conclude that glyphosate causes DNA strand breaks, which is indicative of genotoxicity.

The micronucleus assays in rodents examining glyphosate genotoxicity are either all positive in one strain or all negative in one strain with the exception of the three studies in CD-1 mice and four studies in Swiss Albino mice. For the positive studies, we can ask the question of whether, in this strain, the actual number of micronuclei are consistent.

Table 17: Summary of *in vivo* and *in vitro* genotoxicity studies of glyphosate and glyphosate formulations in mammals[1]

In vivo or *in vitro*	Species	Cell type or tissue	Glyphosate[2]		Glyphosate Formulations	
			Number Positive	Number Negative	Number Positive	Number Negative
In vivo	Humans	Peripheral blood			2	1
in vitro	Humans	lymphocytes	5	2(1)	2	
		Hep 2	1			
		GM 38 HT1080	1			
		GM 5757	1			
		TR146	1		1	
In vivo	Swiss CD-1 Mouse	Liver/Kidney	1	1	2	
In vivo (micro-nucleus assay)	NMRI mouse	Erythrocytes		4(3)		2(1)
	Swiss CD-1 mouse		1		2	
	Balb C mouse		1			
	B6C3F$_1$ mouse			1		
	Swiss mouse		1(1)			3(2)
	CD-1 mouse		2(2)	1(1)	2 (2)	6 (6)
	Swiss albino mouse		1(1)	3(3)	1	
	C57BL mouse					1
	Mouse (not specified)				1	
	Rats (all)			2(1)		1(1)
In vitro	Mouse	L5178 lymphoma		2(2)		
	Chinese hamster	Lung		3(3)		
	Chinese hamster	ovary	1	1		
	Fischer rat	liver		1		
	Rat	Lymphocytes		1(1)		
	Bovine	Lymphocytes	1		2	

[1]each entry in the table corresponds to a single study where a study is positive if at least one valid positive finding emerged from the study $p<0.05$; entries in the table are only for studies where data was available to review including data from EFSA[88] and Kier and Kirkland (2000)[174]; [2]numbers are the total number of studies in this category, numbers in parentheses are the subset of studies that are regulatory studies

In Swiss Albino mice, all four studies were done with males and females. Exposures were by oral gavage for the positive study (in female mice) and IP injection by the negative studies. The positive study was at 5000 mg/kg and the highest dose in any of the negative studies was 3024 mg/kg. Finally, the control response in the positive study was 6.7 micronucleated PCE per 1000 PCE whereas the controls in the three negative studies were between 0 and 0.6 micronucleated PCE per 1000 PCE. Any of these differences could easily explain the differences in response so the positive result in Swiss Albino mice should be accepted.

For CD-1 mice, the one negative micronucleus study was by oral gavage in males and females at a single dose of 5000 mg/kg. One of the positive studies was also by oral gavage in males at a single dose of 2000 mg/kg. Because of the nature of statistical noise, these two studies could both occur whether there is a true effect or not. For the other positive study, the dose was by IP injection in male mice with a positive response at 600 mg/kg that was more than double the response of the controls. These data support the finding that glyphosate can cause micronuclei in male CD-1 mice, which is indicative of genotoxicity.

The remaining *in vitro* assays in mammalian cells exposed to glyphosate show mixed results. The mouse lymphoma assay and the Chinese hamster ovary assays are looking for specific mutations that will allow these cells to grow in culture. The Chinese hamster lung, the two rat assays and the assay in bovine lymphocytes are measuring DNA damage and provide mixed results. In general, these responses appear to be negative with the exception of those seen in bovine lymphocytes that appear to show a positive increase in SCEs following exposure to glyphosate.

For glyphosate formulations, the main difference between the findings for glyphosate and those for the glyphosate formulations is the direct evidence for genotoxicity in humans and the micronucleus assays in Swiss mice. The observation of genotoxicity in humans following exposure to glyphosate formulations must carry the greatest weight in the overall analysis and two of the three studies were positive with the strongest study by **Bolognesi et al. (2009)**[117] showing the strongest response.

For the Swiss mouse studies of micronuclei, the fact that all three studies are negative for glyphosate formulations while one study is positive for glyphosate creates a clear disagreement. The positive study is an oral gavage study with an effect seen in male mice at 30 mg/kg/day. The two negative regulatory studies for glyphosate formulations were done at 2000 mg/kg (about 500 mg/kg glyphosate equivalent), were also oral gavage studies and were replicates done in the same laboratory at different times. The remaining negative study used glyphosate formulation doses of 50-200 mg/kg (25-100 mg/kg glyphosate equivalent) but was done by intraperitoneal injection. With the exception of the different routes of exposure, the differences between these studies cannot be resolved.

In this case, a pooled analysis of the data is not possible because in almost every case, no one study is a clear replicate of another. Instead, the appropriate approach would be to do a meta-analysis and evaluate which aspects of the experimental designs are

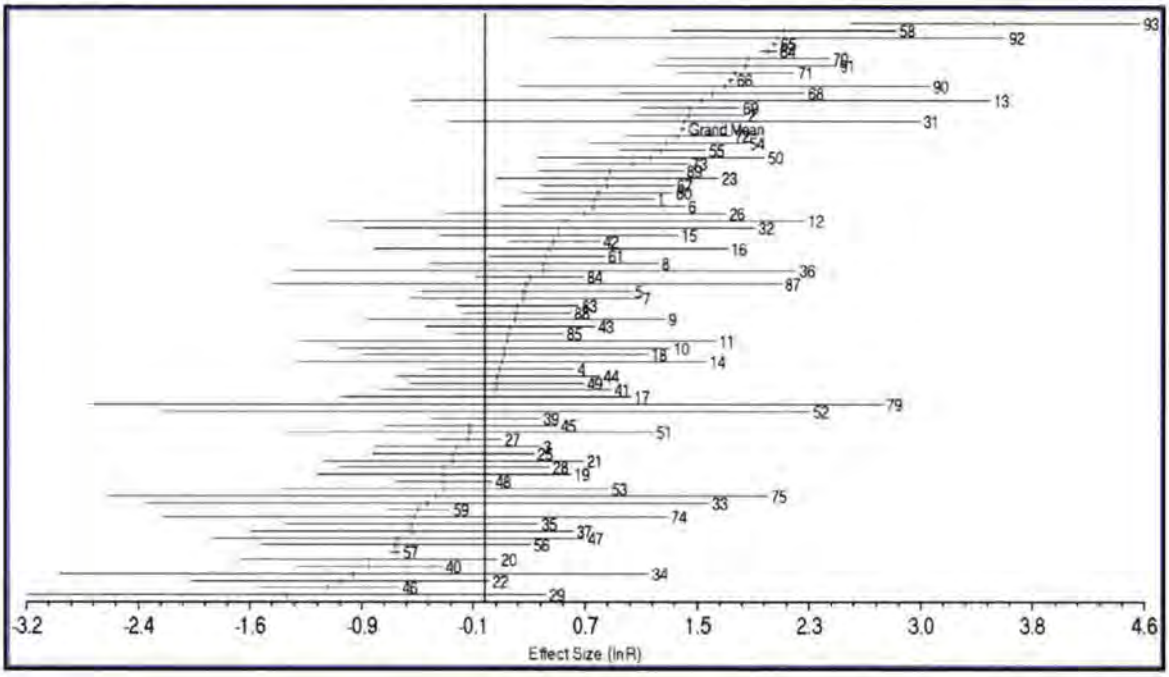
important to producing positive findings of genotoxicity. The studies with the most data for this type of analysis are the various *in vivo* assays of micronucleus formation. **Ghisi et al. (2016)**[177] did a systematic search to identify all published studies evaluating the ability of glyphosate or glyphosate formulations to induce micronuclei *in vivo*. The authors also used the data from **Kier and Kirkland (2013)**[174] summarized above. An experiment, in their evaluation, was defined by sex/species/route/form of glyphosate so that some studies doing both sexes using glyphosate and a glyphosate formulation will enter multiple times into the analysis. They identified 93 experiments from which it was possible to do a meta-analysis. Data were extracted for each study and the log ratio of the mean of each experimental group to the mean control response (E+) was used to evaluate effect sizes in the meta-analysis. For this meta-analytic mean, a value below zero suggests no genotoxicity while a value above zero suggests increased genotoxicity. A test of heterogeneity (Cochran's Q statistic discussed earlier for the epidemiological data) was also evaluated.

Figure 2 is a reprint of Figure 1 from the study by **Ghisi et al. (2016)**[177] and is a forest plot from all studies they evaluated for glyphosate and glyphosate formulations. It is clear from this plot that the predominant response is positive in these data with an overall grand mean response across all studies of E+=1.37 and a 95% confidence interval of (1.356-1.381) (this is highly statistically significant with a $p<0.0001$). The Qt value for the grand mean was also statistically significant suggesting there are other explanatory variables in the data that would help to explain the overall variance.

Categorical variables were then used to make comparisons across the various strata in the data to identify which experimental conditions show the largest impacts on the mean response. Mammalian species presented a higher mean effect (E+=1.379; 1.366-1.391) than non-mammalian species (E+=0.740; 0.641-0.840). Glyphosate formulations showed a greater mean response (E+=1.388; 1.375-1.400) than did glyphosate (E+=0.121; 0.021-0.221), but both were significantly greater than zero. The mean response in studies using only male animals (E+=1.833; 1.819-1.847) was significantly different from zero as were studies using both males and females (E+=0.674;0.523-0.825) whereas the mean response in studies using only females (E+0.088; -0.153-0.328) was not. Peer-reviewed studies had higher mean response (E+=1.394; 1.381-1.407) compared to regulatory studies (E+=0.114; 0.027-0.202), but both means were significantly greater than zero, indicating an overall genotoxic effect. Other variables were examined such as length of exposure and magnitude of exposure that had very little impact on the overall findings.

The meta-analysis by **Ghisi et al. (2016)**[177] provides strong support for the hypothesis that exposure to glyphosate and glyphosate formulations increases the formation of micronuclei *in vivo*. This means that glyphosate and glyphosate formulations are damaging DNA in living, functioning organisms with intact DNA repair capacity strengthening the finding that glyphosate is genotoxic to humans.

Figure 2: Forest plot of studies evaluating micronucleus frequency in glyphosate exposure, arranged by effects size. The plot shows the estimate of the response ratio and 95% confidence interval (CI) of each experiment included in the meta-analysis. The number beside the bars represents the reference number of each experiment as in Table 1 of Ghisi et al. (2016)[177]. Grand Mean is the overall mean effects size of all studies. [Reprinted from Ghisi et al. (2016)[177]]



From a simply statistical perspective, there is another way in which one can decide if the positive findings in the micronucleus assays in the mice are due to chance. For the glyphosate studies, if one adds up all of the individual experimental groups, there are 79 total groups which correspond to 79 statistical tests. Assuming the critical testing level is 0.05 for all of the tests, one would expect to see just under four positive findings, yet six are observed. For the glyphosate formulations, there were 70 experimental groups so one expects 3.5 positive findings yet 12 are observed ($p<0.01$). Overall, there were a total of 149 experimental groups examined in mice for micronucleus formation and we observed 18 (7.5 expected, $p<0.01$). Repeating this analysis on the basis of studies instead of experimental groups, there were 15 studies for glyphosate (expected number is 0.75 positive) yet six positive were observed ($p<0.01$). For the glyphosate formulations, there were 18 studies (expected number is 0.9 positive) yet six positive are observed ($p<0.01$). Now expanding to all 69 studies presented in Table 17, there were 33 positive studies, but the expectation is a mere 3.5 ($p<0.01$).

It is clear that both glyphosate and glyphosate formulations have genotoxic potential. But which is worse? Of the 69 experiments in Table 17, there were eight experiments from five research publications that addressed both glyphosate and a glyphosate formulation in the same laboratory. Of these, two were negative for both glyphosate and the formulation and do not contribute to a discussion of relative potency. The

remaining six can provide some guidance on the relative potency of glyphosate to glyphosate formulations. In **Koller et al. (2007)**[124], tail intensity for the comet assay were virtually identical when the amount of glyphosate in the formulation was compared to the results using glyphosate alone. In the same paper, micronuclei and related biomarkers were consistently higher in the glyphosate formulation by 10-20%. In **Bolognesi et al. (1997),** DNA strand breaks in liver and kidney in Swiss CD-1 mice were virtually identical under equivalent doses of glyphosate and glyphosate formulations. In their micronucleus assay, the glyphosate formulation was approximately 50% more potent. Finally, **Bolognesi et al. (1997),** in their analysis of SCEs in human lymphocytes, the glyphosate formulation was approximately twice as effective as glyphosate alone. In **Peluso et al. (1988)**[130], DNA adducts in livers and kidneys were only seen in mice treated with the glyphosate formulation, so these findings are not likely to be due to glyphosate. The data suggest a small increase in the potential for genotoxicity for glyphosate formulations relative to the genotoxicity one would see with glyphosate alone.

In summary, the data support a conclusion that both glyphosate and glyphosate formulations are genotoxic. Thus, there is a reasonable mechanism supporting the increases in tumors caused by glyphosate and glyphosate formulations in humans and animals.

Oxidative Stress

Oxidative stress refers to an imbalance between the production of reactive oxygen species (free radicals) in a cell and the antioxidant defenses the cell has in place to prevent this. Oxidative stress has been linked to both the causes and consequences of several diseases[178-183] including cancer[37, 184-188]. Multiple biomarkers exist for oxidative stress; the most common being the increased antioxidant enzyme activity, depletion of glutathione or increases in lipid peroxidation. In addition, many studies evaluating oxidative stress used antioxidants following exposure to glyphosate to demonstrate that the effect of the oxidative stress can be diminished.

Oxidative Stress in Human Cells (*in vitro*)

Mladinic et al. (2009)[119] examined the induction of oxidative stress from exposure to glyphosate (98% purity) in lymphocytes from three healthy human donors (questionnaires were used to exclude other genotoxic exposures) at concentrations of 0.5, 2.91, 3.5, 92.8 and 580 µg/ml. Cells with and without S9 activation saw increases in total antioxidant capacity at only the highest dose for cells without S9 activation although a clear concentration response pattern was seen with S9 activation.

Kwiatkowska et al. (2014)[189] examined the induction of oxidative stress from exposure to glyphosate (purity not given) in erythrocytes obtained from healthy donors in the Blood Bank of Lodz, Poland. Erythrocytes were exposed to concentrations of 1.7, 8.4, 17, 42.3, 85 and 845 µg/ml and incubated for 1 hour. Oxidative stress (oxidation of dihydrorhodamine 123) was significantly increased at 42.3, 85 and 845 µg/l with a clear concentration-response pattern.

Chaufan et al. (2014)[190] examined the induction of oxidative stress from exposure to glyphosate (95% purity) and Roundup UltraMax (74.7% glyphosate) in HepG2 cells (human hepatoma cell line). Exposure concentrations were 900 µg/ml for glyphosate and 40 µg/ml for the glyphosate formulation. After incubation for 24 hours, oxidative stress (expressed as the activity of superoxide dismutase (SOD), catalase (CAT), glutathione (GSH) and glutathione-S-transferase (GST)) was significantly increased ($p<0.0\text{-}5$) for the glyphosate formulation (increased SOD activity) but not for glyphosate alone.

Coalova et al. (2014)[191] examined the induction of oxidative stress from exposure to a glyphosate formulation (Atanor, 48% glyphosate) or with a surfactant (Impacto) in Hep-2 cells (human epithelial cell line). Exposure concentrations were 376.4 µg/ml for Atanor, 12.1 µg/ml for Impacto and 180.2 µg/ml for a mixture of the two. After incubation for 24 hours, oxidative stress (measured as activity of SOD, CAT, GSH, and GST) was significantly increased for Impacto, Atanor and the mixture (CAT and GSH only, $p<0.05$ or $p<0.01$).

Gehin et al. (2005)[192] examined the induction of oxidative stress from exposure to glyphosate (purity unknown) and a glyphosate formulation (Roundup 3 plus, 21% glyphosate) in HaCaT cells (human keratinocyte cell line). Glyphosate induced cytotoxicity in the cells which was reduced or eliminated by antioxidants. The authors attributed the cytotoxicity to oxidative stress.

Elie-Caille et al. (2010)[193] examined the induction of oxidative stress from exposure to glyphosate (purity unknown) in HaCaT cells (human keratinocyte cell line). Exposure concentrations ranged from 1700 µg/l to almost 12,000 µg/ml. Glyphosate induced cytotoxicity in the cells and increased hydrogen peroxide H_2O_2 (dichlorodihydrofluorescein diacetate assay). This study used exceptionally high concentrations that may be inducing cytotoxicity by means that are independent of the oxidative stress observed. Measuring oxidative stress using the dichlorodihydrofluorescein diacetate assay has limitations[194, 195].

George and Shukla (2013)[196] examined the induction of oxidative stress from exposure to a glyphosate formulation (Roundup Original, 41% glyphosate) in HaCaT cells (human keratinocyte cell line). Exposure concentration ranged from 1.7 µg/ml to 17,000 µg/ml and exposure was for 24 hours. Glyphosate significantly induced the formation of reactive oxygen species (dichlorodihydrofluorescein diacetate assay) at all exposures in a concentration-dependent fashion. Prior treatment of the cells with N-Acetylcysteine reduced the impact of glyphosate, but did not eliminate it. Measuring oxidative stress using dichlorodihydrofluorescein diacetate has limitations[194, 195] that affect the clear interpretation of these results.

Oxidative Stress in Non-Human Mammals (*in vivo*)

Bolognesi et al. (1997)[127] exposed groups of three Swiss CD-1 male mice by IP injection with a single dose of glyphosate (99.9% purity, 300 mg/kg) or Roundup (900 mg/kg, equivalent to 270 mg/kg glyphosate). Animals were sacrificed at eight and 24 hours after injection and livers and kidney were removed to obtain crude nuclei from the

adhering tissues. Samples of liver and kidneys from these mice were evaluated for levels of 8-hydroxy-2'-deoxyguanosine (8-OHdG) which is a biomarker of oxidative stress[197]. There was a significant increase in the liver of 8-OHdG at 24 hours following glyphosate exposure, but not at eight hours and not in the kidney. At both eight hours and 24 hours, Roundup increased 8-OHdG in the kidneys, but the mild increase seen in the liver at 24 hours was not significant.

Cavusoglu et al. (2011)[136] exposed groups of six Swiss albino mice by IP injection of a glyphosate formulation (RoundupUltra Max, 450 g/l glyphosate, 50 mg/kg formulation). At the end of dosing, animals were fasted overnight then sacrificed. There was a significant increase in malondialdehyde in both liver and kidney and a significant decrease in GSH in liver and kidney from exposure to the glyphosate formulation. *G. bilboa* eliminated these effects.

Jasper et al. (2012)[198] exposed groups of 10 male and 10 female Swiss albino mice via oral gavage for 15 days to a glyphosate formulation (Roundup Original, 41% glyphosate, 50 mg/kg glyphosate equivalent dose). Animals were sacrificed at three days after injection. There was a significant increase in thiobarbituric acid-reactive substances (TBARS) in the liver for both male and female mice at both doses ($p<0.05$). The concentration of non-protein thiols was elevated in both dose groups for males and for the high dose only in females (no dose-response was seen for this endpoint).

Astiz et al. (2009)[199] exposed groups of four male Wistar rats by IP injection to a single dose of glyphosate (purity unknown, 10 mg/kg). Animals were injected three times per week for five weeks and then sacrificed. Thiobarbituric acid-reactive substances (TBARS assay), protein carbonyls (PCOSs), total glutathione levels, individual glutathione levels, SOD and CAT were all measured as biomarkers for oxidative stress in plasma, brain, liver and kidney. Glyphosate significantly increased TBARS in all tissues ($p<0.01$), total glutathione in brain ($p<0.01$), SOD in liver and brain ($p<0.01$) and CAT in brain. In a follow-up report[200], they demonstrate that lipoic acid eliminates or severely reduces the impacts of glyphosate on the brain.

Cattani et al. (2014)[201] exposed groups of four pregnant Wistar rats to glyphosate formulation (Roundup Original, 360 g/L glyphosate) in drinking water from gestational days 5-15 at a dose of 71.4mg/kg. Fifteen day-old pups (2 per dam) were examined for oxidative stress markers in the hippocampus. Pups had a significant increase in TBARS ($p<0.05$) and a significant decrease in GSH ($p<0.01$).

George et al. (2010)[81] exposed groups of four Swiss albino mice to a glyphosate formulation (Roundup Original, 36g/L glyphosate) at a dose of 50 mg/kg (glyphosate equivalent dose) via a single topical application. Proteomic analysis of skin from the treated animals saw alterations in SOD1, CA III and PRX II, proteins known to play a role in the management of oxidative stress.

Oxidative Stress in Non-Mammalian Systems

As for genotoxicity, oxidative stress from exposure to glyphosate and glyphosate formulations have been studied in various aquatic organisms; reviewed in **Slaninova et**

al. (2009)[202]. Many of the studies reviewed by **Slaninova et al. (2009)** showed associations with glyphosate and oxidative stress in various organs. Since that review, additional studies have been completed that also demonstrate a positive association between glyphosate and oxidative stress[144, 153-156, 203-214].

Summary for Oxidative Stress

Seven studies addressed oxidative stress in human cells and another six studies addressed it in mammalian systems. In lymphocytes and erythrocytes from healthy donors, oxidative stress was detected as low as 580 µg/ml in lymphocytes and at 42.3 µg/ml in erythrocytes. In Hep-G2 cells, no increased oxidative stress was seen for a single concentration of 900 µg/l. In two studies in HaCat cells, glyphosate induced oxidative stress in a continuous model fit to the results in one study and at the lowest concentration (1700 µg/ml) in the other. The most convincing studies in human cells for oxidative stress are the two studies in human blood.

In Swiss CD-1 male mice, increased oxidative stress was seen in the liver at 24 hours, but not at four hours after injection of 300 mg/kg glyphosate. No increase was seen in the kidney. In Wistar rats, repeated IP dosing with glyphosate lead to increased oxidative stress in multiple organs using multiple biomarkers. Thus, all of the laboratory studies demonstrated oxidative stress with a significant finding in the rat study.

In Hep-G2 cells, a glyphosate formulation demonstrated a robust increase in oxidative stress at 40 µg/ml. Given the negative response in this cell line for glyphosate alone, it must be concluded that this response is not due to glyphosate. In HEP-2 cells, a glyphosate formulation demonstrated a robust increase in oxidative stress via multiple biomarkers at 376 µg/ml and when a surfactant is added, at 180.2 µg/ml. In HaCaT cells, a glyphosate formulation demonstrated significant increases in oxidative stress from doses starting as low as 1.7 µg/ml in a concentration-dependent fashion. No studies were available in human lymphocytes.

In Swiss CD-1 mice, a glyphosate formulation significantly increased oxidative stress in the kidney but only demonstrated a mild (non-significant) increase in the liver. This study evaluated oxidative stress at two different time points following exposure and saw responses that differed over time. The strong increase in the liver for glyphosate but not glyphosate formulation, suggests a complicated response pattern for pure glyphosate versus the formulation that could be linked to the time since exposure. In Swiss Albino mice, a glyphosate formulation demonstrated increased oxidative stress by two separate biomarkers in both the liver and the kidney. In a second study in Swiss albino mice using a different biomarker but a similar dose, increased oxidative stress was seen in both the liver and the kidney. In Wistar rat pups exposed in utero, an increase in oxidative stress was seen in the hippocampus. In Swiss albino mice, topical application of a glyphosate formulation to the skin resulted in a proteomic fingerprint suggesting oxidative stress was increased.

Though there are fewer studies for oxidative stress than there are for genotoxicity, the robust response seen here in human cells and in rodent studies clearly supports a role for both glyphosate and glyphosate formulations in inducing oxidative stress. Thus,

there is a second reasonable mechanism through which the tumors seen in humans and those seen in animals can be caused by glyphosate and glyphosate formulations.

Summary for Biological Plausibility

In the evaluation of causality, the evidence for biological plausibility is overwhelming. Glyphosate clearly causes multiple cancers in mice, two cancers in the hematopoietic system similar to what is seen in humans, causes cancer in rats, is genotoxic and induces oxidative stress. The findings are clear for both glyphosate alone and for glyphosate formulations. **There is strong support for biological plausibility in support of a causal association of glyphosate and glyphosate formulations with NHL.**

Biological Gradient

Only three of the epidemiological studies provided information on biological gradients in their publications.

Eriksson et al. (2008)[46] divided their cases and controls into those with ≤10 days per year of exposure and those with >10 days per year of exposure. The ORs were calculated using a multivariate analysis that included agents with statistically significant increased OR, or with an OR > 1.50 and at least 10 exposed subjects. ORs for glyphosate were 1.69 (0.70-4.07) for ≤10 days per year and 2.36 (1.04-5.37) for >10 days per year. In their multivariate analysis, latency periods of 1-10 years showed an OR of 1.11 (0.24-5.08) and >10 years had an OR of 2.26 (1.16-4.40). Thus, they show an increase with intensity of exposure and with latency.

McDuffie et al. (2001)[50], using a conditional logistic regression analysis controlling for major chemical classes of pesticides and all other covariates with $p<0.05$, the OR for ≤2 days per year of exposure was 1.0 (0.63-1.57) and for >2 days per year, the OR was 2.12 (1.20-3.73). Thus, they show an increase with intensity of exposure.

De Roos et al. (2005)[45] used three exposure metrics in their analyses: a) ever personally mixed or applied pesticides containing glyphosate; b) cumulative exposure days of use of glyphosate (years of use times days per year); and c) intensity weighted cumulative exposure days (years of use times days per year times intensity of use). For exposure measurements b and c, they divided the respondents into tertiles chosen *a priori* to avoid having sparse data when dealing with rare tumors. For cumulative exposure days and using the lowest exposed tertile as the reference group, the RRs drop with values of 0.7 (0.4-1.4) and 0.9 (0.5-1.6) for tertiles 2 and 3 respectively adjusted for demographic and lifestyle factors and other pesticides (30,699 subjects). When intensity-weighted exposure days are examined, the RRs drop with values of 0.6 (0.3-1.1) and 0.8 (0.5-1.4) for tertiles 2 and 3, respectively adjusted for demographic and lifestyle factors and other pesticides (30,699 subjects). Thus, they do not see a biological gradient in their responses. However, the high frequency of exposure to many pesticides (e.g. 73.8% were exposed to 2,4-D) means subjects with low exposure to glyphosate were likely to be exposed to other agents that may also induce NHL; this could reduce the RRs in the higher exposure classes because it would inflate the RR in the low-exposure referent group.

Eriksson et al. (2008)[46] and **McDuffie et al. (2001)**[50] had consistent results for intensity of exposure per year (≤2 days per year, OR=1.0; ≤10 days per year, OR=1.69; >2 days per year, OR=2.12; >10 days per year, OR=2.26). It is not possible to resolve the remaining differences between these three studies nor is it easy to argue that one study has more weight on this question than any other. The studies use different measures of exposure or time since exposure, are done on different populations and have different statistical power to detect a trend.

In rodent carcinogenicity studies, there is clear evidence of a biological gradient.

In general, there is support that a biological gradient exists for the epidemiological data and thus support from this aspect of the Bradford-Hill evaluation.

Temporal Relationship

Exposure must come before the cancers occur otherwise the epidemiology studies are useless. In this case, it is clear that exposure came before the onset of NHL. **The need for a temporal relationship in the data supporting a causal association between glyphosate and NHL is satisfied.**

Specificity

There are other causes of NHL[215-218] so this group of cancers is not specific to glyphosate. **There is little support for specificity**.

Coherence

Humans, coming into contact with glyphosate, can absorb the compound into their bodies where it has been measured in blood and in urine[56, 219-223]. In laboratory animals, absorption, distribution and elimination of glyphosate and glyphosate compounds have been studied[137, 224] and show that glyphosate gets into the animal's bodies, distributes to numerous organs and is eliminated in urine. The animal cancer studies clearly demonstrate that glyphosate in mammals can have toxic effects.

Mouse models have long served as surrogates for humans in understanding and developing treatments for many diseases. The same holds true for lymphoid tumors seen in humans. For over 30 years, mouse models have been studied and evaluated as surrogates for NHL[225-229]. These publications and the associated classification systems for humans and mice indicate a close linkage between the diseases in humans and mice. Thus, coherence is supported by the increased risk of malignant lymphomas in CD-1 mice, the marginal increase in these tumors in Swiss mice and the strong similarity between malignant lymphomas in mice and NHL in humans.

There is strong support for coherence in the data supporting a causal association of glyphosate and glyphosate formulations with NHL.

Experimental Evidence in Humans

There is no experimental evidence in humans since purposely exposing humans to a pesticide, especially one that is probably carcinogenic, is not ethical and would never pass review by a human subject's advisory board.

Analogy

I am unaware of any analogous compounds from the scientific literature. This, however, is not an area where I have sufficient background to express an opinion.

Summary

Table 18 summarizes the information for each of Hill's aspects of causality. For these data, causality is strengthened because the available epidemiological studies show a consistent positive association between cancer and the exposure. The studies do not show different responses with some studies being positive and others negative, nor do they show any heterogeneity when analyzed together. And, in answer to Hill's question, the relationship between NHL and glyphosate exposure has been observed by different persons, in different places, circumstances, and times.

Causality is strengthened for these data because the strength of the observed associations, when evaluated simultaneously, are statistically significant, the findings are uni-directional and the results are unlikely to be due to chance. Even though none of the individual studies provide relative risks or odds ratios that are large and precise, the meta-analysis has objectively shown that the observed association across these studies is significant and supports a positive association between NHL and glyphosate.

Biological plausibility is strongly supported by the animal carcinogenicity data and the mechanistic data on genotoxicity and oxidative stress. When addressing biological plausibility, the first question generally asked is "Can you show that glyphosate causes cancers in experimental animals?" In this case, the answer to that question is clearly yes. Glyphosate has been demonstrated to cause cancer in two strains of rats and one strain of mice. Glyphosate has been demonstrated to cause cancer in two strains of rats and one strain of mice. Glyphosate causes hepatocellular adenomas in male Wistar rats and male Sprague-Dawley rats, mammary gland adenomas and adenocarcinomas in female Wistar rats and kidney adenomas in male Sprague-Dawley rats. Glyphosate causes hemangiosarcomas, kidney tumors and malignant lymphomas in male CD-1 mice and hemangiosarcomas in female CD-1 mice and possibly causes malignant lymphomas in male Swiss albino mice. Thus, it is biologically plausible that glyphosate alone can cause cancer in mammals.

The next question generally asked is "Does the mechanism by which glyphosate causes cancer in experimental animals also work in humans?" The best understood mechanism by which chemicals cause cancer in both humans and animals is through damaging DNA that leads to mutations in cells that then leads to uncontrolled cellular replication and

eventually cancer. It is absolutely clear from the available scientific data that both glyphosate and glyphosate formulations are genotoxic. This has been amply demonstrated in humans that were exposed to glyphosate, in human cells *in vitro*, in experimental animal models and their cells *in vitro* and *in vivo*, and in wildlife. One way in which DNA can be damaged is through the presence of free oxygen radicals that overwhelm a cell's antioxidant defenses. Glyphosate induces this type of oxidative stress, providing additional support for a biological mechanism that works in humans.

Table 18: Summary conclusions for Hill's nine aspects of epidemiological data and related science

Aspect	Conclusion	Reason
Consistency of the observed association	Strong	Multiple studies, all are positive, meta-analysis shows little heterogeneity, different research teams, different continents, different questionnaires, no obvious bias or confounding
Strength of the observed association	Strong	Six core epidemiology studies all show the same modest increase, significant meta-analyses
Biological plausibility	Very Strong	Multiple cancers in multiple species, not due to chance, increased risk of rare tumors, convincing evidence for genotoxicity and oxidative stress
Biological gradient	Moderate	Clearly seen in the two case-control studies that evaluated it, not seen in the cohort study
Temporal relationship of the observed association	Satisfied	Exposure clearly came before cancers
Specificity of the observed association	Not needed	NHL has other causes, this does not subtract from the causal argument
Coherence	Strong	Glyphosate is absorbed, distributed and excreted from the body, cancers seen in the mice have strong similarity to human NHL
Evidence from human experimentation	No data	No studies are available
Analogy	No data	No studies available in the literature

In general, there is support that a biological gradient exists for the epidemiological data and thus support from this aspect of the Bradford-Hill evaluation. Glyphosate ORs increased with time since first exposure and with intensity of use per year in the two case-control studies that evaluated at least one of these issues.

There is clearly the proper temporal relationship with the exposure coming before the cancers.

The human evidence is coherent. The basic findings in humans agree with the animal evidence for absorption, distribution and elimination of glyphosate. Also, one of the tumors seen in mice has almost the same etiology as NHL.

NHL is not specific to glyphosate exposure. There is no experimental evidence in humans and I did not find any references where researchers looked for analogous compounds with similar toxicity.

Hill (1965)[36] asks "*is there any other way of explaining the set of facts before us, is there any other answer equally, or more, likely than cause and effect?*" There is no better way of explaining the scientific evidence relating glyphosate to an increase in NHL in humans than cause and effect.

In my opinion, glyphosate probably causes NHL and, given the human, animal and experimental evidence, I assert that, to a reasonable degree of scientific certainty, the probability that glyphosate causes NHL is high.

The IARC Assessment of Glyphosate

In March 2015, the International Agency for Research on Cancer (an agency of the World Health Organization) brought together seventeen scientists (the Working Group) to evaluate the scientific evidence on whether glyphosate can cause cancer in humans. This group also contained one invited specialist (myself) to aid the Working Group (WG) in going through the science but who was not allowed to join discussions on the final conclusion or write any part of the document. The Working Group concluded that glyphosate falls in the category *"probably carcinogenic to humans (Group 2A)"*[56].

The IARC preamble[30] guides Working Groups on how to evaluate scientific literature to determine if something is a hazard. All Working Groups follow these guidelines and this process is accepted worldwide as a proper way to evaluate the literature for a hazard (e.g., the European Chemical Agency cites the IARC review process as guidance and then uses the exact same wording as IARC does to guide their own hazard evaluation process[34]).

The WG examined the epidemiological data and classified it as "*limited evidence of carcinogenicity,*" which is defined to mean "*a positive association has been observed between exposure to the agent and cancer for which a causal interpretation is considered to be credible, but chance, bias or confounding could not be ruled out with reasonable confidence.*" This is a precise and clear description of the strength of the evidence from the epidemiological studies.

The WG examined the evidence from animal carcinogenicity studies and classified it as "*sufficient evidence of carcinogenicity,*" which IARC defines as: "*a causal relationship has been established between the agent and an increased incidence of malignant neoplasms or of an appropriate combination of benign and malignant neoplasms in (a) two or more species of animals or (b) two or more independent studies in one species carried out at different times or in different laboratories or under different protocols. A single study in*

one species and sex might be considered to provide sufficient evidence of carcinogenicity when malignant neoplasms occur to an unusual degree with regard to incidence, site, type of tumour or age at onset, or when there are strong findings of tumours at multiple sites." Based on the data available to IARC at the time of their review and the restrictions placed on the studies they can review by the Preamble, this conclusion is justified and correct.

One of the major criticisms of the WG review was that the WG did not review all of the animal carcinogenicity data that was available to the regulatory bodies and thus came to the wrong conclusions on the animal cancer data. In this review, I evaluated all 19 animal carcinogenicity experiments that have been collectively mentioned by any agency that reviews glyphosate. Where possible, I have analyzed the original data and used sound statistical methods to test for significant increases in cancer incidence in animals exposed to glyphosate. My conclusion is that the WG would have called this data "*sufficient evidence*" to support their findings despite not reviewing the additional studies analyzed herein. Despite the fact the industry kept these studies confidential, nothing contained in the withheld studies would have changed the WG conclusion.

On the mechanistic data, the IARC Working Group reviewed the same data that I reviewed, but I also evaluated, where possible, the proprietary data supporting the regulatory decisions. Where possible, I reanalyzed that data to be certain the results being presented were accurate. The IARC Working Group, using the guidelines set forth in their Preamble, declared strong support for the biological mechanisms of genotoxicity and oxidative stress. As I have shown here, there is strong support for these two mechanisms, even with the proprietary evidence from the industry studies. Thus, the IARC Working Group reached the correct conclusion.

To decide on a final classification for a compound, the IARC Preamble provides guidance on how the classification of the three areas are to be used. If the data in humans is "*limited*" and the data from animal carcinogenicity studies is "*sufficient*," the discussions should begin with Class 2A, "*the agent is probably carcinogenic to humans*." Then, given the overall quality of the data set, the strength of the evidence from the mechanistic studies and any additional scientific issues that need to be considered, the Working Group will determine whether the data justifies a different category. In this case, the Working Group concluded 2A was the right category and I still believe the evidence supports that finding.

The EPA Assessment of Glyphosate

Like IARC, the EPA has guidelines that are to be followed when evaluating scientific literature and making a determination about the carcinogenic potential of a chemical. Those guidelines have been developed over many years and are based on sound scientific guidance that myself and many other scientists have provided to the Agency. For their evaluation of glyphosate, the Agency did not follow their own guidelines, nor did they follow sound scientific practice. This opinion is consistent with the review done by the **EPA FIFRA Scientific Advisory Panel**[54]. In addition, the Agency failed to find all of the relevant animal cancer studies and misinterpreted several of them. The major

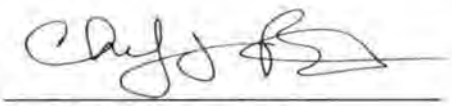
problems with the Agency evaluation are:

- Misinterpretation of the epidemiological evidence, confusing the potential for bias and potential for confounding with real bias and real confounding, allowing them to give almost no weight to the case-control studies in favor of the one cohort study;
- Misinterpretation of the findings in the meta-analysis;
- Failure to properly use historical controls in the analysis of the animal carcinogenicity studies; declaring a significant finding as not due to the compound if it is in the range of the historical controls;
- Failure to analyze all tumors in all studies relying upon the industry submissions to have done this correctly;
- Failure to follow their guidelines on what constitutes a positive finding, disregarding significant trend tests when no corresponding pairwise comparisons are also significant;
- Disregarding positive findings in doses that are clearly not above the maximum dose the animals could be given with compromising the integrity of the study;
- Using unreasonable arguments about the overall false positive rates in the study without actually doing an analysis of this issue;
- Failing to recognize the similar findings in similar studies and to do a pooled analysis to determine if the negative effects in one study cancel out the positive effects in another;
- Giving very little weight to studies from the literature and relying almost entirely on studies provided by industry that have not undergone peer review for both quality and, more importantly in some cases, interpretation of the findings; and
- Comparing results across different species and strains for the animal cancer studies and the mechanistic studies with little regard for unique findings in any one study and consistent findings across multiple studies.

Similar comments apply to the evaluation done by the **European Food Safety Authority**[88] and the **European Chemical Agency**[230]. My detailed comments to these agencies on their risk assessments are attached. There were comments to my comments to EPA by other scientists and I also responded to those comments in the EPA docket for glyphosate. These are also included in the attached Appendices.

Compensation

I am being compensated at $450 per hour for my expert work in this case, plus travel expenses.



Dr. Christopher J. Portier

Cited References

1. Portier, C. and D. Hoel, *Optimal design of the chronic animal bioassay.* J Toxicol Environ Health, 1983. **12**(1): p. 1-19.
2. Portier, C.J. and D.G. Hoel, *Design of the Chronic Animal Bioassay for Goodness of Fit to Multistage Models.* Biometrics, 1983. **39**(3): p. 809-809.
3. Bailer, A.J. and C.J. Portier, *Effects of treatment-induced mortality and tumor-induced mortality on tests for carcinogenicity in small samples.* Biometrics, 1988. **44**(2): p. 417-31.
4. Portier, C.J. and A.J. Bailer, *Testing for increased carcinogenicity using a survival-adjusted quantal response test.* Fundam Appl Toxicol, 1989. **12**(4): p. 731-7.
5. Portier, C.J., J.C. Hedges, and D.G. Hoel, *Age-specific models of mortality and tumor onset for historical control animals in the National Toxicology Program's carcinogenicity experiments.* Cancer Res, 1986. **46**(9): p. 4372-8.
6. Portier, C.J. and A.J. Bailer, *2-Stage Models of Tumor-Incidence for Historical Control Animals in the National Toxicology Programs Carcinogenicity Experiments.* Journal of Toxicology and Environmental Health, 1989. **27**(1): p. 21-45.
7. Portier, C.J. and L. Edler, *Two-stage models of carcinogenesis, classification of agents, and design of experiments.* Fundam Appl Toxicol, 1990. **14**(3): p. 444-60.
8. Portier, C.J., et al., *Biologically based models for risk assessment.* IARC Sci Publ, 1990(104): p. 20-8.
9. Kopp-Schneider, A. and C.J. Portier, *Distinguishing between models of carcinogenesis: the role of clonal expansion.* Fundam Appl Toxicol, 1991. **17**(3): p. 601-13.
10. Kopp-Schneider, A., C.J. Portier, and F. Rippmann, *The application of a multistage model that incorporates DNA damage and repair to the analysis of initiation/promotion experiments.* Math Biosci, 1991. **105**(2): p. 139-66.
11. Kopp-Schneider, A., C.J. Portier, and F. Rippmann, *The Application of a Multistage Model That Incorporates DNA Damage and Repair to the Analysis of Initiation Promotion Experiments.* Mathematical Biosciences, 1991. **105**(2): p. 139-166.
12. Kopp-Schneider, A. and C.J. Portier, *Birth and Death Differentiation Rates of Papillomas in Mouse Skin.* Carcinogenesis, 1992. **13**(6): p. 973-978.
13. Portier, C.J., A. Kopp-Schneider, and C.D. Sherman, *Using Cell Replication Data in Mathematical-Modeling in Carcinogenesis.* Environmental Health Perspectives, 1993. **101**: p. 79-86.
14. Kopp-Schneider, A., C.J. Portier, and C.D. Sherman, *The Exact Formula for Tumor-Incidence in the 2-Stage Model.* Risk Analysis, 1994. **14**(6): p. 1079-1080.
15. Portier, C., et al., *Modeling the number and size of hepatic focal lesions following exposure to 2378-TCDD.* Organohalogen Compounds, 1994. **21**: p. 393-397.

16. Kopp-Schneider, A. and C.J. Portier, *Carcinoma formation in NMRI mouse skin painting studies is a process suggesting greater than two stages.* Carcinogenesis, 1995. **16**(1): p. 53-9.
17. Portier, C.J., A. Kopp-Schneider, and C.D. Sherman, *Calculating tumor incidence rates in stochastic models of carcinogenesis.* Mathematical Biosciences, 1996. **135**(2): p. 129-146.
18. Portier, C.J., et al., *Modeling the number and size of hepatic focal lesions following exposure to 2,3,7,8-TCDD.* Toxicology and Applied Pharmacology, 1996. **138**(1): p. 20-30.
19. Kopp-Schneider, A., C. Portier, and P. Bannasch, *A model for hepatocarcinogenesis treating phenotypical changes in focal hepatocellular lesions as epigenetic events.* Math Biosci, 1998. **148**(2): p. 181-204.
20. Toyoshiba, H., et al., *Gene interaction network analysis suggests differences between high and low doses of acetaminophen.* Toxicol Appl Pharmacol, 2006. **215**(3): p. 306-16.
21. Gohlke, J.M. and C.J. Portier, *The forest for the trees: a systems approach to human health research.* Environ Health Perspect, 2007. **115**(9): p. 1261-3.
22. Gohlke, J.M., et al., *Genetic and environmental pathways to complex diseases.* BMC Syst Biol, 2009. **3**: p. 46.
23. Gohlke, J.M., et al., *Estimating the global public health implications of electricity and coal consumption.* Environ Health Perspect, 2011. **119**(6): p. 821-6.
24. Aylward, L.L., et al., *Evaluation of biomonitoring data from the CDC National Exposure Report in a risk assessment context: perspectives across chemicals.* Environ Health Perspect, 2013. **121**(3): p. 287-94.
25. Trong, l. and C. Portier. *Proceedings of the Viet Nam – United States Scientific Conferences on Human Health and Environmental Effects of Agent Orange/Dioxin, Part 1 and 2.* 2002. Ha Noi, Vietnam: US National Institute of Environmental Sciences and the Government of Vietnam.
26. Portier, C.J. and M.S. Wolfe, eds. *EMF Science Review Symposium Breakout Group Report for Epidemiology Research Findings*. 1998, National Institute of Environmental Health Sciences: Research Triangle Park, North Carolina.
27. Portier, C.J. and M.S. Wolfe, eds. *EMF Science Review Symposium Breakout Group Report for Clinical and In Vivo Laboratory Findings*. NIH Publication Number 98-4400. 1998, National Institute of Environmental Health Sciences: Research Triangle Park, North Carolina.
28. Portier, C.J. and M.S. Wolfe, eds. *Assessment of Health Effects from Exposure to Power-Line Frequency Electric and Magnetic Fields*. NIH Publication Number 98-3981. 1998, National Institute of Environmental Health Sciences: Research Triangle Park, North Carolina. 508.
29. Portier, C.J., et al., *A Human Health Perspective on Climate Change*, N.I.o.E.H.S. Health and Human Services, Editor. 2010, Environmental Health Perspectives: Research Triangle Park, NC72.
30. IARC. *Preamble to the IARC Monographs*. 2006 [cited 2017 3/8/17]; Available from: http://monographs.iarc.fr/ENG/Preamble/CurrentPreamble.pdf.

31. National Research Council. Science and Judgment in Risk Assessment. 1994, Washington (DC): National Academy of Sciences Press.
32. National Research Council. Science and Decisions: Advancing Risk Assessment. 2009, Washington (DC): National Academies Press.
33. USEPA, *Guidelines for Carcinogen Risk Assessment*, U.E.P. Agency, Editor. 2005: Washington DC166.
34. European Chemicals Agency, *Guidance on the Application of the CLP Criteria: Guidance to Regulation (EC) No 1272/2008 on classification, labelling and packaging (CLP) of substances and mixtures*. 2015, European Chemicals Agency: Helsinki, Finland.
35. National Toxicology Program. *Handbook for Preparing Report on Carcinogens Monographs*. 2015 [cited 2017 3/8/17]; Available from: https://ntp.niehs.nih.gov/ntp/roc/handbook/roc_handbook_508.pdf.
36. Hill, A.B., *The Environment and Disease: Association or Causation?* Proc R Soc Med, 1965. **58**: p. 295-300.
37. Smith, M.T., et al., *Key Characteristics of Carcinogens as a Basis for Organizing Data on Mechanisms of Carcinogenesis*. Environ Health Perspect, 2016. **124**(6): p. 713-21.
38. Chang, E.T. and E. Delzell, *Systematic review and meta-analysis of glyphosate exposure and risk of lymphohematopoietic cancers*. J Environ Sci Health B, 2016: p. 1-27.
39. Cantor, K.P., et al., *Pesticides and other agricultural risk factors for non-Hodgkin's lymphoma among men in Iowa and Minnesota*. Cancer Res, 1992. **52**(9): p. 2447-2455.
40. Nordstrom, M., et al., *Occupational exposures, animal exposure and smoking as risk factors for hairy cell leukaemia evaluated in a case-control study*. Br.J Cancer, 1998. **77**(11): p. 2048-2052.
41. Hardell, L. and M. Eriksson, *A case-control study of non-Hodgkin lymphoma and exposure to pesticides*. Cancer, 1999. **85**(6): p. 1353-1360.
42. Hardell, L., M. Eriksson, and M. Nordstrom, *Exposure to pesticides as risk factor for non-Hodgkin's lymphoma and hairy cell leukemia: pooled analysis of two Swedish case-control studies*. Leuk.Lymphoma, 2002. **43**(5): p. 1043-1049.
43. De Roos, A.J., et al., *Integrative assessment of multiple pesticides as risk factors for non-Hodgkin's lymphoma among men*. Occup Environ Med, 2003. **60**(9): p. E11.
44. Lee, W.J., et al., *Non-Hodgkin's lymphoma among asthmatics exposed to pesticides*. Int J Cancer, 2004. **111**(2): p. 298-302.
45. De Roos, A.J., et al., *Cancer incidence among glyphosate-exposed pesticide applicators in the Agricultural Health Study*. Environ Health Perspect, 2005. **113**(1): p. 49-54.
46. Eriksson, M., et al., *Pesticide exposure as risk factor for non-Hodgkin lymphoma including histopathological subgroup analysis*. Int J Cancer, 2008. **123**(7): p. 1657-63.

47. Orsi, L., et al., *Occupational exposure to pesticides and lymphoid neoplasms among men: results of a French case-control study.* Occup Environ Med, 2009. **66**(5): p. 291-8.
48. Hohenadel, K., et al., *Exposure to multiple pesticides and risk of non-Hodgkin lymphoma in men from six Canadian provinces.* Int J Environ Res Public Health, 2011. **8**(6): p. 2320-30.
49. Cocco, P., et al., *Lymphoma risk and occupational exposure to pesticides: results of the Epilymph study.* Occup Environ Med, 2013. **70**(2): p. 91-8.
50. McDuffie, H.H., et al., *Non-Hodgkin's lymphoma and specific pesticide exposures in men: cross-Canada study of pesticides and health.* Cancer Epidemiol Biomarkers Prev, 2001. **10**(11): p. 1155-1163.
51. Zahm, S.H., et al., *A case-control study of non-Hodgkin's lymphoma and the herbicide 2,4-dichlorophenoxyacetic acid (2,4-D) in eastern Nebraska.* Epidemiology, 1990. **1**(5): p. 349-356.
52. Hoar, S.K., et al., *Agricultural herbicide use and risk of lymphoma and soft-tissue sarcoma.* JAMA, 1986. **256**(9): p. 1141-1147.
53. Blair, A., et al., *Reliability of reporting on life-style and agricultural factors by a sample of participants in the Agricultural Health Study from Iowa.* Epidemiology, 2002. **13**(1): p. 94-9.
54. FIFRA Scientific Advisory Panel Meeting Minutes, *Meeting Minutes and Final Report of the December 13-16, 2016 FIFRA SAP Meeting Held to Consider and Review Scientific Issues Associated with EPA's Evaluation of the Carcinogenic Potential of Glyphasate*, O.o.P. Programs, Editor. 2017, US Environmental Protection Agency: Washington, DC101.
55. Portier, C.J., et al., *Differences in the carcinogenic evaluation of glyphosate between the International Agency for Research on Cancer (IARC) and the European Food Safety Authority (EFSA).* J Epidemiol Community Health, 2016. **70**(8): p. 741-5.
56. IARC Working Group, *Glyphosate*, In: *Some Organophosphate Insecticides and Herbicides: Diazinon, Glyphosate, Malathion, Parathion, and Tetrachlorvinphos*. 2015, IARC Monogr Prog. V. 112. p. 1-92.
57. Schinasi, L. and M.E. Leon, *Non-Hodgkin lymphoma and occupational exposure to agricultural pesticide chemical groups and active ingredients: a systematic review and meta-analysis.* Int J Environ Res Public Health, 2014. **11**(4): p. 4449-527.
58. Ioannidis, J.P., *Interpretation of tests of heterogeneity and bias in meta-analysis.* J Eval Clin Pract, 2008. **14**(5): p. 951-7.
59. OECD, *Guidance Document 116 on the Conduct and Design of Chronic Toxicity and Carcinogenicity Studies*, H.a.S.P. Environment, Editor. 2012, OECD: Paris.
60. Haseman, J.K., *Statistical issues in the design, analysis and interpretation of animal carcinogenicity studies.* Environ Health Perspect, 1984. **58**: p. 385-92.
61. Environmental Protection Agency, *Glyphosate Issue Paper: Evaluation of Carcinogenic Potential*. 2016: Washington, DC.
62. NRC Committee to Review the Styrene Assessment in the National Toxicology Program 12th Report on Carcinogens, in *Review of the Styrene Assessment in the*

National Toxicology Program 12th Report on Carcinogens: Workshop Summary. 2014, National Academies Press: Washington (DC).

63. Bieler, G.S. and R.L. Williams, *Ratio estimates, the delta method, and quantal response tests for increased carcinogenicity*. Biometrics, 1993. **49**(3): p. 793-801.
64. Haseman, J.K., G.A. Boorman, and J. Huff, *Value of historical control data and other issues related to the evaluation of long-term rodent carcinogenicity studies*. Toxicol Pathol, 1997. **25**(5): p. 524-7.
65. Haseman, J.K., J. Huff, and G.A. Boorman, *Use of historical control data in carcinogenicity studies in rodents*. Toxicol Pathol, 1984. **12**(2): p. 126-35.
66. OECD, *Combined Chronic Toxicity\Carcinogenicity Studies, OECD Guidelines for Testing of Chemicals, No. 453*, OECD, Editor. 2009: Paris.
67. Atkinson, C., Strutt, A., Henderson, W., et al. , *104-Week Chronic Feeding/ Oncogenicity study in rats with 52-week interim kill*. 1993.
68. Brammer., *Glyphosate Acid: Two Year Dietary Toxicity and Oncogenicity Study in Wistar Rats*. 2001: Central Toxicology Laboratory, Alderley Park Macclesfield, Cheshire, UK.
69. Burnett, P., Borders, J.;,Kush, J. , *Report to Monsanto Company: Two Year Chronic Oral Toxicity Study with CP- 76100 in Albino Rats*. 1979, Industrial Bio-Test Laboratories, Inc.: on behalf of Monsanto Co.
70. Chruscielska, K.B., J.; Kita, K, et al., *Glyphosate: Evaluation of chronic activity and possible far - reaching effects - Part 1. Studies on chronic toxicity*. Pestycydy, 2000. **3-4**: p. 10.
71. Enemoto, K., *24-Month Oral Chronic Toxicity and Oncogenicity Study in Rats, Vol. 1*. . 1997: The Institute of Environmental Toxicology, Kodaira-shi, Tokyo, Japan.
72. Excel, *Combined chronic toxicity/carcinogenicity study of glyphosate technical in Sprague Dawley rats*. . 1997: Indian Institute of Toxicology, Pune, India.
73. Lankas, G., P., *A Lifetime Study of Glyphosate in Rats*. 1981, Monsanto: Report No. 77-2062 prepared by Bio Dynamics, Inc.
74. Pavkov, K.L., Wyand, S. , *Two-Year Chronic Toxicity and Oncogenecity Dietary Study with SC-0224 in Rats*. 1987: Stauffer Chemical Company.
75. Reyna, M.S., Gordon, D.E. , *Two- Year Chronic Oral Toxicity Study with CP67573 in Albino Rats: Monsanto*. 1974.
76. Seralini, G.E., et al., *Republished study: long-term toxicity of a Roundup herbicide and a Roundup-tolerant genetically modified maize*. Environmental Sciences Europe, 2014. **26**(1): p. 14.
77. Stout, L.D.a.R., P.A., *Chronic Study of Glyphosate Administered in Feed to Albino Rats*. 1990: Monsanto.
78. Suresh, T.P., *Combined Chronic Toxicity and Carcinogenicity Study with Glyphosate Technical in Wistar Rats*. 1996, Syngenta: Toxicology Department Rallis Research Centre, Rallis India Limited.
79. Wood, E., Dunster, J., Watson, P., and Brooks, P. , *Glyphosate Technical: Dietary Combined Chronic Toxicity/Carcinogenicity Study in the Rat*. 2009: Harlan Laboratories Limited, Shardlow Business Park, Shardlow, Derbyshire DE72 2GD, UK. Study No. 2060-012. April, 23, 2009.

80. Atkinson, C., Martin, T., Hudson, P., and Robb, D., *Glyphosate: 104 week dietary carcinogenicity study in mice*. 1993: Inveresk Research International, Tranent, EH33 2NE, Scotland. IRI Project No. 438618. April 7, 1993. .
81. George, J., et al., *Studies on glyphosate-induced carcinogenicity in mouse skin: a proteomic approach.* J Proteomics, 2010. **73**(5): p. 951-64.
82. Knezevich, A.L. and G.K. Hogan, *A chronic feeding study of glyphosate in mice: Monsanto*. 1983: Bio/Dynamic Inc., dated July 21, 1983. Report No. 77-2011. .
83. Kumar, D.P.S., *Carcinogenicity Study with Glyphosate Technical in Swiss Albino Mice*. 2001: Toxicology Department Rallis Research Centre, Rallis India Limited. Study No. TOXI: 1559.CARCI-M. .
84. Pavkov, K.L., Turnier, J.C. , *Two-Year Chronic Toxicity and Oncogenecity Dietary Study with SC-0224 in Mice*. 1987: Stauffer Chemical Company.
85. Reyna, M.S. and D.E. Gordon, *18-Month Carcinogenicity Study with CP67573 in Swiss White Mice: Monsanto*. 1973: Industrial Bio-Test Laboratories, Inc.
86. Sugimoto, K., *18-Month Oral Oncogenicity Study in Mice, Vol. 1 and 2*. . 1997: The Institute of Environmental Toxicology, 2-772, Suzuki-cho, Kodaira-shi, Tokyo, 187, Japan, Study No.:IET 94-0151. .
87. Wood, E., Dunster, J., Watson, P., and Brooks, P. , *Glyphosate Technical: Dietary Carcinogenicity Study in the Mouse*. 2009: Harlan Laboratories Limited, Shardlow Business Park, Shardlow, Derbyshire DE72 2GD, UK. Study No. 2060-011. April, 22, 2009.
88. (BfR)., G.F.I.f.R.A., *Glyphosate Renewal Assessment Report: Toxicology and Metabolism*. 2013. V. 3. p.
89. (BfR)., G.F.I.f.R.A., *Glyphosate Renewal Assessment Report: Assessment of IARC Monographies Volume 112 (2015)*. 2015. V. 3Glyphosate Addendum I to RAR. p.
90. Greim, H., et al., *Evaluation of carcinogenic potential of the herbicide glyphosate, drawing on tumor incidence data from fourteen chronic/carcinogenicity rodent studies.* Crit Rev Toxicol, 2015. **45**(3): p. 185-208.
91. Blettner, M., et al., *Traditional reviews, meta-analyses and pooled analyses in epidemiology*. Int J Epidemiol, 1999. **28**(1): p. 1-9.
92. Friedenreich, C.M., *Methods for pooled analyses of epidemiologic studies.* Epidemiology, 1993. **4**(4): p. 295-302.
93. Portier, C.J., *Comments on Glyphosate Issue Paper (EPA-HQ-OPP-2016-0385-0094)*. 2016, US EPA Docket Number (EPA-HQ-OPP-2016-0385-0094): Washington DC.
94. Hogan, g.k., *A chronic Feeding Study of Glyphosate (Roundup Technical) in Rats*. 1981, Monsanto: Bio/dynamics Inc., E. Millstone, NJ.
95. EPA, *EPA Memo Stout and Ruecker*, I. William Dykstra.Toxicology Branch, Editor. 1991. V. MRID 416438-01 Tox review 008897. p.
96. Uno, H., et al., *A versatile test for equality of two survival functions based on weighted differences of Kaplan-Meier curves.* Stat Med, 2015. **34**(28): p. 3680-95.
97. Giknis, M. and C. Clifford. *Neoplastic and Non-Neoplastic Lesions in the Charles River Wistar Hannover [Crl:WI(Han)] Rat*. 2011; Available from:

http://www.criver.com/files/pdfs/rms/wistarhan/rm_rm_r_wistar_han_tox_data_2011.aspx.

98. Acquavella, J., et al., *Glyphosate epidemiology expert panel review: a weight of evidence systematic review of the relationship between glyphosate exposure and non-Hodgkin's lymphoma or multiple myeloma.* Crit Rev Toxicol, 2016. **46**(sup1): p. 28-43.
99. Lacayo, H., *Memorandum: Use of historical data in determiningthe weight of evidence from kidney tumor incidence in the Glyphosate two-year feeding study and some remarks on false positives*, S.M.S. Branch, Editor. 1985, US EPA: Washington, DCC.
100. Chandra, M. and C.H. Frith, *Spontaneous neoplasms in aged CD-1 mice.* Toxicol Lett, 1992. **61**(1): p. 67-74.
101. Giknis, M. and C. Clifford, *Spontaneous Neoplastic Lesions in the Crl:CD-1(ICR)BR Mouse*. 2000, Charles River Laboratories.
102. Giknis, M. and C. Clifford, *Spontaneous Neoplastic Lesions in the Crl:CD-1(ICR)BR Mouse in Control Groups from 18 Month to 2 year Studies*. 2005, Charles River Laboratories.
103. Eustis, S.L., et al., *The utility of multiple-section sampling in the histopathological evaluation of the kidney for carcinogenicity studies*. Toxicol Pathol, 1994. **22**(5): p. 457-72.
104. de Vivar Chevez, A.R., J. Finke, and R. Bukowski, *The role of inflammation in kidney cancer.* Adv Exp Med Biol, 2014. **816**: p. 197-234.
105. Dixon, D., et al., *Summary of chemically induced pulmonary lesions in the National Toxicology Program (NTP) toxicology and carcinogenesis studies.* Toxicol Pathol, 2008. **36**(3): p. 428-39.
106. Son, W.C. and C. Gopinath, *Early occurrence of spontaneous tumors in CD-1 mice and Sprague-Dawley rats.* Toxicol Pathol, 2004. **32**(4): p. 371-4.
107. OECD, *Carcinogenicity Studies, OECD Guideline for the Testing of Chemicals, No. 451*, OECD, Editor. 2009: Paris.
108. Haseman, J.K., *Comments on the laboratory animal carcinogenicity studies evaluated in EPA's Glyphosate Issue Paper: Evaluation of Carcinogenic Potential, and an assessment of the comments by Dr. Christopher Portier on these studies*. 2016, US EPA Docket Number (EPA-HQ-OPP-2016-0385-0094): Washington DC.
109. Hanahan, D. and R.A. Weinberg, *Hallmarks of cancer: the next generation.* Cell, 2011. **144**(5): p. 646-74.
110. Higgins, J.P.T. and S. Green, *Cochrane Handbook for Systematic Reviews of Interventions*. 2011, The Cochrane Collaboration.
111. Birnbaum, L.S., et al., *Implementing systematic review at the National Toxicology Program: status and next steps.* Environ Health Perspect, 2013. **121**(4): p. A108-9.
112. Murray, H.E. and K.A. Thayer, *Implementing systematic review in toxicological profiles: ATSDR and NIEHS/NTP collaboration.* J Environ Health, 2014. **76**(8): p. 34-5.

113. Rooney, A.A., et al., *Systematic review and evidence integration for literature-based environmental health science assessments.* Environ Health Perspect, 2014. **122**(7): p. 711-8.
114. Vandenberg, L.N., et al., *A proposed framework for the systematic review and integrated assessment (SYRINA) of endocrine disrupting chemicals.* Environ Health, 2016. **15**(1): p. 74.
115. Paz-y-Miño, C., et al., *Evaluation of DNA damage in an Ecuadorian population exposed to glyphosate.* Genetics and Molecular Biology, 2007. **30**(2).
116. Paz-y-Miño, C., et al., *Baseline determination in social, health, and genetic areas in communities affected by glyphosate aerial spraying on the northeastern Ecuadorian border.* Reviews on Environmental Health, 2011. **26**(1).
117. Bolognesi, C., et al., *Biomonitoring of genotoxic risk in agricultural workers from five colombian regions: association to occupational exposure to glyphosate.* J Toxicol Environ Health A, 2009. **72**(15-16): p. 986-97.
118. Kier, L.D., *Review of genotoxicity biomonitoring studies of glyphosate-based formulations.* Crit Rev Toxicol, 2015. **45**(3): p. 209-18.
119. Mladinic, M., et al., *Evaluation of genome damage and its relation to oxidative stress induced by glyphosate in human lymphocytes in vitro.* Environ Mol Mutagen, 2009. **50**(9): p. 800-7.
120. Alvarez-Moya, C., et al., *Comparison of the in vivo and in vitro genotoxicity of glyphosate isopropylamine salt in three different organisms.* Genet.Mol Biol, 2014. **37**(1): p. 105-110.
121. Manas, F., et al., *Genotoxicity of glyphosate assessed by the comet assay and cytogenetic tests.* Environ Toxicol Pharmacol, 2009. **28**(1): p. 37-41.
122. Monroy, C.M., et al., *Citotoxicidad y genotoxicidad en células humanas expuestas in vitro a glifosato.* Biomédica, 2005. **25**: p. 335-45.
123. Lueken, A., et al., *Synergistic DNA damage by oxidative stress (induced by H2O2) and nongenotoxic environmental chemicals in human fibroblasts.* Toxicology Letters, 2004. **147**(1): p. 35-43.
124. Koller, V.J., et al., *Cytotoxic and DNA-damaging properties of glyphosate and Roundup in human-derived buccal epithelial cells.* Arch Toxicol, 2012. **86**(5): p. 805-13.
125. Gasnier, C., et al., *Glyphosate-based herbicides are toxic and endocrine disruptors in human cell lines.* Toxicology, 2009. **262**(3): p. 184-91.
126. Mladinic, M., P. Perkovic, and D. Zeljezic, *Characterization of chromatin instabilities induced by glyphosate, terbuthylazine and carbofuran using cytome FISH assay.* Toxicol Lett, 2009. **189**(2): p. 130-7.
127. Bolognesi, C., et al., *Genotoxic Activity of Glyphosate and Its Technical Formulation Roundup.* J. Agric. Food Chem., 1997. **45**: p. 1957-1962.
128. Lioi, M.B., et al., *Genotoxicity and oxidative stress induced by pesticide exposure in bovine lymphocyte cultures in vitro.* Mutat Res, 1998. **403**(1-2): p. 13-20.
129. Vigfusson, N.V. and E.R. Vyse, *The effect of the pesticides, Dexon, Captan and Roundup, on sister-chromatid exchanges in human lymphocytes in vitro.* Mutat Res, 1980. **79**(1): p. 53-57.

130. Peluso, M., et al., *32P-postlabeling detection of DNA adducts in mice treated with the herbicide Roundup.* Environ Mol Mutagen, 1998. **31**(1): p. 55-59.
131. Rank, J., et al., *Genotoxicity testing of the herbicide Roundup and its active ingredient glyphosate isopropylamine using the mouse bone marrow micronucleus test, Salmonella mutagenicity test, and Allium anaphase-telophase test.* Mutat Res, 1993. **300**(1): p. 29-36.
132. Dimitrov, B.D., et al., *Comparative genotoxicity of the herbicides Roundup, Stomp and Reglone in plant and mammalian test systems.* Mutagenesis, 2006. **21**(6): p. 375-82.
133. Prasad, S., et al., *Clastogenic effects of glyphosate in bone marrow cells of swiss albino mice.* J Toxicol, 2009. **2009**: p. 308985.
134. Grisolia, C.K., *A comparison between mouse and fish micronucleus test using cyclophosphamide, mitomycin C and various pesticides.* Mutat Res, 2002. **518**(2): p. 145-150.
135. Coutinho do Nascimento A and G. C., *Comparative analysis between micronuclei tests in mice and in peripheral erythrocytes of Oreochromis niloticus in evaluation of mutagenic potential of the agrotoxins deltamethrin, dicofol, glyphosate, and imazapyr.* Pesticides: R Ecotoxicol E Meio Ambiente, Curitiba, 2000. **10**: p. 8.
136. Cavusoglu, K., et al., *Protective effect of Ginkgo biloba L. leaf extract against glyphosate toxicity in Swiss albino mice.* J Med Food, 2011. **14**(10): p. 1263-72.
137. Chan, P. and J. Mahler, *Toxicity Studies of Glyphosate (CAS No. 1071836) Administered in Dosed Feed to F344/N Rats and B6C3F1 Mice*, N.T. Program, Editor. 1992: Research Triangle Park, NC.
138. Li, A.P. and T.J. Long, *An evaluation of the genotoxic potential of glyphosate.* Fundam.Appl Toxicol, 1988. **10**(3): p. 537-546.
139. Roustan, A., et al., *Genotoxicity of mixtures of glyphosate and atrazine and their environmental transformation products before and after photoactivation.* Chemosphere, 2014. **108**: p. 93-100.
140. Sivikova, K. and J. Dianovsky, *Cytogenetic effect of technical glyphosate on cultivated bovine peripheral lymphocytes.* Int J Hyg Environ Health, 2006. **209**(1): p. 15-20.
141. HOLEČKOVÁ, B., *EVALUATION OF THE IN VITRO EFFECT OF GLYPHOSATE-BASED HERBICIDE ON BOVINE LYMPHOCYTES USING CHROMOSOME PAINTING.* Bull Vet Inst Pulawy, 2006. **50**: p. 533-536.
142. Moreno, N.C., S.H. Sofia, and C.B. Martinez, *Genotoxic effects of the herbicide Roundup Transorb and its active ingredient glyphosate on the fish Prochilodus lineatus.* Environ Toxicol Pharmacol, 2014. **37**(1): p. 448-54.
143. Lopes, F.M., et al., *Effect of glyphosate on the sperm quality of zebrafish Danio rerio.* Aquat Toxicol, 2014. **155**: p. 322-6.
144. Guilherme, S., et al., *Are DNA-damaging effects induced by herbicide formulations (Roundup(R) and Garlon(R)) in fish transient and reversible upon cessation of exposure?* Aquat Toxicol, 2014. **155**: p. 213-21.
145. Akcha, F., C. Spagnol, and J. Rouxel, *Genotoxicity of diuron and glyphosate in oyster spermatozoa and embryos.* Aquat Toxicol, 2012. **106-107**: p. 104-13.

146. Kaya, B., et al., *Use of the Drosophila wing spot test in the genotoxicity testing of different herbicides.* Environ Mol Mutagen, 2000. **36**(1): p. 40-6.
147. Cavalcante, D.G., C.B. Martinez, and S.H. Sofia, *Genotoxic effects of Roundup on the fish Prochilodus lineatus.* Mutat Res, 2008. **655**(1-2): p. 41-6.
148. Cavas, T. and S. Konen, *Detection of cytogenetic and DNA damage in peripheral erythrocytes of goldfish (Carassius auratus) exposed to a glyphosate formulation using the micronucleus test and the comet assay.* Mutagenesis, 2007. **22**(4): p. 263-8.
149. de Castilhos Ghisi, N. and M.M. Cestari, *Genotoxic effects of the herbicide Roundup((R)) in the fish Corydoras paleatus (Jenyns 1842) after short-term, environmentally low concentration exposure.* Environ Monit Assess, 2013. **185**(4): p. 3201-7.
150. De Souza Filho, J., et al., *Mutagenicity and genotoxicity in gill erythrocyte cells of Poecilia reticulata exposed to a glyphosate formulation.* Bull Environ Contam Toxicol, 2013. **91**(5): p. 583-7.
151. Gholami-Seyedkolaei, S.J., et al., *Optimization of recovery patterns in common carp exposed to roundup using response surface methodology: evaluation of neurotoxicity and genotoxicity effects and biochemical parameters.* Ecotoxicol Environ Saf, 2013. **98**: p. 152-61.
152. Guilherme, S., et al., *European eel (Anguilla anguilla) genotoxic and pro-oxidant responses following short-term exposure to Roundup--a glyphosate-based herbicide.* Mutagenesis, 2010. **25**(5): p. 523-30.
153. Guilherme, S., et al., *Differential genotoxicity of Roundup((R)) formulation and its constituents in blood cells of fish (Anguilla anguilla): considerations on chemical interactions and DNA damaging mechanisms.* Ecotoxicology, 2012. **21**(5): p. 1381-90.
154. Marques, A., et al., *Progression of DNA damage induced by a glyphosate-based herbicide in fish (Anguilla anguilla) upon exposure and post-exposure periods--insights into the mechanisms of genotoxicity and DNA repair.* Comp Biochem Physiol C Toxicol Pharmacol, 2014. **166**: p. 126-33.
155. Marques, A., et al., *Erratum To: Progression of DNA damage induced by a glyphosate-based herbicide in fish (Anguilla anguilla) upon exposure and post-exposure periods--insights into the mechanisms of genotoxicity and DNA repair.* Comp Biochem Physiol C Toxicol Pharmacol, 2015. **168**: p. 1.
156. Nwani, C.D., et al., *DNA damage and oxidative stress modulatory effects of glyphosate-based herbicide in freshwater fish, Channa punctatus.* Environ Toxicol Pharmacol, 2013. **36**(2): p. 539-47.
157. Vera-Candioti, J., S. Soloneski, and M.L. Larramendy, *Evaluation of the genotoxic and cytotoxic effects of glyphosate-based herbicides in the ten spotted live-bearer fish Cnesterodon decemmaculatus (Jenyns, 1842).* Ecotoxicol Environ Saf, 2013. **89**: p. 166-73.
158. Poletta, G.L., et al., *Genetic, enzymatic and developmental alterations observed in Caiman latirostris exposed in ovo to pesticide formulations and mixtures in an*

experiment simulating environmental exposure. Ecotoxicol Environ Saf, 2011. **74**(4): p. 852-9.

159. Poletta, G.L., et al., *Genotoxicity of the herbicide formulation Roundup (glyphosate) in broad-snouted caiman (Caiman latirostris) evidenced by the Comet assay and the Micronucleus test.* Mutat Res, 2009. **672**(2): p. 95-102.
160. Clements, C., S. Ralph, and M. Petras, *Genotoxicity of select herbicides in Rana catesbeiana tadpoles using the alkaline single-cell gel DNA electrophoresis (comet) assay.* Environ Mol Mutagen, 1997. **29**(3): p. 277-288.
161. Meza-Joya, F.L., M.P. Ramirez-Pinilla, and J.L. Fuentes-Lorenzo, *Toxic, cytotoxic, and genotoxic effects of a glyphosate formulation (Roundup(R)SL-Cosmoflux(R)411F) in the direct-developing frog Eleutherodactylus johnstonei.* Environ Mol Mutagen, 2013. **54**(5): p. 362-73.
162. Yadav, S.S., et al., *Toxic and genotoxic effects of Roundup on tadpoles of the Indian skittering frog (Euflictis cyanophlyctis) in the presence and absence of predator stress.* Aquat Toxicol, 2013. **132-133**: p. 1-8.
163. dos Santos, K.C. and C.B. Martinez, *Genotoxic and biochemical effects of atrazine and Roundup((R)), alone and in combination, on the Asian clam Corbicula fluminea.* Ecotoxicol Environ Saf, 2014. **100**: p. 7-14.
164. Conners, D.E. and M.C. Black, *Evaluation of Lethality and Genotoxicity in the Freshwater Mussel Utterbackia imbecillis (Bivalvia: Unionidae) Exposed Singly and in Combination to Chemicals Used in Lawn Care.* Archives of Environmental Contamination and Toxicology, 2004. **46**(3).
165. Mohamed, A.H., *Sublethal toxicity of Roundup to immunological and molecular aspects of Biomphalaria alexandrina to Schistosoma mansoni infection.* Ecotoxicol Environ Saf, 2011. **74**(4): p. 754-60.
166. Muangphra, P., W. Kwankua, and R. Gooneratne, *Genotoxic effects of glyphosate or paraquat on earthworm coelomocytes.* Environ Toxicol, 2014. **29**(6): p. 612-20.
167. Piola, L., et al., *Comparative toxicity of two glyphosate-based formulations to Eisenia andrei under laboratory conditions.* Chemosphere, 2013. **91**(4): p. 545-51.
168. Kale, P.G., et al., *Mutagenicity testing of nine herbicides and pesticides currently used in agriculture.* Environ Mol Mutagen, 1995. **25**(2): p. 148-153.
169. Chen, L., et al., *The combined effects of UV-B radiation and herbicides on photosynthesis, antioxidant enzymes and DNA damage in two bloom-forming cyanobacteria.* Ecotoxicol Environ Saf, 2012. **80**: p. 224-30.
170. Wang, G., et al., *Damage to DNA caused by UV-B radiation in the desert cyanobacterium Scytonema javanicum and the effects of exogenous chemicals on the process.* Chemosphere, 2012. **88**(4): p. 413-7.
171. Alvarez-Moya, C., et al., *Evaluation of genetic damage induced by glyphosate isopropylamine salt using Tradescantia bioassays.* Genet.Mol Biol, 2011. **34**(1): p. 127-130.
172. Williams, G.M., R. Kroes, and I.C. Munro, *Safety evaluation and risk assessment of the herbicide Roundup and its active ingredient, glyphosate, for humans.* Regul Toxicol Pharmacol, 2000. **31**(2 Pt 1): p. 117-65.

173. Moriya, M., et al., *Further mutagenicity studies on pesticides in bacterial reversion assay systems.* Mutat Res, 1983. **116**(3-4): p. 185-216.
174. Kier, L.D. and D.J. Kirkland, *Review of genotoxicity studies of glyphosate and glyphosate-based formulations.* Crit Rev Toxicol, 2013. **43**(4): p. 283-315.
175. Lioi, M.B., et al., *Cytogenetic damage and induction of pro-oxidant state in human lymphocytes exposed in vitro to gliphosate, vinclozolin, atrazine, and DPX-E9636.* Environ Mol Mutagen, 1998. **32**(1): p. 39-46.
176. Chruscielska, K.B., J.; Kita, K, et al., *Glyphosate: Evaluation of chronic activity and possible far - reaching effects - Part 2. Studies on Mutagenic Activity.* Pestycydy, 2000. **3-4**: p. 5.
177. Ghisi Nde, C., E.C. de Oliveira, and A.J. Prioli, *Does exposure to glyphosate lead to an increase in the micronuclei frequency? A systematic and meta-analytic review.* Chemosphere, 2016. **145**: p. 42-54.
178. Aghadavod, E., et al., *Role of Oxidative Stress and Inflammatory Factors in Diabetic Kidney Disease.* Iran J Kidney Dis, 2016. **10**(6): p. 337-343.
179. Kamceva, G., et al., *Cigarette Smoking and Oxidative Stress in Patients with Coronary Artery Disease.* Open Access Maced J Med Sci, 2016. **4**(4): p. 636-640.
180. Qureshi, M.A., Y.O. Kim, and D. Schuppan, *Hepatocellular carcinoma in nonalcoholic fatty liver disease: A link between oxidative stress and T-cell suppression.* Hepatology, 2016. **64**(5): p. 1794-1797.
181. Sayanthooran, S., et al., *Upregulation of Oxidative Stress Related Genes in a Chronic Kidney Disease Attributed to Specific Geographical Locations of Sri Lanka.* Biomed Res Int, 2016. **2016**: p. 7546265.
182. Turkmen, K., *Inflammation, oxidative stress, apoptosis, and autophagy in diabetes mellitus and diabetic kidney disease: the Four Horsemen of the Apocalypse.* Int Urol Nephrol, 2016.
183. Vakonaki, E., et al., *Complex interplay of DNA damage, DNA repair genes, and oxidative stress in coronary artery disease.* Anatol J Cardiol, 2016. **16**(12): p. 939.
184. Hecht, F., et al., *The role of oxidative stress on breast cancer development and therapy.* Tumour Biol, 2016. **37**(4): p. 4281-91.
185. Li, L. and F. Chen, *Oxidative stress, epigenetics, and cancer stem cells in arsenic carcinogenesis and prevention.* Curr Pharmacol Rep, 2016. **2**(2): p. 57-63.
186. Perse, M., *Oxidative stress in the pathogenesis of colorectal cancer: cause or consequence?* Biomed Res Int, 2013. **2013**: p. 725710.
187. Prasad, S., et al., *Oxidative Stress and Cancer: Advances and Challenges.* Oxid Med Cell Longev, 2016. **2016**: p. 5010423.
188. Toyokuni, S., *Oxidative stress as an iceberg in carcinogenesis and cancer biology.* Arch Biochem Biophys, 2016. **595**: p. 46-9.
189. Kwiatkowska, M., B. Huras, and B. Bukowska, *The effect of metabolites and impurities of glyphosate on human erythrocytes (in vitro).* Pestic Biochem Physiol, 2014. **109**: p. 34-43.
190. Chaufan, G., I. Coalova, and C. Rios de Molina Mdel, *Glyphosate commercial formulation causes cytotoxicity, oxidative effects, and apoptosis on human cells: differences with its active ingredient.* Int J Toxicol, 2014. **33**(1): p. 29-38.

191. Coalova, I., C. Rios de Molina Mdel, and G. Chaufan, *Influence of the spray adjuvant on the toxicity effects of a glyphosate formulation.* Toxicol In Vitro, 2014. **28**(7): p. 1306-11.
192. Gehin, A., et al., *Vitamins C and E reverse effect of herbicide-induced toxicity on human epidermal cells HaCaT: a biochemometric approach.* Int J Pharm, 2005. **288**(2): p. 219-26.
193. Elie-Caille, C., et al., *Morphological damages of a glyphosate-treated human keratinocyte cell line revealed by a micro- to nanoscale microscopic investigation.* Cell Biol Toxicol, 2010. **26**(4): p. 331-9.
194. Bonini, M.G., et al., *The oxidation of 2',7'-dichlorofluorescin to reactive oxygen species: a self-fulfilling prophesy?* Free Radic Biol Med, 2006. **40**(6): p. 968-75.
195. Kalyanaraman, B., et al., *Measuring reactive oxygen and nitrogen species with fluorescent probes: challenges and limitations.* Free Radic Biol Med, 2012. **52**(1): p. 1-6.
196. George, J. and Y. Shukla, *Emptying of Intracellular Calcium Pool and Oxidative Stress Imbalance Are Associated with the Glyphosate-Induced Proliferation in Human Skin Keratinocytes HaCaT Cells.* ISRN Dermatol, 2013. **2013**: p. 825180.
197. Valavanidis, A., T. Vlachogianni, and C. Fiotakis, *8-hydroxy-2' -deoxyguanosine (8-OHdG): A critical biomarker of oxidative stress and carcinogenesis.* J Environ Sci Health C Environ Carcinog Ecotoxicol Rev, 2009. **27**(2): p. 120-39.
198. Jasper, R., et al., *Evaluation of biochemical, hematological and oxidative parameters in mice exposed to the herbicide glyphosate-Roundup((R)).* Interdiscip Toxicol, 2012. **5**(3): p. 133-40.
199. Astiz, M., M.J. de Alaniz, and C.A. Marra, *Antioxidant defense system in rats simultaneously intoxicated with agrochemicals.* Environ Toxicol Pharmacol, 2009. **28**(3): p. 465-73.
200. Astiz, M., M.J. de Alaniz, and C.A. Marra, *The oxidative damage and inflammation caused by pesticides are reverted by lipoic acid in rat brain.* Neurochem Int, 2012. **61**(7): p. 1231-41.
201. Cattani, D., et al., *Mechanisms underlying the neurotoxicity induced by glyphosate-based herbicide in immature rat hippocampus: involvement of glutamate excitotoxicity.* Toxicology, 2014. **320**: p. 34-45.
202. Slaninova, A., et al., *A review: oxidative stress in fish induced by pesticides.* Neuro.Endocrinol Lett, 2009. **30 Suppl 1**: p. 2-12.
203. Cattaneo, R., et al., *Toxicological responses of Cyprinus carpio exposed to a commercial formulation containing glyphosate.* Bull Environ Contam Toxicol, 2011. **87**(6): p. 597-602.
204. Costa, M.J., et al., *Oxidative stress biomarkers and heart function in bullfrog tadpoles exposed to Roundup Original.* Ecotoxicology, 2008. **17**(3): p. 153-63.
205. de Menezes, C.C., et al., *Roundup effects on oxidative stress parameters and recovery pattern of Rhamdia quelen.* Arch Environ Contam Toxicol, 2011. **60**(4): p. 665-71.
206. Ferreira, D., et al., *Assessment of oxidative stress in Rhamdia quelen exposed to agrichemicals.* Chemosphere, 2010. **79**(9): p. 914-21.

207. Geret, F., et al., *Effects of low-dose exposure to pesticide mixture on physiological responses of the Pacific oyster, Crassostrea gigas.* Environ Toxicol, 2013. **28**(12): p. 689-99.
208. Glusczak, L., et al., *Acute exposure to glyphosate herbicide affects oxidative parameters in piava (Leporinus obtusidens).* Arch Environ Contam Toxicol, 2011. **61**(4): p. 624-30.
209. Guilherme, S., et al., *DNA damage in fish (Anguilla anguilla) exposed to a glyphosate-based herbicide -- elucidation of organ-specificity and the role of oxidative stress.* Mutat Res, 2012. **743**(1-2): p. 1-9.
210. Lushchak, O.V., et al., *Low toxic herbicide Roundup induces mild oxidative stress in goldfish tissues.* Chemosphere, 2009. **76**(7): p. 932-7.
211. Modesto, K.A. and C.B. Martinez, *Roundup causes oxidative stress in liver and inhibits acetylcholinesterase in muscle and brain of the fish Prochilodus lineatus.* Chemosphere, 2010. **78**(3): p. 294-9.
212. Ortiz-Ordonez, E., et al., *Effect of Yerbimat herbicide on lipid peroxidation, catalase activity, and histological damage in gills and liver of the freshwater fish Goodea atripinnis.* Arch Environ Contam Toxicol, 2011. **61**(3): p. 443-52.
213. Sinhorin, V.D., et al., *Effects of the acute exposition to glyphosate-based herbicide on oxidative stress parameters and antioxidant responses in a hybrid Amazon fish surubim (Pseudoplatystoma sp).* Ecotoxicol Environ Saf, 2014. **106**: p. 181-7.
214. Uren Webster, T.M., et al., *Effects of glyphosate and its formulation, roundup, on reproduction in zebrafish (Danio rerio).* Environ Sci Technol, 2014. **48**(2): p. 1271-9.
215. Adamson, P., et al., *Time trends in the registration of Hodgkin and non-Hodgkin lymphomas in Europe.* Eur J Cancer, 2007. **43**(2): p. 391-401.
216. Engels, E.A., *Infectious agents as causes of non-Hodgkin lymphoma.* Cancer Epidemiol Biomarkers Prev, 2007. **16**(3): p. 401-4.
217. Hennessy, B.T., E.O. Hanrahan, and P.A. Daly, *Non-Hodgkin lymphoma: an update.* Lancet Oncol, 2004. **5**(6): p. 341-53.
218. Muller, A.M., et al., *Epidemiology of non-Hodgkin's lymphoma (NHL): trends, geographic distribution, and etiology.* Ann Hematol, 2005. **84**(1): p. 1-12.
219. Acquavella, J.F., et al., *Glyphosate Biomonitoring for Farmers and Their Families: Results from the Farm Family Exposure Study.* Environmental Health Perspectives, 2003. **112**(3): p. 321-326.
220. Curwin, B.D., et al., *Urinary pesticide concentrations among children, mothers and fathers living in farm and non-farm households in iowa.* Ann Occup Hyg, 2007. **51**(1): p. 53-65.
221. Wester, R.C., et al., *Glyphosate skin binding, absorption, residual tissue distribution, and skin decontamination.* Fundam.Appl Toxicol, 1991. **16**(4): p. 725-732.
222. Wester, R.C., D. Quan, and H.I. Maibach, *In vitro percutaneous absorption of model compounds glyphosate and malathion from cotton fabric into and through human skin.* Food Chem Toxicol, 1996. **34**(8): p. 731-735.

223. Zouaoui, K., et al., *Determination of glyphosate and AMPA in blood and urine from humans: about 13 cases of acute intoxication.* Forensic Sci Int, 2013. **226**(1-3): p. e20-5.
224. Brewster, D.W., J. Warren, and W.E. Hopkins, *Metabolism of glyphosate in Sprague-Dawley rats: tissue distribution, identification, and quantitation of glyphosate-derived materials following a single oral dose.* Fundam.Appl Toxicol, 1991. **17**(1): p. 43-51.
225. Begley, D.A., et al., *Finding mouse models of human lymphomas and leukemia's using the Jackson laboratory mouse tumor biology database.* Exp Mol Pathol, 2015. **99**(3): p. 533-6.
226. Hori, M., et al., *Non-Hodgkin lymphomas of mice.* Blood Cells Mol Dis, 2001. **27**(1): p. 217-22.
227. Morse, H.C., 3rd, J.M. Ward, and M.A. Teitell, *Mouse models of human B lymphoid neoplasms*, in *The Lymphoid Neoplasms*, I.T. Magrath, Editor. 2010, CRC Press: Boca Raton, FL.
228. Pattengale, P.K. and C.R. Taylor, *Experimental models of lymphoproliferative disease. The mouse as a model for human non-Hodgkin's lymphomas and related leukemias.* Am J Pathol, 1983. **113**(2): p. 237-65.
229. Ward, J.M., *Lymphomas and leukemias in mice.* Exp Toxicol Pathol, 2006. **57**(5-6): p. 377-81.
230. German Federal Institute for Occupational Safety and Health, *Proposal for Harmonized Classification and Labeling: Glyphosate*, F.O.f. Chemicals, Editor, 2016: Dortmund, Germany.

Additional Reliance Materials

1. Ahsan, N., et al., *Glyphosate-induced oxidative stress in rice leaves revealed by proteomic approach*. Plant Physiol. Biochem., 2008. 46(12): p. 1062-70.
2. Anadon, A., et al., *Toxicokinetics of glyphosate and its metabolite aminomethyl phosphonic acid in rats*. Toxicol. Lett., 2009. 190(1): p. 91-5.
3. Baer, K.N. and B.J. Marcel, *Glyphosate*. 2014: p. 767-769.
4. Bernal, J., et al., *Development and validation of a liquid chromatography-fluorescence-mass spectrometry method to measure glyphosate and aminomethylphosphonic acid in rat plasma*. J Chromatogr. B Analyt. Technol. Biomed. Life. Sci., 2010. 878(31): p. 3290-6.
5. Brusick, D., et al., *Genotoxicity Expert Panel review: weight of evidence evaluation of the genotoxicity of glyphosate, glyphosate-based formulations, and aminomethylphosphonic acid*. Crit. Rev. Toxicol., 2016. 46(sup1): p. 56-74.
6. De Roos, A.J., et al., *Glyphosate Results Revisited: De Roos et al*. Respond. Environ. Health Perspect, 2005. 113(6): p. A366-A367.
7. Devesa, S.S. and T. Fears, *Non-Hodgkin's lymphoma time trends: United States and international data*. Cancer Res, 1992. 52(19 Suppl): p. 5432s-5440s.
8. EPA, *Memo on the Second Peer Review of Glyphosate*, October 30, 1991, From: William Dykstra and George Ghali, To: Robert Taylor
9. EPA, *Memo: Peer Review of Glyphosate*, June 5, 1991, From: William Dykstra, to: Esther Rind
10. Farmer, D.R., T.L. Lash, and J.F. Acquavella, *Glyphosate results revisited*. Environ. Health Perspect., 2005. 113(6): p. A365-A366.
11. Hartge, P. and S.S. Devesa, *Quantification of the impact of known risk factors on time trends in non-Hodgkin's lymphoma incidence*. Cancer Res., 1992. 52(19 Suppl): p. 5566s-5569s.
12. Hoppin, J.A., et al., *Accuracy of self-reported pesticide use duration information from licensed pesticide applicators in the Agricultural Health Study*. J. Expo. Anal. Environ. Epidemiol., 2002. 12(5): p. 313-8.
13. Larsen, K., et al., *Effects of Sub-lethal Exposure to a Glyphosate-Based Herbicide Formulation on Metabolic Activities of Different Xenobiotic-Metabolizing Enzymes in Rats*. Int. J. Toxicol., 2014. 33(4): p. 307-318.
14. Mink, P.J., et al., *Epidemiologic studies of glyphosate and cancer: a review*. Regul. Toxicol. Pharmacol., 2012. 63(3): p. 440-52.
15. Morse, H.C., 3rd, et al., *B lymphoid neoplasms of mice: characteristics of naturally occurring and engineered diseases and relationships to human disorders*. Adv Immunol, 2003. 81: p. 97-121.
16. Morse, H.C., 3rd, et al., *Bethesda proposals for classification of lymphoid neoplasms in mice*. Blood, 2002. 100(1): p. 246-58.
17. Presutti, R., et al., *Pesticide exposures and the risk of multiple myeloma in men: An analysis of the North American Pooled Project*. Int J Cancer, 2016. 139(8): p. 1703-14.

18. Seralini, G.E., et al., *Long term toxicity of a Roundup herbicide and a Roundup-tolerant genetically modified maize*. Food Chem Toxicol, 2012. 50(11): p. 4221-31.
19. Williams, G.M., et al., *A review of the carcinogenic potential of glyphosate by four independent expert panels and comparison to the IARC assessment*. Crit Rev Toxicol, 2016. 46(sup1): p. 3-20.
20. Acquavella, J., et al., *Comments on Charge Questions for Consideration by EPA FIFRA SAP: Review of Glyphosate, Presentation*, 2016.
21. Portier, C., *IARC Monograph Review Process and Glyphosate: Cancer by Glyphosate - how dangerous is the herbicide?* Deutscher Bundestag, Berlin, June 6, 2015
22. Pahwa, et al., *The North American Pooled Project (NAPP): Pooled analysis of case-control studies of pesticides and agricultural expostures, lympohematopoietic cancers and sarcoma*. Occup. Environ. Med., 2014. 41: A116
23. van Burgsteden, J. A., *In vitro percutaneous absorption study with [14C]glyphosate using viable rat skin membranes*, TNO Nutrition and Food Research, 2002.
24. Williams, G.M., et al., *Expert Panel Reivew of the Carcinogenic Potential of the Herbicide Glyphosate*, Poster Presentation, 2015.
25. Parry, J., *Evaluation of the potential genotoxicity of Glyphosate, Glyphosate mixtures and component surfactants, Centre for Molecular Genetics Toxicology*, School of Biological Sciences, University of Wales Swansea, 1999.
26. *EPA, Glyphosate: Evaluation of Carcinogenic Potential* - Charge to FIFRA SAP for October 18-21, 2016 Meeting, 2016.
27. Portier, C., *Carcinogenicity of Glyphosate: A Systematic Review of the Available Evidence*, Presentation to EChA, November 21, 2016.
28. Lan, et al., *Genetic polymorphisms in the oxidative stress pathway and susceptibility to non-Hodgkin Lymphoma*, Hum. Genet., 2007. 121:161-168
29. Lioi, et al., *Cytogenetic Damage and Induction of Pro-Oxidant State in Human Lymphocytes Exposed In Vitro to Gliphosate, Vinclozolin, Atrazine, and DPX-E9636*, Env. And Molecular Mutagenesis, 1998. 32:39-46.
30. Lei Luo, Fei Wang, Yiyuan Zhang, Ming Zeng, Caigao Zhong & Fang Xiao (2017), *In vitro cytotoxicity assessment of roundup (glyphosate) in L-02 hepatocytes, Journal of Environmental Science and Health, Part B*, DOI: *10.1080/03601234.2017.1293449*
31. Hannah, L. H., *Two-year Oral Toxicity Study with CP 67573 in albino rats*, BTL-71-32; IBT No. B564, 1974.
32. International Research and Development Corporation, *Teratology Study in Rabbits*, 1980.
33. Response to EPA Memo, and further Author Unknown (date unknown) Response to EPA memo (Lacayo, Memorandum: Use of historical data in determining the weight of evidence from kidney tumor incidence in the Glyphosate two-year feeding study and some remarks on false positives, S.M.S. Branch, Editor. 1985, US EPA: Washington, DCC), Monsanto.

34. Serdy, Frank S. and Monsanto. Roundup Herbicide EPA Reg. Nos. 524-308, 524-330, 524-339, 524-332, 524-343: Chronic Mouse Study with Glyphosate. Letter to Douglas Campt US EPA. 13 Mar. 1985.
35. Sher, S. P. and Merck Institute for Therapeutic Research. Control data on CD-1 mouse: Enclosure, Letter to David McFadden, Monsanto Company. 22 Mar. 1985
36. Street, Robert W. and Monsanto, Roundup Herbicide EPA Reg. No. 524-308, Additional Information Relating to Chronic Mouse Study, BD-77-420. Letter to Director, Office of Pesticide Programs and US EPA. 20 Mar. 1984.
37. Serdy, Frank S. and Monsanto. Roundup Herbicide EPA Reg. Nos. 524-308, 524-330, 524-339, 524-332, 524-343: Chronic Mouse Study with Glyphosate. Letter to Robert Taylor US EPA. 21 May 1985.
38. Lacayo, H., Memorandum: *Preliminary [sic] Risk Assessment for Glyphosate* - Draft, Toxicology Branch/HED. 1985, US EPA.
39. Long, Timothy, J. Memorandum: *Addendum to Chronic Feeding Study with Glyphosate in Mice BD-77-420*, Dept. of Med. and Env. Health. 1985, Monsanto.
40. Serdy, Frank S. Glyphosate mouse study: Additional Steps. Letter to Tim Long. 28 Aug. 1985.
41. Street, Robert W. and Monsanto, Roundup Herbicide. Letter to Director, Office of Pesticide Programs and US EPA. 21 Aug. 1985.
42. Holden, L. R., *Examination of Crump's Multistage Model*. Memorandum to E. E. Debus. 24 Jul. 1985, Monsanto.
43. Long, Timothy, J. *Possible Response to Press Release of EPA Internal Document on Glyphosate* - Revised Draft. 26 Mar. 1985, Monsanto.
44. Serdy, Frank S. and Monsanto. Roundup Herbicide EPA Reg. No. 524-308, Chronic Mouse Study with Glyphosate. Letter to Director, Office of Pesticide Progams and US EPA. 26 Mar. 1985.
45. *EPA, Memorandum: Consensus Review of Glyphosate Casewell No. 661A*, Office of Pesticides and Toxic Substances. 4 Mar. 1985.
46. Johannsen, F.R. [No Subject]. Memorandum to R. W. Street. 1 Mar. 1985, Monsanto.
47. Gingerich, Lyle L. EPA Toxicology Branch/ Roundup, Letter to T. F. Armstrong, et al., 22 Feb. 1985, Monsanto.
48. Uelner, A. F. IBT B564 - BTL 71-32; Two-Year Chronic Oral Toxicity Study with CP 67573 in Albino Rats, Letter to T. J. Long, 8 Sept. 1986. Monsanto.
49. Pathco Inc. *Lifetime Feeding in Mice, 77-420*, Report, 10 Oct. 1985.
50. Townsend, M., Peck, C., Meng, W., Heaton, M., Robison, R., O'Neill, K., *Evaluation of various glyphosate concentrations on DNA damage in human Raji cells and its impact on cytotoxicity*, Regulatory Toxicology and Pharmacology (2017), doi: 10.1016/j.yrtph.2017.02.002.
51. Wang, S., et al., *Polymorphisms in oxidative stress genes and risk for non-Hodgkin lymphoma*, Carcinogenesis, 2006. 27(9): p. 1828-1834.
52. Acquavella, J. Review: Comments of Chris J. Portier, PhD., USEPA (EPA-HQ_OPP-2016-0385-0094); Glyphosate Issue Paper: Evaluation of Carcinogenic Potential. 20 Nov. 2016.

53. Oleson, R. *Dominant lethal mutagenicity assay with technical glyphosate in mice*. Memorandum to W. D. Carpenter. 24 Apr. 1980, Monsanto.
54. Li, A. P., *In Vivo Bone Marrow Cytogenetics Study of Glyphosate in Sprague-Dawley Rats*, Monsanto Company Environmental Health Laboratory, 20 Oct. 1983.
55. Dirks R. C., Study Summary: *The Hepatocyte Primary Culture/DNA Repair Assay on Compound JJN-1020 (Glyphosate) Using Rat Hepatocytes in Culture*. 21 Oct 1983. Monsanto.
56. Stout, L..D. and Johnson, C. *90 Day Study of Glyphosate Administered in Feed to Sprague/Dawley Rats*. Monsanto Environmental Health Laboratory. 30 Nov. 1987.
57. Reyna, M and Reucker, F. A. *Twelve Month Study of Glyphosate Administered by Gelatin Capsule to Beagle Dogs*. Monsanto Environmental Health Laboratory. 22 Aug. 1985.
58. Shirasu, Y., et al. *The report of mutagenic study with bacteria for CP67573.* Institute of Environmental Toxicology. 20 Jul. 1978.
59. Li, A. P., *CHO/HGPRT Gene Mutation Assay with Glyphosate*, Monsanto Company Environmental Health Laboratory, 20 Oct. 1983.
60. Li, A. P., *Effects of Glyphosate on Rat Bone Marrow Cells*, Monsanto Company Environmental Health Laboratory, 21 Oct. 1983.
61. Haas, M. C. *Immunotoxicity Study in Female B6C3F1 Mice*. WIL Research Laboratory. 21 Mar. 2012.
62. Reagan, E. L. and Laveglia, J. *Volume 3: Primary Irritation Study of Glyphosate*, Food and Drug Research Laboratories. 8 June 1988.
63. Reagan, E. L. and Laveglia, J. *Acute Oral Toxicity Study of Glyphosate Batch/Lot/NBR No. XLI-55 in Sprague-Dawley Rats*, Food and Drug Research Laboratories. 8 June 1988.
64. BDN-77-428, *Acute Oral Toxicity Study in Rats*, 6 Aug. 1979. Bio/dynamics Laboratory.
65. BDN-77-428, *Primary Dermal Irritation Study in Rabbits*. 6 Aug. 1979. Bio/dynamics Laboratory.
66. BDN-77-419, *A Three Month Feeding Study of Glyphosate (Roundup Technical) in Mice*. 31 Dec. 1979. Bio/dynamics Laboratory.
67. BDN-77-418, *A Four Week Pilot Study with Glyphosate in Mice*. 29 Aug. 1978. Bio/dynamics Laboratory.
68. BDN-77-417, *A Three Generation Reproduction Study in Rats with Glyphosate*. 31 Mar. 1981. Bio/dynamics Laboratory.
69. Reyna, M. S. and Thake, D. C., *Rangefinding Study in Glyphosate Administered in Feed to Sprague-Dawley Rats*, Monsanto Company Environmental Health Laboratories. 10 Apr. 1989.
70. BDN-88-114, *Acute Oral Toxicity in Rats*, 20 Sept. 1988. Bio/dynamics Laboratory.
71. IR-79-016, *Teratology Study in Rats*, 21 Mar. 1980. International Research and Development Corporation.

72. Kier, L., *Final Report on Salmonella Mutagenicity Assay of Glyphosate*. 16 June 1978. Monsanto Company Environmental Health Laboratory.
73. van de Waart, E. J., Report: *Evaluation of the Ability of Glyfosaat to Induce Chromosome Aberrations in Cultured Peripheral Human Lymphocytes (with Independent Repeat)*. 1995 June 30. Notox.
74. Martens, M., Study Summary. MON-297-05.
75. Martens, M., Study Summary. MON-298-09-05.
76. Martens, M., Study Summary. MON-MA-81-349-09-05.
77. Martens, M., Study Summary. MON-ML-83-218-09-05.
78. Martens, M., Study Summary. MON-MSL-309-09-05.
79. Martens, M., Study Summary. MON-MSL-7206-09-05.
80. Martens, M., Study Summary. MON-MSL-7215-09-05.
81. Martens, M., Study Summary. SYN-CTL-P-4940-09-05.
82. Martens, M., Study Summary. SYN-CTL-P-4942-09-05.
83. Martens, M., Study Summary. SYN-CTL-P-4943-09-05.
84. Martens, M., Study Summary. SYN-CTL-P-4944-09-05.
85. Martens, M., Study Summary. SYN-CTL-P-5058-09-05.
86. Martens, M., Study Summary. JMPR-2a1-09-05.
87. Martens, M., Study Summary. JMPR-2a2-09-05.
88. Martens, M., Study Summary. JMPR-2a3-09-05.
89. Martens, M., Study Summary. JMPR-2a4-09-05.
90. Martens, M., Study Summary. JMPR-2a5-09-05.
91. Martens, M., Study Summary. JMPR-2a6-09-05.
92. Martens, M., Study Summary. BDN-77-416-09-05.
93. Martens, M., Study Summary. BDN-77-418-09-05.
94. Martens, M., Study Summary. BDN-77-419-09-05.
95. Martens, M., Study Summary. MON-ML-86-351-09-05.
96. Martens, M., Study Summary. MON-ML-88-272-09-05.
97. Martens, M., Study Summary. SYN-CTL-P-1599-09-05.
98. Martens, M., Study Summary. SYN-CTL-P-1802-09-05.
99. Martens, M., Study Summary. SYN-CTL-P-4985-09-05.
100. Martens, M., Study Summary. SYN-CTL-P-5079-09-05.
101. Martens, M., Study Summary. MON-BDN-77-420-09-05.
102. Martens, M., Study Summary. MON-ML-83-137-09-05.
103. Martens, M., Study Summary. MON-ML-87-148-09-05.
104. Martens, M., Study Summary. SYN-CTL-P-5143-09-05.
105. Martens, M., Study Summary. SYN-CTL-PR-1111-09-05.
106. Martens, M., Study Summary. SYN-T-11813-09-05.
107. Holland, M. E., *Results of the Stability Analyses of AMPA (aminomethyl phosphonic acid) Test Material Used in 90-Day Dog Study at WIL Laboratories (WI-90-354)*. 5 Aug. 1994.
108. Holson, J. F., *A Dose Range-Finding Developmental Toxicity Study of AMPA in Rats*. 5 Aug. 1994.
109. Holson, J. F., *A Developmental Toxicity Study of AMPA in Rats*. 5 Aug. 1994.

110. Campbell, S., et al., *AMPA: An Acute Oral Toxicity with the Northern Bobwhite*. 5 Aug. 1994.
111. Long, R. D., et al., *AMPA: A Dietary LC 50 Study with the Northern Bobwhite*. 5 Aug. 1994.
112. Long, R. D., et al., *AMPA: A Dietary LC 50 Study with the Mallard*. 5 Aug. 1994.
113. Schermes, S., et al., *Results of the Analyses of Avian Diet Samples for AMPA (aminomethyl phosphonic acid)*. 5 Aug. 1994.
114. Bowman, J. H., *Acute Toxicity of AMPA to Rainbow Trout (Oncorhychus mykiss)*. 5 Aug. 1994.
115. Holland, M. E., *Results of the Stability Analyses of AMPA in a 96-Hour Acute Study with Rainbow Trout*. 5 Aug. 1994.
116. Burgess, D. and Hicks, S. L., *Acute Toxicity of AMPA to Daphnia magna*. 5. Aug. 1994.
117. Holland, M. E., *Results of the Analyses of AMPA in a 48-Hour Acute Study with Daphnia magna*. 5 Aug. 1994.
118. Long, T. Memorandum: *Aminomethyl Phosphonic Acid (CP 50435): Mutagenicity Studies*. 3 May 1983. Monsanto.
119. Bakke, J. P., *Evaluation of Potential of AMPA to Induce Unscheduled DNA Synthesis in the in vitro Hepatocyte DNA Repair Assay Using the Male F-344 Rat*. 5 Aug. 1994.
120. Kier, L, et al. *Mouse Micronucleus Study of AMPA*. 5 Aug. 1994. Monsanto.
121. Stout, L. D., *One Month Study of AMPA Administered by Capsule to Beagle Dogs*. 5 Aug. 1994.
122. Colvin, L. B., et al., *Final Report on CP 67573 Residue and Metabolism*. Aug. 1973. Monsanto.
123. Tompkins, E. C., *90-Day Oral (Capsule) Toxicity Study in Dogs with AMPA*, 5 Aug. 1994. Monsanto.
124. Deposition of Daniel Goldstein. Monsanto Company. 18 Jan. 2017.
125. Deposition of David Saltmiras, vol. I. Monsanto Company. 31 Jan. 2017.
126. Deposition of David Saltmiras, vol. II. Monsanto Company. 1 Feb. 2017.
127. Deposition of Donna Farmer, vol. I. Monsanto Company. 11 Jan. 2017.
128. Deposition of Donna Farmer, vol. II. Monsanto Company. 12 Jan. 2017.
129. Deposition of William Heydens, vol. I. Monsanto Company. 23 Jan. 2017.
130. Deposition of William Heydens, vol. II. Monsanto Company. 24 Jan. 2017.
131. Deposition of Aaron Blair. Independent. 20 Mar. 2017.

CURRICULUM VITAE

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Education:

1981	Ph.D.	(Biostatistics), University of North Carolina, Chapel Hill
1979	M.S.	(Biostatistics), University of North Carolina, Chapel Hill
1977	B.S.	(Mathematics), summa cum laude, Nicholls State University

Employment:

2013-present	**Consultant** to various governmental agencies (multiple countries)
2013-2014	**Senior Visiting Scientist**, International Agency for Research on Cancer, Lyon, France
2013-present	**Senior Contributing Scientist**, Environmental Defense Fund, New York City, NY
2010-2013	**Director**, National Center for Environment Health, Centers for Disease Control and Prevention, Atlanta, GA
2010-2013	**Director**, Agency for Toxic Substances and Disease Registry, Atlanta, GA
2009 – 2010	**Senior Advisor to the Director**, National Institute of Environmental Health Sciences and National Toxicology Program, Research Triangle Park, North Carolina.
2009 – 2010	**Visiting Scientist**, National Research Centre for Environmental Toxicology (EnTox), Queensland, Australia
2006 - 2009	**Associate Director,** National Institute of Environmental Health Sciences, Research Triangle Park, North Carolina.
2006 - 2009	**Director, Office of Risk Assessment Research,** National Institute of Environmental Health Sciences, Research Triangle Park, North Carolina.
1993 – 2010	**Head, Environmental Systems Biology** (originally Stochastic Modeling), Laboratory of Molecular Toxicology, National Institute of Environmental Health Sciences, Research Triangle Park, North Carolina.
2000 - 2006	**Associate Director, National Toxicology Program**, National Institute of Environmental Health Sciences, Division of Intramural Research, Research Triangle Park, North Carolina.
2000 - 2006	**Director, Environmental Toxicology Program**, National Institute of Environmental Health Sciences, Division of Intramural Research, Research Triangle Park, North Carolina.
2006-2007	**Scientific Advisor to the Director**, Public Health and the Environment Department, World Health Organization, Geneva, Switzerland (detail from NIEHS – four months)
1993 - 2005	**Chief, Laboratory of Computational Biology and Risk Analysis** (originally the Laboratory of Quantitative and Computational Biology), National Institute of Environmental Health Sciences, Division of Intramural Research, Research Triangle Park, North Carolina.

1996 - 2000	**Associate Director for Risk Assessment**, Environmental Toxicology Program National Institute of Environmental Health Sciences, Division of Intramural Research, Research Triangle Park, North Carolina.
1990 - 1993	**Head, Risk Methodology Section**, National Institute of Environmental Health Sciences, Division of Biometry and Risk Assessment, Research Triangle Park, North Carolina.
1987, 1992, 1990	**Guest Scientist**, German Cancer Research Center, Heidelberg, Germany.
1978 - 1990	**Mathematical Statistician**, National Institute of Environmental Health Sciences, Division of Biometry and Risk Assessment, Research Triangle Park, North Carolina.
1977	**Mathematician**, Computer Sciences Division, Oak Ridge National Laboratory, Oak Ridge, Tennessee.
1976	**Undergraduate Research Trainee**, Neutron Physics Division, Oak Ridge National Laboratory, Oak Ridge, Tennessee.

University Affiliations:

2014 – present	Visiting Professor, Department of Toxicogenomics, Maastricht University, The Netherlands
2013 – present	Honorary Professor, National Research Centre for Environmental Toxicology, University of Queensland, Brisbane, Australia
2011 – present	Adjunct Professor, Department of Environmental Health, Emory University, Atlanta, GA, USA
2009 – 2010	Visiting Professor, University of Queensland, Brisbane, Australia
1986 - 2007	Adjunct Professor of Biostatistics, University of North Carolina, School of Public Health, Chapel Hill, North Carolina.
1990-1992	Adjunct Professor of Statistics, University of Waterloo, Waterloo, Ontario, Canada

Honors & Awards:

- 2013 President's Dream Green Team Award for "A Human Health Perspective on Climate Change"
- Fellow, World Innovation Foundation, 2006
- Society of Toxicology, Risk Assessment Specialty Section, Paper of the Year, 2006
- Society of Toxicology, Risk Assessment Specialty Section, Paper of the Year, 2005
- Outstanding Risk Practitioner Award, International Society for Risk Analysis, 2000.
- Elected Fellow, International Statistical Institute, 2000.
- Outstanding Performance Award, National Institute of Environmental Health Sciences, numerous dates.
- Commendation for Sustained High Quality Work Performance, National Institute of Environmental Health Sciences, numerous dates.
- Merit Award, National Institute of Health, 1998.
- Board of Publications, Best Paper Award, Society of Toxicology, 1995.
- Distinguished Achievement Award, Section on Statistics and the Environment, American Statistical Association, 1995.
- Spiegelman Award presented by the American Public Health Association to the most outstanding public health statistician under the age of 40, 1995.
- Best-applied statistics paper, Centers for Disease Control, 1993.
- Elected Fellow, American Statistical Association, 1992.

- Elected Foreign Correspondent, Russian National Academy of Natural Sciences, 1992.
- First recipient of the James E. Grizzle Distinguished Alumnus Award, The Department of Biostatistics, The University of North Carolina, 1991.

Professional Societies Membership:

Society of Toxicology, American Public Health Association, International Statistics Institute, Society for Risk Analysis, EcoHealth Society, American Association for Cancer Research, American Association for the Advancement of Science

Editorial Activities:

- Editor in Chief - The Open Environmental Journal (2008 to 2010)
- Associate Editor – Frontiers in Predictive Toxicity (2010 to present)
- Associate Editor - Environmental Health Perspectives (1987-2006)
- Associate Editor - Risk Analysis: An International Journal (1989-2003)
- Editorial Board – Environmental and Ecological Statistics (2004-2007)
- Associate Editor – Statistics in Medicine (1998-2002)
- Associate Editor - Biometrics (1997-99)
- Editorial Board Member/Reviewer (different dates): Biometrika, Cancer Research, Communications in Statistics, Fundamental and Applied Toxicology, Journal of Applied Toxicology, Journal of the American Statistical Association, Journal of Toxicology and Environmental Health, Science, Mathematical Biosciences, Journal of Mathematical Biology, Carcinogenesis, Science, PNAS, Toxicological Sciences

Advisory & Review Committees:

2015 – 2016	Member, Committee to Review the Draft Interagency Report on the Impacts of Climate Change on Human Health in the United States, National Research Council, National Academy of Sciences, USA
2010 – present	Member, Science Advisory Group on Electromagnetic Fields and Health, Netherlands Organisation for Health Research and Development
2009 – 2010	Coordinating Lead Author, Interagency Working Group on Climate Change and Health
2009 – present	Member, Institute of Medicine Roundtable on Environmental Health Sciences Research and Medicine
2009 – 2012	Member, National Academies of Science Roundtable on Science and Technology for Sustainability
2009	Member, WHO Advisory group on the health implications of the use of DDT to reduce risks of malaria.
2005 – 2010	Chair, Subcommittee on Toxics and Risk, President's National Council on Science and Technology
1997 - 2012	Advisor, *World Health Organization*, International Program on Chemical Safety, EMF Project.
2008 – 2010	Member, Environmental Protection Agency, Science Advisory Board
2007 – 2010	Member, International Life Sciences Institute, Health and Environmental Sciences Institute, Subcommittee on Susceptible Populations
2008	Center Review Committee, Canadian National Science and Engineering Research Council Chair in Risk Assessment

2008	Chair, International Agency for Research on Cancer Monographs Advisory Group, Lyon, France
2008	Advisory Group, Center for Environmental Oncology, University of Pittsburgh Cancer Institute
2007.	Chair, WHO Workshop on Low Cost Options for Reducing Exposures to ELF-EMF, Geneva
2007.	Invited Participant, International Program on Chemical Safety Workshop on Aggregate and Cumulative Risk Assessment, Washington, DC.
2006	Rapporteur, International Agency for Research on Cancer, Scientific Advisory Group to Plan Volume 100 of the IARC Monograph Series
2005	Chair, International Agency for Research on Cancer, Scientific Advisory Board on the Preamble to the Cancer Monograph Series
2005	Chair, World Health Organization Expert Panel on Health Criteria Document for Extremely Low Frequency Electric and Magnetic Fields
2003 – 2005	Co-Chair, Subcommittee on Health and Environment, President's National Council on Science and Technology
2003	Ad-Hoc member, EPA Science Advisory Board, Review of Children's Cancer Risk Assessment Supplement to Cancer Guidelines
2002 – 2006	Co-Chair, Subcommittee on Mercury, President's National Council on Science and Technology
2000 – 2007	Member, Finish Academy of Sciences Centers of Excellence Program Science Advisory Committee
2000	Reviewer, *Congressional Research Service, Library of Congress*; Research needs relevant to children's environmental health risks.
1998 - 2004	Member and Chair, *Environmental Protection Agency*, FIFRA Science Advisory Panel.
1997 - 2006	Member, National Occupational Research Agenda Team, *National Institute of Occupational Safety and Health*.
1995 - 2000	Advisor, *Australian Health Council*, Risk Assessment Methodology, Member *NHMRC* Steering Committee on Cancer Risk Assessment Guidelines.
1992 - 2000	Member, *EPA* Dioxin Reassessment Working Group.
1985 - 2007	Thesis director for graduate students, Department of Biostatistics, *University of North Carolina - Chapel Hill, North Carolina*.
1997	Advisor, *Netherlands National Health Council*, Risk Assessment Methodology.
1997	Reviewer, *Air Force Office of Scientific Research*.
1996 - 1997	Temporary Advisor, *World Health Organization*, Expert Committee on Food Additives.
1996	Advisor, *Environmental Protection Agency;* Evaluation of the benchmark dose methodology.
1996	Advisor, *Environmental Protection Agency;* Evaluation of risks from exposure to PCBs.
1996	Expert Review Committee, *Environmental Protection Agency;* Cancer dose-response for PCB's.
1995 - 1996	Member, *California Environmental Protection Agency*, Risk Assessment Advisory Committee.

1994 - 1997	Science Advisory Panel, *Public Broadcasting System Production* "Poisons in the Womb".
1991 - 1995	Ad-Hoc Member, *Environmental Protection Agency*, Science Advisory Panel.

Legislative Hearings:

- Glyphosate Carcinogenicity, European Parliament, Brussels, December 2015
- Glyphosate Carcinogenicity, German Parliament, Berlin, July 2015
- Lead and Children's Health, Senate Committee on Environment and Public Works, July, 2012
- Asthma and Children's Health, Senate Committee on Environment and Public Works, May, 2012
- Contaminated Drywall, Senate Committee on Commerce, Science and Transportation, December, 2012.
- Camp Lejeune Contaminated Drinking Water, House Committee on Science and Technology, September, 2010.
- Autism and Vaccines, House Committee on Government Reform, December, 2002.

US Government Service Activities:

- Member, President's Task Force on Environmental Justice 2010-2013
- Member, President's Task Force on Children's Environmental Health 2009-2013
- Member, National Toxicology Program Executive Committee 2010-2013
- Financial Support and International Press Conference for research on "The Health Benefits of Tackling Climate Change" appearing as a series in Lancet, November 25, 2009
- Organizing Committee, White House Stakeholder briefing on Climate Change and Human Health, Old Executive Office Building, November 2009.
- Member, US Delegation, World Climate Congress, Geneva (September 2009)
- Member, US Delegation, Global Risk Communication Dialogue (2008-2009)
- Member, NIEHS Corrective Action Plan Management Committee (2008-2009)
- Primary focus, all interagency activities on hazards and risk (2006 to present)
- Co-Organizer, NIEHS/EPA Workshop on Children's Environmental Health, RTP, NC, January, (2007)
- Co-Organizer, NIEHS/NTP Workshop on the Identification of Targets for the HTS Roadmap Project (2007)
- Coordinator, NIEHS/EPA Review of the Children's Environmental Health Centers Program (2006-2007)
- Organizing Committee, Global Environmental Health Initiative, NIEHS (2006 to 2009)
- NIEHS Leadership Council (2005 to 2009)
- Organizer, formal collaborative agreements between NTP and Ramazzini Foundation (2001 to 2006)
- Organizer, formal collaborative agreements between NTP and Korean NTP (2002 to 2006)
- NIEHS Title 42 Review Committee (2003 to 2004)
- NIEHS Executive Committee and Operations Update Committee (2000 to 2005)
- NIEHS Leadership Retreats, DERT Retreats, DIR Retreats (all years since 1997)
- Presenter, NIEHS-sponsored National Academy of Sciences Committee on Emerging Issues in Environmental Health, November, 2001
- Organizer and presenter, National Toxicology Program Executive Committee Meetings (multiple dates since 2000)
- Organizer and presenter, National Toxicology Program Board of Scientific Counselors (multiple dates since 1998)

- Organizer, Joint NIEHS/US Geological Survey Interagency Program on Exposure Assessment, April 2001 to present)
- Organizer, US-Vietnam Scientific Conference on the Health and Environmental Effects of Agent Orange/Dioxin in Vietnam, March, 2002
- Organizing Committee, National Toxicology Program/EPA/FDA Scientific Conference on the Allergenicity of Genetically Modified Food, November, 2001
- NIEHS Town Hall Meeting, Los Angeles California, November, 2001
- NTP Research Directions, NAEHSC, Research Triangle Park, NC. May, 2001.
- NCI Study Section Center Presite Meeting, Seattle, Washington, January, 2001.
- Program committee member, *NIEHS/Colorado State University* conference on the Application of Technology to Chemical Mixture Research, 2001.
- Coordinating Core Committee, National Center for Toxicogenomics, NIEHS, 2000 to present
- Organizer, Joint US-Vietnam Consultation on Research on Agent Orange Health Effects in Vietnam. Singapore, 2000
- *ICCVAM/NICEATM*, Up-and-Down Procedure Peer Review Meeting, 2000.
- Chairman, *NIEHS* Risk Assessment Research Committee, 1995-present.
- Discussant, *NIEHS/PNNL* Workshop on Human Biology Models for Environmental Health Effects, 2000.
- Risk Assessment Coordinator, *NIEHS* US *RAPID* Program for the Evaluation of Health Risks from Exposure to Electric and Magnetic Fields, 1996-99.
- Organizer and Chair, Four Public Comment Sessions on the report of the *NIEHS/DOE* Working Group on the Health Effects of Exposure to Electric and Magnetic Fields, 1998.
- Organizer and Co-Chair, *NIEHS/DOE* Working Group on the Health Effects of Exposure to Electric and Magnetic Fields, 1998.
- Scientific Organizing Committee, *NIEHS* Workshop on Risk Assessment Issues Associated with Endocrine Disrupting Chemicals, 1998.
- Organizer, *NIEHS/DOE* Science Research Symposium on the Health Effects of Exposure to Electric and Magnetic Fields I: Biophysical Mechanisms and *In Vitro* Experimentation, 1998.
- Organizer, *NIEHS/DOE* Science Research Symposium on the Health Effects of Exposure to Electric and Magnetic Fields II: Epidemiological Findings, 1998.
- Organizer, *NIEHS/DOE* Science Research Symposium on the Health Effects of Exposure to Electric and Magnetic Fields III: *In Vitro* and Clinical Research Findings, 1998.
- Head, Toxicokinetics Faculty, *NIEHS*, 1994-97.
- Coordinator/Director, *NIEHS/ATSDR* Interagency Course on Mechanistic Modeling in Environmental Risk Assessment, 1996.
- Organizer, *NIEHS/EPA* Workshop on Research Priorities for New Risk Assessment Guidelines, 1996.
- Co-Organizer, *National Institute of Statistical Sciences*, *NIEHS/EPA* Workshop on Mechanistic Modeling in Risk Assessment, 1995.
- Scientific Coordinator and Mission Director, *NIEHS* "Mission to Vietnam" to assess the potential for scientific collaboration on the impact of Agent Orange on the Vietnamese Population, 1995.
- Chairman, *NIEHS* Computer Science Focus Group, 1995.
- Discussant, National Toxicology Program Workshop on Mechanistic Modeling in Toxicology, *NIEHS*, 1995.
- Discussant, National Toxicology Program Workshop on Mechanisms of Carcinogenesis, *NIEHS*, 1995.
- Co-Organizer, International Conference on The Role of Cell Proliferation in Carcinogenesis, co-

sponsored by *NIEHS*, *The Chemical Industry Institute of Toxicology*, *The International Life Sciences Institute* and *The American Industrial Health Council*, 1992.

- Organizer and Director, Scientific Basis of Animal Carcinogenicity Testing, Moscow, Russia, co-sponsored by the *International Agency for Research on Cancer*, *NIEHS*, *Health and Welfare Canada* and *The All-Union Cancer Research Center*, 1991.
- Chairman, Computer Technology Advisory Forum, *NIEHS*, 1989.
- Organizer and Director, Design and Analysis of Long-Term Animal Carcinogenicity Experiments, Lyon, France, co-sponsored by the *International Agency for Research on Cancer* and the *NIEHS*, 1988.

Extramural Activities:

- Member, NRC Committee to review the Draft Interagency Report on the Impacts of Climate Change on Human Health in the United States, Washington, DC, 2015
- Expert Scientist, International Agency for Research on Cancer Monograph Meeting on Some Organophosphate Pesticides and Herbicides, Lyon, France, March, 2015
- Overall Chair, International Agency for Research on Cancer Monograph Meeting on Diesel and Gasoline Engine Exhausts and related compounds, Lyon, France, June, 2012
- Advisor to Wellcome Trust at "International Research Futures Symposium on Global Change, Economic Sustainability, and Human Health", London, England, March, 2012.
- Expert Panel Member for review of Hollings Marine Laboratory, National Oceanographic and Atmospheric Agency, Charleston, USA, February, 2012.
- Chair, Mechanism Subgroup, International Agency for Research on Cancer Monograph Meeting on Radiofrequency Electric and Magnetic Fields, Lyon, France, May, 2011
- Advisor, Greek Ministry Health, Working group on hexavalent chromium in the environment, January, 2011
- Member, WHO Consultation on Human Health Risks from DDT, Geneva, Switzerland, November, 2010
- Associate Editor, Frontiers in Predictive Toxicity, 2010 – 2011
- Scientific Advisor, Health Investigation Levels Workshop, Canberra, Australia, January, 2010
- Chair, IARC Working Group, IARC Monograph 100-G, Lyon, France, October, 2009
- Scientific Organizing Committee, VII World Congress on Alternatives and Animal Use in Life Sciences, Rome, Italy, September, 2009
- Chair, Research Directions Working Group, World Health Organization Consultation on Global Research on Climate Change and Health, October, 2008.
- Editor-in-Chief, The Open Environment Journal, May 2008-August, 2010
- Member, EPA Science Advisory Board, July, 2008-present
- Working Group Member, IARC Monograph 98 - Fire-fighting, Painting and Shift-work, Lyon, France, November, 2007
- Chair, WHO Extremely Low Frequency Magnetic and Electric Fields Workshop on Intervention Strategies, June, 2007
- Special Advisor to the Director, Program on Public Health and the Environment, WHO, Geneva, May-July, 2007
- Member, International Life Sciences Institute Working Group on Susceptible Populations, March, 2007 – present
- Special Advisor to the Director, Program on Public Health and the Environment, WHO, Geneva, November, 2006-January, 2007

- Breakout Group Chair, International Workshop on Uncertainty and Variability in PBPK Modeling, RTP, NC USA, October, 2006
- Member, Health Effects Sciences Institute Committee on Sensitive Subpopulations and Groups, Washington, DC, 2006 to present
- Rapporteur, Steering Committee for developing the 100th Monograph of the International Agency for Research on Cancer, Lyon, France, September, 2006
- Co-Organizer, parallel workshops on the advancement of PBPK modeling in risk assessment, Research Triangle Park, November, 2006, Corfu, Greece, April, 2007.
- Organizer, Alternative Models in Developmental Neurotoxicity, Alexandria, Virginia, March, 2006.
- Organizer, NTP High Throughput Screening Workshop, Washington, DC, December, 2005
- Organizer, ISRTP Meeting on Alternative Methods in Toxicology, Baltimore, Maryland, November, 2005
- Organizer, NTP 25th Anniversary Meeting, Washington, DC, May, 2005
- Organizer, IPCS/WHO Workgroup on Dose-Response Modeling, Geneva, Switzerland, September, 2004
- Organizer, Consultation on harmonization of toxicological research between the NTP, Ramazzini Foundation and the European Union, European Congress of Toxicology, Florence, Italy, September, 2003.
- Member, WHO Workgroup on the epidemiology of cellular phone toxicity, Tskuuba, Japan, September, 2003.
- Program Committee, 12th International Conference on Global Warming, Boston, Massachusetts, May 2003
- Program Committee, International Conference on Cancer Risk Assessment, Athens, Greece, August, 2003
- Chair, WHO Public Consultation on Risk Communication, Luxembourg, February, 2003.
- Chair, WHO Committee on Establishing a Plan for Implementation of the Precautionary Principle in Risk Management. Luxembourg, February, 2003.
- Presenter (on behalf of US Government), National Academy of Sciences Panel on the Use of Third Party Toxicity Research with Human Research Participants, December, 2002
- Member, US Science Delegation, United Nations Environmental Program Consultation on Organic Mercury, September, 2002
- Science Panel Member, IARC Carcinogenicity Review of ELF-EMF, Lyon, France, June, 2001.
- Reviewer, Finish Ministry of Health Centers of Excellence Program, Helsinki, April, 2001.
- *EPA* dioxin reassessment peer review workshop and public comment session, Washington, DC, 2000.
- Organizer: Dioxin Dose-Response Working Group Meeting, Fort Collins, Colorado, February, 2000.
- Chair, Spiegelman Award Committee, *American Public Health Association*, 1998.
- Chair, *Bioelectromagnetics Society* Symposium on the use of Transgenic Animals in Evaluating Health Risks from Exposure to Cellular Phones, St. Petersburg, Florida, 1998.
- Member, *World Health Organization* International Program on Chemical Safety, Workshop on Issues in Cancer Risk Assessment, 1998.
- Advisor, *Joint Committee on Food Additives, World Health Organization/Food and Agriculture Organization*. Evaluation of certain food additives and contaminants
- Member, US Government Methylene Chloride Risk Characterization Science Committee, 1996-1998.
- Scientific Organizing Committee, *Colorado State University* Workshop on Biomedical Advances on Chemical Mixtures, 1997.
- *National Academy of Sciences*, Institute of Medicine, Committee on Funding Future Agent Orange

Research in Vietnam, 1996.

- Discussant, Workshop on the role of Endocrine Disruptors in Human Health, 1995.
- Advisor to *Australian Health Council* on Risk Assessment Methodology, Member *NHMRC* Steering Committee on Cancer Risk Assessment Guidelines
- Participant, International Program on Chemical Safety of the *World Health Organization* Workshop on Chemical Risk Assessment, London, England, 1995.
- Participant, *IARC* Workshop on Receptor-Mediated Carcinogenesis, Lyon, France, 1994.
- Co-Organizer, Symposium on Quantitative Risk Assessment, *German Cancer Research Center*, Heidelberg, Germany, 1993.
- Participant, *IARC* Monograph on Risk Assessment Methodology, *International Agency for Research on Cancer*, Lyon, France, 1993.
- Thesis advisor for graduate student, *University of Waterloo*, Waterloo, Ontario, Canada. 1991-93.
- Co-Organizer, *Russian Academy of Sciences* Informatics and Cybernetics Research Award, 1992.
- Official Observer, IARC Monograph on the Biological Effects of Ultraviolet Radiation, *International Agency for Research on Cancer*, Lyon, France, 1992.
- Member, *International Life Sciences Institute*, Dose-Response Working Group, 1991.
- Participant in Banbury Conference on Human Health Risks from Exposures to Dioxins, Banbury Conference Center, Cold Spring Harbor, New York, 1990.
- Co-Chairman, Session on Biostatistical Developments in Cancer Research, *15th International Cancer Congress*, Hamburg, Germany, 1990.
- Participant in *Environmental Protection Agency* Workshop on Risk Assessment Guidelines, Virginia Beach, Virginia, 1989.

Invited Presentations (present-1999)

"Glyphosate Carcinogenicity", Swiss Society of Toxicology, Basel, November, 2016
"Glyphosate Carcinogenicity", Concerned Scientists of Switzerland Annual Meeting, Zurich, December, 2015
"Should the precautionary principle be invoked for RF-EMF", BIOEM 2015, Asilomar, CA, USA, June 2015
"IARC Monograph Review Process and Glyphosate", Deutscher Bundestag, Berlin, Germany, June 2015
"The Exposome: Why does it matter for public health?", Association of Public Health Laboratories, Dr. Katherine Kelly Distinguished Lecture, Indianapolis, USA May 2015
"A bioinformatics/biostatistics approach to systems toxicology", Maastricht University, Maastricht, Netherlands, March, 2015
"Mechanistic Data, Cellular Pathways, and Cancer Classification", 39th Annual Toxicology Forum Meeting, Washington D.C., USA, January, 2015
"Current Issues in Environmental Public Health", Nicholls State University, Thibodaux, LA, April, 2014
"Review of Approaches to the Quantification of Risks", International Agency for Research on Cancer, Lyon, France, November, 2013
"The Gene-Environment-Disease Interactome", Maastricht University, September, 2013
"Toxicogenomics and Electromagnetic Fields", BEMS Annual Meeting, Thesaloniki, Greece, June 2013
"Toxicogenomics and Risk Assessment", International Symposium on Toxicogenomics and Human Health, Paris, France, May, 2013
"Extreme Weather, Climate and Health: Putting Science into Practice", Climate and Health Conference, Washington, DC, February, 2013
"Biofuels and Human Health: CDC's activities", Our Energy Future and Health, National Academy of Sciences, Washington, DC, USA, January, 2013

"Global Environmental Health", Johns Hopkins Bloomberg School of Public Health, Baltimore, MD, USA, January, 2013
"Addressing Superfund Sites", SBRP Annual Meeting, Washington, DC, October, 2012
"The future of cancer risk assessment", 6th NCC International Symposium- Management of Carcinogenic Hazard: Recent Progress and Future Perspectives, Seoul, Korea, June, 2012
"ATSDR Reorganization and New Directions", National Toxicology Program Executive Committee, Washington, DC, May, 2012
"Hydraulic Fracturing, National Concerns", Institute of Medicine Roundtable on Environmental Health, Washington, DC, April, 2012.
"Using Health Impact Assessment to Guide National Policy", National Health Impact Assessment Meeting, Washington, DC, April, 2012
"The Great Debate", Society of Toxicology Meetings, San Francisco, USA, March, 2012.
"CDC and Environmental Health", Society of Toxicology Meetings, San Francisco, USA, March, 2012.
"Keynote Address", National Climate Assessment Initial Workshop, Charleston, SC, February, 2012.
"Integration of Climate, Weather and Health", American Meteorological Society, New Orleans, USA, January, 2012.
"Biomonitoring and Environmental Health", CINVESTAV, Mexico Department of Health, Mexico City, Mexico, January, 2012.
"What is health risk assessment", International Conference on EMF and Health, Brussels, Belgium, November, 2012.
"Risks from multiple chemicals in polluted communities", International Conference on Chemical Mixtures, Wasington, USA, October, 2011.
"Healthy Homes: The Scientific Support", National Healthy Homes Conference, Denver, CO, June, 2011
"Climate Change and Human Health", Keynote Address, Yale School of Public Health Annual Alumni Day, New Haven, Ct, June, 2011
"Environmental Public Health", Keynote Address, International Conference on Sustainable Remediation, Amherst, MA, June, 2011
"The gene-environment disease interactome for humans", Society of Toxicology Annual Meeting, Washington, DC, March, 2011
"Environmental Exposure Science in the 21st Century", NAS Committee on Human and Environmental Exposure Science in the 21st Century, Washington, DC, February, 2011
"Human Health Effects and the Impact of Climate Change on Our Oceans", Annual National Conference on Science, Policy, and the Environment, Washington, Dc, January, 2011
"A Vision for the National Center for Environmental Health and Agency for Toxic Substances and Disease Registry", NAS Committee on Emerging Issues in Environmental Health, Washington, DC, December, 2010
"Predicting Health Risks Using Gene-Expression Data", EPA NextGEN Workshop, Research Triangle Park, NC, October, 2010
"Building Sustainable Environments", NAS Workshop: Pathways to Sustainable Development, Atlanta, GA, September, 2010
"Future Directions in Environmental Public Health", State Environmental Health Directors Annual Meeting, Portland, OR, September 2010
"The gene-environment disease interactome for humans", AACR Workshop on The Future of Molecular Epidemiology, Miami, Fl, June, 2010
"Emerging methods for determining chemical hazards", Keynote Address, Human Health Hazard Indicators Workshop, Sacramento, March, 2010.
"Dose-response and risk assessment considerations of melamine in infants", Society of Toxicology Annual Meetings, Sal Lake City, March, 2010
"The gene-environment disease interactome for humans", University of Queensland Centre for Clinical Research, Brisbane, Australia, February, 2010

"Research Needs for Climate Change and Human Health", Queensland Health and ENTOX, February, 2010

"Using systems biology to develop the gene-environment disease interactome for humans", ENTOX, Brisbane, Australia, December, 2009

"Quantifying the health risks of dioxins and dioxin-like compounds", ENTOX, January, 2010

"Epigenetics and its use in toxicology and risk assessment", Keynote Adress, Australian College of Toxicology and Risk Assessment Annual Meeting, Canberra, December, 2009.

"The changing shape of regulatory toxicology in the United States", Office of Chemical Safety & Environmental Health, Canberra, Australia, December, 2009

"The changing shape of regulatory toxicology in the United States", NICNAS, Sydney, Australia, December, 2009

"Development and Use of the Gene-Environment-Disease Interactome for Humans", 60th Anniversary of the Department of Biostatistics, University of North Carolina, Chapel Hill, NC, October, 2009

"Using systems biology to develop the gene-environment disease interactome for humans", Seventh World Congress on Alternatives and Animal Use in Life Sciences, Rome, Italy, September, 2009

"Use of 'Omics' Technologies to Enhance Risk Assessment", International Workshop on Genomics in Cancer Risk Assessment, Venice, Italy, August, 2009.

"Proteomics, Genomics and the Human Gene-Environment-Disease Interactome", First International Radiation Proteomics Workshop, Munich, Germany, May, 2009.

"Molecular Biology and Regualtory Toxicology", Society of Toxicology, Annual Meeting, Baltimore, Maryland, March, 2009.

"Using Modern Research Tools to Enhance Regulatory Toxicology", SOT Satelite Meeting on Development Of Toxicological And Environmental Public Health Infrastructures In Africa: Understanding The Premise And Mapping The Approach, Baltimore, MD, March 2009.

"Health Implications of Climate Change", Navy and Marine Public Health Conference, Norfolk, Va., March, 2009.

"Climate Change and Minority Health", Second Annual National Conference on Health Disparities, University of the Virgin Islands, St. Croix, December, 2008.

"Climate Change and Human Health", the Environmental Mutagen Society Annual Meetings, Puerto Rico, October, 2008.

"Climate Change and Human Health", The Puerto Rico Chamber of Commerce, October, 2008.

"Using Stem Cells in Environmental Health Research", California Stem Cells and Predictive Toxicology Initiative Workshop, Berkeley, CA, July, 2008.

"Developing the Disease-Environment Interactome", IMBA-FGF Workshop on Developing Omics Technologies to Assess Unclear Risks", Berlin, Germany, May, 2008

"Environmental Systems Biology", 40th Annual Interface Symposium on the interface between computer science and statistics, Durham, NC, May, 2008

"Environmental causes of childhood leukemia and modern environmental health research", Summary Address, ICNIRP/WHO/BS Symposium on Research Advances in Childhood Leukemia, Berlin, Germany, May, 2008.

"Using systems biology as a tool to understand environmental health", Atlantic Coast Symposium on the Mathematical Sciences in Biology and Medicine, Raleigh, NC, April, 2008.

"Biological networks and high-throughput screening", Society of Toxicology, Seattle, Washington, March, 2008.

"Environment and cancer: strategies for identifying new hazards". Workshop on the Environment and Cancer, American Cancer Society, Atlanta, Georgia, USA. January, 2008.

"A systems approach to human health risk assessment", Environmental Health Sciences Decision Making: Risk Management, Evidence and Ethics. US National Academy of Sciences, Washington, DC, USA. January, 2008.

"Finding targets for high-throughput screening linking genetics, genomics, pathways and human disease". NTP Biological Screening Program Seminar Series. January, 2008.

"The forest for the trees: A systems approach to environmental health research". Keynote Address, KTL-DEH Center of Excellence Program, Lamalo, Finland. December, 2007.
"Toxicology for the 21st Century", ECNIS Research Colloquium, Engelheim, Germany, October, 2007.
"Evidence-Based Decision Making in Public Health", First International Workshop on Evidence-Based Toxicology, Cuomo, Italy. October, 2007.
"Genes, Pathways and Diseases", World Health Organization Headquarters, Geneva, Switzerland. September, 2007
"Pharmacokinetics and dynamics of ketamine in horses", University of Bern, June, 2007
"Uncertainty and Variability in Risk Assessment", NAS Workshop on Characterizing Uncertainty: Subgroup on Uncertainty in Estimating Low-Dose Risk from High-Dose Data, June, 2007
"The utility and interpretation of high-throughput screening data in risk assessment with focus on the NTP programs", International Workshop on "Evaluating upstream endpoints for improved decision making and risk assessment", Berkeley, Ca. May, 2007
"Future directions of risk assessment at the World Health Organization", International Program for Chemical Safety Harmonization Steering Committee Meeting, Berlin, Germany, May, 2007
"Chipping away at environmental health risk assessment", GEMS annual meeting, Research Triangle park, NC, April, 2007
"The forest for the trees: A systems approach to environmental health", National Conference on Science, Policy and the Environment, Washington, DC, February, 2007
"Emerging technologies and their application in risk assessment", National Research Council Standing Committee on Risk Analysis Issues and Reviews, Washington, DC, December, 206
"Stochastic systems biology modeling", International Congress on Systems Biology, Tokyo, Japan, October, 2006
"Acceptable risk: environmental health research and public health", National Symposium on Acceptable Risk in Clinical Medicine, RTP, NC, September, 2006
"Stochastic Systems Biology", Society for Industrial and Applied Mathematics Annual Meeting, Raleigh, NC, August, 2006.
"Identifying and Quantifying Gene Interaction Networks", Academia Sinica, Tapei, Taiwan, April, 2006.
"Estimating Health Risks from Environmental Exposures", Medical School, Cheng Kung National University, Taiwan, April, 2006.
"Systems Biology and Environmental Health Research", National Institute of Environmental Studies, Tsukuba, Japan, April, 2006.
"Cancer Research and Risk Assessment: Looking to the Future", National Cancer Institute, Japan, April, 2006
"Cancer Research and Risk Assessment: Looking to the Future", Keynote Address, Cancer and the Environment Symposium, Duke University, March, 2006
"Environmental Systems Biology", Mount St Mary's College, Maryland, March, 2006.
"Bioinformatics in High Throughput Screening: A proposal", ILSI Annual Meeting, Puerto Rico, January, 2006.
"Gene Regulatory Networks in Cancer Risk Assessment, German Cancer Research Center, Heidelberg, Germany, December, 2005.
"Alternative Methods in Toxicology; Problems and Solutions", Keynote Address, Workshop on Alternative Methods in Toxicology, Baltimore, Maryland, November, 2005
"Future Directions of the National Toxicology Program", American Public Health Association Annual Meeting, New Orleans, Louisiana, November, 2005.
"Risk Assessment", A Mini-Course, University of Finland, Kuopio, Finland, October, 2005.
"Mechanism-Based Modeling as an Alternative to Animals in Toxicology", 5th World Conference on Alternatives To Animals Used in the Life Sciences, Berlin, Germany, August, 2005.
"Environmental Systems Biology", Toxicology Forum, Aspen, Colorado, July, 2005.
"Mechanistic Implications of Modifiers of Chemical Toxicity", Keynote Lecture, Workshop on Modifiers of Chemical Toxicity: Implications for Human Health Risk Assessment, Poros, Greece, June, 2005.

"Future Directions in Screening for Toxicants", Committee on the Future of Toxicology, National Academy of Sciences, Washington, DC, May, 2005.
"Toxicogenomics and the Future of Cancer Risk Assessment", International Agency for Research on Cancer, Lyon France, May, 2005.
"Statistical Contributions to Federal Advisory Committees", Eastern North American Region of the Biometrics Society Annual Meeting, Austin, Texas, March, 2005.
"The Future of Long-Term Animal Carcinogenicity Studies", Invited Debate, Society of Toxicology Annual Meetings, New Orleans, Louisiana, March, 2005.
"Future Directions of the National Toxicology Program", Society of Toxicology Annual Meetings, New Orleans, Louisiana, March, 2005.
"Role of the National Toxicology Program in Risk Assessment", Federal-State Oncology and Risk Analysis Committee, Madison, Wisconsin, October, 2004.
"Identifying and Quantifying Cancer Risks using Mechanistic Data", Cancer and The Environment: NCI Science Writers Seminar, National Institutes of Health, Bethesda, Maryland, October, 2004.
"Toxicogenomics and the Future of Risk Assessment", Office of Science and Technology Policy, Washington, DC, August, 2004
"A Vision for the National Toxicology Program", Toxicology Forum, Aspen, Colorado, June, 2004
"Health and Environment", Joint Program on Climate Variability and Human Health, Atlanta, Georgia, March, 2004
"Dose-Response Analysis of Toxicogenomic Data", NIEHS Toxicogenomics Faculty, Research Triangle Park, NC, December, 2003.
"Dose-Response Analysis of Toxicogenomic Data", DERT Retreat, Southern Pines, NC, December, 2003.
"Toxicogenomics and the Research Directions of the National Toxicology Program", Keynote Address, Conference on Toxicology in the 21st Century, Korean Food and Drug Administration, Seoul, Korea, November, 2003.
"Dioxin and Agent Orange: What is known and what is suspected", Keynote Address, NCST/NIEHS Conference of Environmental Mediation for Dioxin Soil Contamination, Hanoi, Vietnam, November, 2003.
"Dose-Response Analysis of Toxicogenomic Data", NCSU/NIEHS Workshop on Bioinformatics and Risk Assessment, Research Triangle Park, NC, October, 2003.
"Health Effects of Electric and Magnetic Fields: Current Research Directions", Joint WHO/Japanese Ministry of Health Public Meeting on Cellular Radiation, Tokyo, Japan, September, 2003.
"Future Directions in Toxicology and the National Toxicology Program", NIEHS Public Liaison Meeting, New York City, September, 2003.
"Chipping Away at Risk Assessment: Genomics, Proteomics, Metabonomics and Cancer Risk Assessment", Society of Toxicologic Pathology, Savannah, Ga. June, 2003.
"Cancer Modeling; An Overview", Statistical Methods in Cancer Research, Radiation Effects Research Foundation, Kyoto, Japan, March, 2003 (presented by H. Toyoshiba due to war in Iraq and US Govt. responsibilities)
"Statistical methods for evaluating population exposures using CDC's Environmental Report Card", CDC, Atlanta, Ga. March, 2003.
"Bystander Effects in Carcinogenesis", Society of Toxicology Meetings, Salt Lake City, Utah, March, 2003
"Validation and use of genetically-modified mouse models as alternatives in carcinogenicity testing", ILSI Workshop on Genetically Modified Mouse Models, Alexandria, Va., February, 2003.
"The Future of Genomics in Toxicology", Mississippi State University, Oxford, Mississippi, February, 2003
"NIEHS priorities in ecological research", USGS Director's Retreat, Washington, DC. January, 2003.
"The US National Toxicology Program", Keynote Lecture at Opening Ceremonies for the Korean National Toxicology Program, Seoul, Korea, November, 2002

"The NIEHS/NTP Initiatives on Alternative Models in Toxicology", Science Advisory Board Meeting of the Center for the Advancement of Alternatives in Toxicology, Johns Hopkins University, Baltimore, Maryland, November, 2002

"The Analysis and Interpretation of Two-Stage Liver Bioassays in the Rat", Workshop on Hepatic Preneoplasia: Quantitative Evaluation in Carcinogenesis Bioassays and Relevance for Human Hepatocarcinogenesis, Heidelberg, Germany, June, 2002

"Chipping Away at Risk Assessment: Genomics, Proteomics, Metabonomics and Cancer Risk Assessment", Office of Environmental Health Hazard Assessment, California Department of Health, Sacramento, California, June, 2002

"Mechanistic Modeling of Stochastic Endpoints", NIEHS Conference on Future Directions in Biostatistics, Research Triangle Park, North Carolina, May, 2002

"Mechanistic Models of Skin Carcinogenesis", North Carolina State University, Raleigh, North Carolina, May, 2002

"Endocrine Dismodulation and Cancer", Workshop on Light, Endocrine Systems and Cancer, University of Cologne, Cologne, Germany, May, 2002

"Mechanism-Based Quantitative Analysis of Carcinogenesis Data", New York Academy of Sciences, April, 2002.

"Emerging Issues in Cancer Risk Assessment", Sunrise Expert Seminar, American Association for Cancer Research Annual Meetings, San Francisco, California, April, 2002

"Quantitative Evaluation of Health Risks from Dioxin", Joint US-Vietnam Scientific Conference on the Health and Environmental Effects of Agent Orange/Dioxin in Vietnam, Hanoi, Vietnam, March, 2002

"Cancer Risk Assessment", Hanoi Medical School/Vietnam Environmental Protection Agency/Vietnam Ministry of Health Joint Seminar, Hanoi Vietnam, January, 2002

"Mechanism-Based Modeling of Genomics Data", University of Berne, Switzerland, October, 2001.

"Multistage Models of Carcinogenesis", International Biometrics Conference Satellite Meeting, Fukuoka, Japan, September, 2001. (given by H. Toyoshiba due to conflict in scheduling)

"Discussion of CDC's Report on Levels of Environmental Contaminants in Human Tissues", National Public Radio Talk of the Nation Science Friday, March, 2001.

"Mixtures and Models in Environmental Health Risk Assessment", *NIEHS/Colorado State University* conference on the Application of Technology to Chemical Mixture Research, January, 2001

"Biological and biophysical research at extremely low- and radio-frequencies", Forschungsgemeinschaft Funk, Bad Münstereifel, Germany, December 2000.

"Harmonization of study evaluation and health risk assessment", 2nd International EMF Seminar, Chinese Ministry of Health, WHO and ICNIRP, Xian, China, October 2000.

"QRA: extrapolations (animals to humans; high dose low dose)", First International Course on Scientific Basis of Carcinogen Assessment – Quo Vadis?, NIVA, Naantali, Finland, September 2000.

"Statistical and biological models and toxicological issues in risk assessment", Center for Environmental Health Risk Assessment (CERA) workshop on Future Needs in Risk Assessment, Stockholm, Sweden, September 2000.

"Innovative use of mechanistic considerations in forming quantitative risk assessment models", Workshop on Future Research For Improving Risk Assessment Methods, National Institute for Occupational Safety and Health, Aspen, Colorado, August 2000.

"Latest scientific findings on dioxin", Dioxin Summit, University of California at Berkeley, California, August 2000.

"Stochastic modeling in carcinogenesis and development", Virtual Human Workshop, Research Triangle Park, NC. June 2000.

"Evaluating health risks; methods and examples", Federal Office for Public Health, Bern, Switzerland. May 2000.

"Decisions about environmental health risks: What are the key questions and how does this apply to melatonin", Low frequency EMF, visible light, melatonin and cancer", International Symposium, University of Cologne and the German Research Society, Cologne, Germany. May 2000.

"Children's environmental health-risk assessment issues and challenges", California Environmental Protection Agency, Office of Environmental Health Assessment, Oakland, CA. April 2000.

"Children's environmental health risks", WHO/NIEHS Pacific Rim Meeting on Children's Health, Manila, April, 2000.

"Toxicology and environmental health seminar series 2000", University of Florida, Gainesville, Florida. April 2000.

"Biologically based population health risk assessment models", University of Ottawa, Department of Epidemiology and Community Medicine and the Institute of Population Health, Ottawa, Canada. March 2000.

"Molecular epidemiology: a new tool in cancer prevention", Applying science to regulatory policy and public health, Keystone Symposium, Taos, New Mexico. February 2000.

"Statistical methods used for evaluating chemical safety in the environment", Mathematical Research Institute, Oberwolfach, Germany. February 2000.

"Mechanism-based mathematical modeling in health risk assessment" NC State University, January, 2000.

"Methods for analyzing and quantifying EMF health risks", WHO/ICNIRP scientific meeting, Erice, Sicily, Italy. November 1999.

"Harmonization of cancer and non-cancer risk assessment", The Society of Toxicology, Washington, DC. November 1999.

"Risky business: evaluating the safety of environmental exposures", Illinois Environmental Health Association, 1999 Annual Education Conference, Peoria, IL. October 1999.

"Challenges in the use of experimental and epidemiological data in health risk assessment", Symposium on "Statistical methods in epidemiology and demography", Department of Biostatistics at the University of North Carolina in Chapel Hill, Chapel Hill, North Carolina. October 1999.

"Linking toxicokinetics and toxicodynamics in biologically-based dose response models", Dioxin 99, 19th International Symposium on Halogenated Environmental Organic Pollutants and Persistent Organic Pollutants (POPs), Venice, Italy. September 1999.

"How did WHO decide on a TDI for dioxin", Faculty of Environmental Studies in Nagasaki University and the Department of Medical Informatics in Kyushu Medical School, Nagasaki, Japan. September 1999.

"PBPK Models for Estrogen", Workshop To Evaluate Research Priorities For Endocrine Active Compound Risk Assessment Methods, CMA, EPA, NIEHS, Research Triangle Park, North Carolina. August 1999.

"Risk assessment activities in the National Toxicology Program; risk assessment methodology, and mechanistic modeling of endocrine disruptors and dioxins", Harvard Symposium on Persistent Organic Pollutants (POPS): A Public Health Perspective, Boston, Massachusetts. June 1999.

"Do extremely low frequency electric and magnetic fields pose a health risk?", 21st Annual Meeting of the Bioelectromagnetics Society, Long Beach, California. June 1999.

"Probabilistic assessment of cancer dose response data", Probabilistic Risk Assessment Workshop, Sarasota, Florida. February 1999.

"Linking toxicology and epidemiology: the role of mechanistic modeling", CDC and ATSDR Symposium on Statistical Methods, Emerging Statistical Issues In Public Health For The 21st Century, Atlanta, Georgia. January 1999.

"Evaluation Of Health Risks From Exposure To Electric And Magnetic Fields; Completion Of A 2-Year Review Process", Toxicology Round Table, Virginia-Maryland Regional College of Veterinary Medicine, Blacksburg, Virginia. January 1999.

Interagency Agreements and Intramural Research Grants

US Environmental Protection Agency (1993-1994). Development and implementation of a mechanistic model for 2,3,7,8-TCDD. $25K.

Agency for Toxic Substances and Disease Registry (1994-1996). Study of the relationship between chemical structure/activity and dose-response shape and magnitude for carcinogens and development of physiologically based pharmacokinetic models for risk assessment. $796K.

NIEHS, Intramural Research Grant (1995-1998). Development of a mechanistic model of the impact of electromagnetic fields on melatonin. $180K.

NIEHS, Interagency Coordinator for Risk Assessment, EMFRAPID Program (1996-1998). Evaluation of potential risks from exposure to electric and magnetic fields. $2 million.

US Environmental Protection Agency (1999). Update to Dose-Response Chapter, Dioxin Reassessment. $25K.

NIH, Office of Research on Minority Health (1999-2002) GIS/Resampling method for evaluating data on environmental justice. $70K.

Direction of Ph.D. Theses:

A Bailer. *The effects of treatment lethality on tests of carcinogenicity*. Department of Biostatistics, University of North Carolina, Chapel Hill, North Carolina, 1986.

P Williams. *Estimating tumor incidence rates using the method of moments and maximum likelihood estimation combined*. Department of Biostatistics, University of North Carolina, Chapel Hill, North Carolina, 1989.

G Carr. *The analysis of data on adverse reactions to chemicals in developmental toxicology*. Department of Biostatistics, University of North Carolina, Chapel Hill, North Carolina, 1989.

S Liu. *Estimating parameters in a two-stage model of carcinogenesis using information on enzyme-altered foci from initiation-promotion experiments*. Department of Biostatistics, University of North Carolina, Chapel Hill, North Carolina, 1993.

CD Sherman. *Multipath/multistage models of carcinogenesis*. Department of Statistics and Actuarial Sciences, University of Waterloo, Waterloo, Ontario, Canada, 1994.

C Lyles. *Cell labeling data: Models and parameter estimation*. Department of Biostatistics, University of North Carolina, Chapel Hill, North Carolina, 1995.

F Ye. *The equal slopes test for benchmark doses*. Department of Biostatistics, University of North Carolina, Chapel Hill, North Carolina, 2001

S Whitaker. *Development of a biologically-based mathematical model of fetal development*. Department of Mathematics, North Carolina State University, Raleigh, North Carolina, 2000.

R Helms. *Homeostatic feedback control of growth on multistage cancer models*. Department of Biostatistics, University of North Carolina, Chapel Hill, North Carolina, 2001.

Journal Articles: (peer reviewed)

1. Cote I, Andersen ME, Ankley GT, Barone S, Birnbaum LS, Boekelheide K, et al. The Next Generation of Risk Assessment Multi-Year Study-Highlights of Findings, Applications to Risk Assessment, and Future Directions. *Environ Health Perspect* (2016) **124**(11):1671-82. doi: 10.1289/EHP233. PubMed PMID: 27091369; PubMed Central PMCID: PMCPMC5089888.

2. Parham F, Portier CJ, Chang X, Mevissen M. The Use of Signal-Transduction and Metabolic Pathways to Predict Human Disease Targets from Electric and Magnetic Fields Using in vitro Data in Human Cell Lines. *Frontiers in public health* (2016) **4**:193. doi: 10.3389/fpubh.2016.00193. PubMed PMID: 27656641; PubMed Central PMCID: PMCPMC5013261.

3. Portier CJ, Armstrong BK, Baguley BC, Baur X, Belyaev I, Belle R, et al. Differences in the carcinogenic evaluation of glyphosate between the International Agency for Research on Cancer (IARC) and the European Food Safety Authority (EFSA). *Journal of epidemiology and community health* (2016) **70**(8):741-5. doi: 10.1136/jech-2015-207005. PubMed PMID: 26941213; PubMed Central PMCID: PMCPMC4975799.

4. Sand S, Parham F, Portier CJ, Tice RR, Krewski D. Comparison of Points of Departure for Health Risk Assessment Based on High-Throughput Screening Data. *Environ Health Perspect* (2016). doi: 10.1289/EHP408. PubMed PMID: 27384688.

5. Scinicariello F, Portier C. A simple procedure for estimating pseudo risk ratios from exposure to non-carcinogenic chemical mixtures. *Archives of toxicology* (2016) **90**(3):513-23. doi: 10.1007/s00204-015-1467-z. PubMed PMID: 25667015.

6. Smith MT, Guyton KZ, Gibbons CF, Fritz JM, Portier CJ, Rusyn I, et al. Key Characteristics of Carcinogens as a Basis for Organizing Data on Mechanisms of Carcinogenesis. *Environ Health Perspect* (2016) **124**(6):713-21. doi: 10.1289/ehp.1509912. PubMed PMID: 26600562; PubMed Central PMCID: PMCPMC4892922.

7. McPartland, J., Dantzker, H.C., Portier, C. J. Building a robust 21st century chemical testing program at the U.S. Environmental Protection Agency: recommendations for strengthening scientific engagement, Environ Health Perspect 2015. 123 (1): p. 1-5.

8. Smith, M.T., Gibbons, C.F., Fritz, J.M., Rusyn, I., Lambert, P., Kavlock, R., Hecht, S.S., Bucher, J., Caldwell, J.C., Demarini, D., Cogliano, V., Portier, C., Paan, R., Straif, K., Guyton, K.Z., Key Characteristics of Carcinogens and an Approach to using Mechanistic Data in their Classification, Environ Health Perspect 2015 (in press)

9. Thomas, R., Thomas, R.S., Auerbach, S. S., Portier, C. J., Biological networks for predicting chemical hepatocarcinogenicity using gene expression data from treated mice and relevance across human and rat species. PLoS One, 2013. **8**(5): p. e63308.

10. Scinicariello, F., Buser, M.C., Mevissen, M., Portier, C.J., Blood lead level association with lower body weight in NHANES 1999-2006. Toxicol Appl Pharmacol, 2013. 273(3): p. 516-23.

11. Thomas R, Portier CJ., Gene Expression Networks, Methods Mol Biol. 2013;930:165-78.

12. Aylward LL, Kirman CR, Schoeny R, Portier CJ, Hays SM., Evaluation of Biomonitoring Data from the CDC National Exposure Report in a Risk Assessment Context: Perspectives across Chemicals. Environ Health Perspect. 2012 Dec 11. [Epub ahead of print] PMID: 23232556

13. Sand, S., Portier, C.J., Krewski, D. A Signal-to-noise crossover dose as the point of departure for risk assessment. Environmental Health Perspectives. 119(12):1766-74, 2011

14. Gohlke, J.M., Thomas, R., Woodward, A., Campbell-Lundrum, D., Pruss-Ustun, A., Hales, S., Portier, C.J. Estimating the global public health implications of electricity and coal consumption. Environmental Health Perspectives 2011 119 (6): 821-6

15. McHale CM, Zhang L, Lan Q, Vermeulen R, Li G, Hubbard AE, Porter KE, Thomas R, Portier CJ, Shen M, Rappaport SM, Yin S, Smith MT, Rothman N. Global gene expression profiling of a population exposed to a range of benzene levels. Environ Health Perspect. 2011 May;119(5):628-34.

16. Prause AS, Guionaud CT, Stoffel MH, Portier CJ, Mevissen M. Expression and function of 5-hydroxytryptamine 4 receptors in smooth muscle preparations from the duodenum, ileum, and pelvic flexure of horses without gastrointestinal tract disease. Am J Vet Res. 2010 Dec;71(12):1432-42.

17. Luke, N.S., DeVito, M.J., Portier, C.J., El-Masri, H.A., Employing a mechanistic model for the MAPK pathway to examine the impact of cellular all-or-none behavior on overall tissue response, Dose-Response 2010 8(3): 347-67.

18. Crump, KS, Chen, C., Chiu, W.A., Louis, T.A., Portier, C. J., Subramaniam, R.P., Wgite, P.D., What role for biologically-based Dose-Response Models in Estimating Low-Dose Risk. Env. Health Persp. 2010 118(5):585-8

19. Parham F, Austin C, Southall N, Huang R, Tice R, Portier C. Dose-Response modeling of High-Throughput Screening Data. J Biomol Screen. 2009 **14**(10), 1216-27

20. Hines RN, Sargent D, Autrup H, Birnbaum LS, Brent RL, Doerrer NG, Cohen Hubal EA, Juberg DR, Laurent C, Luebke R., Olejniczak K, Portier CJ, Slikker W. Approaches for assessing risks to sensitive populations: lessons learned from evaluating risks in the pediatric population. Tox. Sci. 2010 **113** (4), 4-26.

21. Portier, C. Toxicological decision making on hazards and risks – status quo and the way forward: current concepts and schemes of science-driven decision making – an overview. *Human and Experimental Toxicology* 2009 **28**(2-3), 123-125

22. Prause, A.S., Stoffel, M.H., Portier, C.J., Mevissen, M., Expression and function of 5-HT7 receptors in smooth muscle preparation from equine duodenum, ileum, and pelvic flexure, *Research in Veterinary Science* 2009 **87**(2), 292-299

23. Boyd, W.A., Smith, M. V., Kissling, G. E., Rice, J., R., Snyder, D. W., Portier, C. J., Freedman, J. H. Application of a Mathematical Model to Describe the Effects of Chlorpyrifos on Caenorhabditis elegans Development, *PLoS ONE* 2009 **4**(9): e7024. doi:10.1371/journal.pone.0007024

24. Smith MV, Boyd WA, Kissling GE, Rice JR, Snyder DW, et al. A Discrete Time Model for the Analysis of Medium-Throughput C. elegans Growth Data. *PLoS ONE* 2009 **4**(9): e7018. doi:10.1371/journal.pone.0007018

25. Gohlke, J. M., Stockton, P.S., Sieber, S., Foley, J., Portier, C. J. AhR-mediated gene expression in the developing mouse telencephalon. *Reproductive Toxicology* 2009 (in press) doi:10.1016/j.reprotox.2009.05.067

26. Thomas, R., Gohlke, J., Parham, F., Smith, M., Portier, C. (2009) Choosing the right path: enhancement of biologically-relevant sets of genes or proteins using pathway structure. *Genome Biology* 2009 **10**(4), R44.

27. Julia M Gohlke, Reuben Thomas, Yonqing Zhang, Michael C Rosenstein, Allan P Davis, Cynthia Murphy, Carolyn J Mattingly, Kevin G Becker, Christopher J Portier, Genetic and Environmental Pathways to Complex Disease. *BMC Systems Biology* 2009 May 5, 3:46.

28. Schmitz, A., Portier, C. J., Thurmann, W., Theurillat, R., Mevissen, M. Stereoselective biotransformation of ketamine in equine liver and lung microsomes. *J. Vet. Pharm. And Therapeutics* 2008 **31** (5): 446-455

29. Xia, M; Huang, R; Witt, KL; Southall, N; Fostel, J; Cho, MH; Jadhav, A; Smith, CS; Inglese, J; Portier, CJ; Tice, RR; Austin, CP Compound cytotoxicity profiling using quantitative high-throughput screening. *Env. Health Perspectives* 2008 **116** (3): 284-291

30. Gohlke, J. M., Armant, O., Parham, F., M., Smith, M., V., Zimmer, C., Castro, D., S., Nguyen, L., Parker, J., S., Gradwohl, G., Guillemot, F., Portier, C. J. Characterization of proneural gene regulatory network during mouse telencephalon development., *BMC Biology* 2008 **6** (15)

31. Subramaniam, R. P., Chen, C., Crump, K. S., Devoney, D., Fox, J., F., Portier, C. J., Schlosser, P. M., Thompson, C. M., White, P. Uncertainties in Biologically-based modeling of formaldehyde-induced respiratory cancer risk: Identification of key issues. *Risk Analysis* 2008 28(4): 907-923

32. Buehler, M., Steiner, A., Meylan, M., Portier, C. J., and Mevissen, M., In vitro effects of bethanechol on smooth muscle preparations obtained from abomasal fundus, corpus and antrum of dairy cows. Research in Vet. Sci. **2008 84** (3), 444-451

33. Barton, H.A., W.A. Chiu, R.W. Setzer, M.E. Andersen, A.J. Bailer, F.Y. Bois, R.S. Dewoskin, S. Hays, G. Johanson, N. Jones, G. Loizou, R.C. Macphail, C.J. Portier, M. Spendiff, and Y.M. Tan, Characterizing Uncertainty and Variability in Physiologically-based Pharmacokinetic (PBPK) Models: State of the Science and Needs for Research and Implementation. *Toxicol Sciences* 2007 **99** (2) 395-402.

34. Pfeiffer, J.B., M. Mevissen, A. Steiner, C.J. Portier, and M. Meylan, In vitro effects of bethanechol on specimens of intestinal smooth muscle obtained from the duodenum and jejunum of healthy dairy cows. Am J Vet Res, 2007. **68**(3): p. 313-22.

35. Smith, M., Miller, C., Kohn, M., Walker, N.J., Portier, C. J., Absolute estimation of initial concentrations of amplicon in a real-time PT-PCR process, *BMC Bioinformatics* 2007 **8**(1), 409

36. Toyoshiba, H., Sone, H., Yamanaka, T., Parham, F., Irwin, R., Boorman, G., and Portier, C. Gene network analysis suggests differences between high and low doses of acetaminophen. Toxicology and Applied Pharmacology *2006* 215 (3), 306-316

37. Knobloch M, Portier CJ, Levionnois OL, Theurillat R, Thormann W, Spadavecchia C, Mevissen M. Antinociceptive effects, metabolism and disposition of ketamine in ponies under target-controlled drug infusion. Toxicology And Applied Pharmacology 2005 216 (3): 373-386

38. Portier, C.J., H. Toyoshiba, H. Sone, F. Parham, R.D. Irwin, and G.A. Boorman, Comparative analysis of gene networks at multiple doses and time points in livers of rats exposed to acetaminophen. Altex 2005 **23 Suppl**: p. 380-4.

39. Resnik, D.B. and C. Portier, Pesticide testing on human subjects: weighing benefits and risks. Environ Health Perspect. 2005 **113**(7): p. 813-7.

40. Walker, N. J., Crockett, P. W., Nyska, A., Brix, A. E., Jokinen, M. P., Sells, D. M., Hailey, J. R., Easterling, M., Haseman, J. K., Yin, M., Wyde, M. E., Bucher, J. R., and Portier, C. J. Dose-additive carcinogenicity of a defined mixture of "dioxin-like compounds". *Environ Health Perspect* 2005 113, 43-48.[1]

41. Boorman, G. A., Irwin, R. D., Vallant, M. K., Gerken, D. K., Lobenhofer, E. K., Hejtmancik, M. R., Hurban, P., Brys, A. M., Travlos, G. S., Parker, J. S., and Portier, C. J. Variation in the hepatic gene expression in individual male Fischer rats. *Toxicol Pathol* 2005 33, 102-110.

42. Toyoshiba, H., Walker, N.J., Bailer, A.J. and Portier, C.J. Evaluation of toxic equivalency factors for induction of cytochromes P450 CYP1A1 and CYP1A2 enzyme activity by dioxin-like compounds. *Toxicol Appl Pharmacol 2004 194*, 156-68.

43. Trachsel, D., Tschudi, P., Portier, C.J., Kuhn, M., Thormann, W., Scholtysik, G. and Mevissen, M. Pharmacokinetics and pharmacodynamic effects of amiodarone in plasma of ponies after single intravenous administration. *Toxicology and Applied Pharmacology 2004, 195*, 113-125.

44. Bucher, J. R., and Portier, C. Human carcinogenic risk evaluation, Part V: The national toxicology program vision for assessing the human carcinogenic hazard of chemicals. *Toxicol Sci* 2004 82, 363-366.

45. Germolec, D. R., Kashon, M., Nyska, A., Kuper, C. F., Portier, C., Kommineni, C., Johnson, K. A., and Luster, M. I. The accuracy of extended histopathology to detect immunotoxic chemicals. *Toxicol Sci* 2004 82, 504-514.

46. Germolec, D. R., Nyska, A., Kashon, M., Kuper, C. F., Portier, C., Kommineni, C., Johnson, K. A., and Luster, M. I.. Extended histopathology in immunotoxicity testing: interlaboratory validation studies. *Toxicol Sci* 2004 78, 107-115.

47. Shelby, M., Portier, C., Goldman, L., Moore, J., Iannucci, A., Jahnke, G., and Donkin, S.. NTP-CERHR expert panel report on the reproductive and developmental toxicity of methanol. *Reprod Toxicol* 2004 18, 303-390.

48. Smith, M. V., Nyska, A., and Portier, C. Application of a statistical dynamic model investigating the short-term cellular kinetics induced by riddelliine, a hepatic endothelial carcinogen. *Toxicol Sci* 2004 80, 258-267.

49. Toyoshiba, H., Yamanaka, T., Sone, H., Parham, F. M., Walker, N. J., Martinez, J., and Portier, C. J. Gene interaction network suggests dioxin induces a significant linkage between aryl hydrocarbon receptor and retinoic acid receptor beta. *Environ Health Perspect* 2004 112, 1217-1224.[2]

---

[1] Awarded outstanding published paper in 2005 by the Risk Assessment Specialty Section of the Society of Toxicology

[2] Awarded outstanding published paper in 2004 by the Risk Assessment Specialty Section of the Society of Toxicology

50. Yamanaka, T., Toyoshiba, H., Sone, H., Parham, F. M., and Portier, C. J. The TAO-Gen algorithm for identifying gene interaction networks with application to SOS repair in E. coli. *Environ Health Perspect* 2004 112, 1614-1621

51. Schecter, A., Lucier, G. W., Cunningham, M. L., Abdo, K. M., Blumenthal, G., Silver, A. G., Melnick, R., Portier, C., Barr, D. B., Barr, J. R., Stanfill, S. B., Patterson, D. G., Jr., Needham, L. L., Stopford, W., Masten, S., Mignogna, J., and Tung, K. C. Human consumption of methyleugenol and its elimination from serum. *Environ Health Perspect* 2004 112, 678-680.

52. Bucher, J and Portier, C. Human carcinogenic risk evaluation, Part V: The national toxicology program vision for assessing the human carcinogenic hazard of chemicals. Toxicol Sci. 2004 Dec;82(2):363-6

53. Chhabra, R.S., Bucher, J.R., Wolfe, M. and Portier, C. Toxicity characterization of environmental chemicals by the US National Toxicology Program: an overview. *Int J Hyg Environ Health, 2003 206*, 437-45.

54. Germolec, D.R., Nyska, A., Kashon, M., Kuper, C.F., Portier, C., Kommineni, C., Johnson, K.A. and Luster, M.I. Extended Histopathology in Immunotoxicity Testing: Interlaboratory Validation Studies. *Toxicol Sci.* 2003 78(1) 107-115

55. Koken, P.J., Piver, W.T., Ye, F., Elixhauser, A., Olsen, L.M. and Portier, C.J. Temperature, air pollution, and hospitalization for cardiovascular diseases among elderly people in Denver. *Environ Health Perspect 2003 111*, 1312-7.

56. Whitaker, S.Y., Tran, H.T. and Portier, C.J. (2003) Development of a biologically-based controlled growth and differentiation model for developmental toxicology. *J Math Biol, 46*, 1-16.

57. Hauser, D.S., Mevissen, M., Weiss, R., Portier, C.J., Scholtysik, G., Studer, U.E. and Danuser, H. Effects of ketanserin and DOI on spontaneous and 5-HT-evoked peristalsis of the pig ureter in vivo. *Br J Pharmacol 2002 135*, 1026-32.

58. Kim, A.H., Kohn, M.C., Portier, C.J. and Walker, N.J. Impact of physiologically based pharmacokinetic modeling on benchmark dose calculations for TCDD-induced biochemical responses. *Regul Toxicol Pharmacol 2002 36*, 287-96.

59. Kohn, M.C., Melnick, R.L., Ye, F. and Portier, C.J. Pharmacokinetics of sodium nitrite-induced methemoglobinemia in the rat. *Drug Metab Dispos* 2002, *30*, 676-83.

60. Koo, J.W., Parham, F., Kohn, M.C., Masten, S.A., Brock, J.W., Needham, L.L. and Portier, C.J. The association between biomarker-based exposure estimates for phthalates and demographic factors in a human reference population. *Environ Health Perspect* 2002, *110*, 405-10.

61. Parham, F.M., Matthews, H.B. and Portier, C.J. A physiologically based pharmacokinetic model of p,p'-dichlorodiphenylsulfone. *Toxicol Appl Pharmacol* 2002, *181*, 153-63.

62. Portier, C.J. Endocrine dismodulation and cancer. *Neuroendocrinol Lett* 2002, *23 Suppl 2*, 43-7.

63. Simmons, P.T. and Portier, C.J. Toxicogenomics: the new frontier in risk analysis. *Carcinogenesis* 2002, *23*, 903-5.

64. Weiss, R., Mevissen, M., Hauser, D.S., Scholtysik, G., Portier, C.J., Walter, B., Studer, U.E. and Danuser, H. Inhibition of human and pig ureter motility in vitro and in vivo by the K(+) channel openers PKF 217-744b and nicorandil. *J Pharmacol Exp Ther* 2002, *302*, 651-8.

65. Whitaker, S.Y. and Portier, C. A controlled growth and differentiation model for non-monotonic dose-response. *Human and Ecological Risk Assessment* 2002, *8*, 1739-1755.

66. Kohn, M.C., Walker, N.J., Kim, A.H. and Portier, C.J. Physiological modeling of a proposed mechanism of enzyme induction by TCDD. *Toxicology* 2001, *162*, 193-208.

67. Mevissen, M., Denac, H., Schaad, A., Portier, C.J. and Scholtysik, G. Identification of a cardiac sodium channel insensitive to synthetic modulators. *J Cardiovasc Pharmacol Ther* 2001, *6*, 201-12.

68. Portier, C.J. Linking toxicology and epidemiology: the role of mechanistic modeling. *Stat Med* 2001, *20*, 1387-93.

69. Willems, B.A., Melnick, R.L., Kohn, M.C. and Portier, C.J. A physiologically based pharmacokinetic model for inhalation and intravenous administration of naphthalene in rats and mice. *Toxicol Appl Pharmacol* 2001, *176*, 81-91.

70. Ye, F., Piver, W.T., Ando, M. and Portier, C.J. Effects of temperature and air pollutants on cardiovascular and respiratory diseases for males and females older than 65 years of age in Tokyo, July and August 1980-1995. *Environ Health Perspect* 2001, *109*, 355-9.

71. Halmes, N.C., Roberts, S.M., Tolson, J.K. and Portier, C.J. Reevaluating cancer risk estimates for short-term exposure scenarios. *Toxicol Sci* 2000, *58*, 32-42.

72. Kohn, M.C., Parham, F., Masten, S.A., Portier, C.J., Shelby, M.D., Brock, J.W. and Needham, L.L. Human exposure estimates for phthalates. *Environ Health Perspect* 2000, *108*, A440-2.

73. Portier, C.J. and Parham, F.M. Comments on a biochemical model of cyclophosphamide hematotoxicity. *J Toxicol Environ Health A* 2000, *61*, 525-8.

74. Portier, C., Sherman, C.D. and Kopp-Schneider, A. Multistage, stochastic models of the cancer process: a general theory for calculating tumor incidence. *Stochastic Environmental Research and Risk Assessment* 2000, *14*, 173-179.

75. Sherman, C.D. and Portier, C.J. Calculation of the cumulative distribution function of the time to a small observable tumor. *Bull Math Biol* 2000, *62*, 229-40.

76. Smith, M.V. and Portier, C.J. Incorporating observability thresholds of tumors into the two-stage carcinogenesis model. *Math Biosci* 2000, *163*, 75-89.

77. Tritscher, A.M., Mahler, J., Portier, C.J., Lucier, G.W. and Walker, N.J. Induction of lung lesions in female rats following chronic exposure to 2,3,7,8-tetrachlorodibenzo-p-dioxin. *Toxicol Pathol* 2000, *28*, 761-9.

78. Walker, N.J., Tritscher, A.M., Sills, R.C., Lucier, G.W. and Portier, C.J. Hepatocarcinogenesis in female Sprague-Dawley rats following discontinuous treatment with 2,3,7,8-tetrachlorodibenzo-p-dioxin. *Toxicol Sci* 2000, *54*, 330-7.

79. Andersen, M.E., Conolly, R.B., Faustman, E.M., Kavlock, R.J., Portier, C.J., Sheehan, D.M., Wier, P.J. and Ziese, L. Quantitative mechanistically based dose-response modeling with endocrine-active compounds. *Environ Health Perspect* 1999, *107 Suppl 4*, 631-8.

80. el Masri, H. and Portier, C. Replication potential of cells via the protein kinase C-MAPK pathway: application of a mathematical model. *Bulletin of Mathematical Biology* 1999, *61*, 379-398.

81. El-Masri, H.A., Bell, D.A. and Portier, C.J. Effects of glutathione transferase theta polymorphism on the risk estimates of dichloromethane to humans. *Toxicol Appl Pharmacol* 1999, *158*, 221-30.

82. Melnick, R.L., Sills, R.C., Portier, C.J., Roycroft, J.H., Chou, B.J., Grumbein, S.L. and Miller, R.A. Multiple organ carcinogenicity of inhaled chloroprene (2-chloro-1,3-butadiene) in F344/N rats and B6C3F1 mice and comparison of dose-response with 1,3-butadiene in mice. *Carcinogenesis* 1999, *20*, 867-78.

83. Piver, W.T., Ando, M., Ye, F. and Portier, C.J. Temperature and air pollution as risk factors for heat stroke in Tokyo, July and August 1980-1995. *Environ Health Perspect* 1999, *107*, 911-6.

84. Walker, N.J., Portier, C.J., Lax, S.F., Crofts, F.G., Li, Y., Lucier, G.W. and Sutter, T.R. Characterization of the dose-response of CYP1B1, CYP1A1, and CYP1A2 in the liver of female Sprague-Dawley rats following chronic exposure to 2,3,7,8-tetrachlorodibenzo-p-dioxin. *Toxicol Appl Pharmacol* 1999, *154*, 279-86.

85. El-Masri, H.A. and Portier, C.J. Physiologically based pharmacokinetics model of primidone and its metabolites phenobarbital and phenylethylmalonamide in humans, rats, and mice. *Drug Metab Dispos* 1998, *26*, 585-94.

86. Jung, D., Berg, P.A., Edler, L., Ehrenthal, W., Fenner, D., Flesch-Janys, D., Huber, C., Klein, R., Koitka, C., Lucier, G., Manz, A., Muttray, A., Needham, L., Papke, O., Pietsch, M., Portier, C., Patterson, D., Prellwitz, W., Rose, D.M., Thews, A. and Konietzko, J. Immunologic findings in workers formerly exposed to 2,3,7,8-tetrachlorodibenzo-p-dioxin and its congeners. *Environ Health Perspect* 1998, *106 Suppl 2*, 689-95.

87. Kopp-Schneider, A., Portier, C. and Bannasch, P. A model for hepatocarcinogenesis treating phenotypical changes in focal hepatocellular lesions as epigenetic events. *Math Biosci* 1998, *148*, 181-204.

88. Murrell, J.A., Portier, C.J. and Morris, R.W. Characterizing dose-response: I: Critical assessment of the benchmark dose concept. *Risk Anal* 1998, *18*, 13-26.

89. Parham, F.M. and Portier, C.J. Using structural information to create physiologically based pharmacokinetic models for all polychlorinated biphenyls. II. Rates of metabolism. *Toxicol Appl Pharmacol* 1998, *151*, 110-6.

90. Portier, C.J. and Bell, D.A. Genetic susceptibility: significance in risk assessment. *Toxicol Lett* 1998, *102-103*, 185-9.

91. Portier, C.J. and Ye, F. U-shaped dose-response curves for carcinogens. *Hum Exp Toxicol* 1998, *17*, 705-7; discussion 716-8.

92. Sherman, C.D. and Portier, C.J. Eyes closed: simple, intuitive, statistically sound, and efficient methods for estimating parameters of clonal growth cancer models. *Risk Anal* 1998, *18*, 529-34.

93. Blumenthal, G.M., Kohn, M.C. and Portier, C.J. A mathematical model of production, distribution, and metabolism of melatonin in mammalian systems. *Toxicol Appl Pharmacol* 1997, *147*, 83-92.

94. Gaido, K.W., Leonard, L.S., Lovell, S., Gould, J.C., Babai, D., Portier, C.J. and McDonnell, D.P. Evaluation of chemicals with endocrine modulating activity in a yeast-based steroid hormone receptor gene transcription assay. *Toxicol Appl Pharmacol* 1997, *143*, 205-12.

95. Parham, F.M., Kohn, M.C., Matthews, H.B., DeRosa, C. and Portier, C.J. Using structural information to create physiologically based pharmacokinetic models for all polychlorinated biphenyls. *Toxicol Appl Pharmacol* 1997, *144*, 340-7.

96. Portier, C. and el Masri, H. Statistical research needs in mechanistic modeling for carcinogenic risk assessment. *Stat Methods Med Res* 1997, *6*, 305-15.

97. Sherman, C.D. and Portier, C.J. The two-stage model of carcinogenesis: overcoming the nonidentifiability dilemma. *Risk Anal* 1997, *17*, 367-74.

98. Buchanan, J.R. and Portier, C.J. The use of data on biologically reactive intermediates in risk assessment. *Adv Exp Med Biol* 1996, *387*, 429-37.

99. Bucher, J.R., Portier, C.J., Goodman, J.I., Faustman, E.M. and Lucier, G.W. Workshop overview. National Toxicology Program Studies: principles of dose selection and applications to mechanistic based risk assessment. *Fundam Appl Toxicol* 1996, *31*, 1-8.

100. Kohn, M.C., Sewall, C.H., Lucier, G.W. and Portier, C.J. A mechanistic model of effects of dioxin on thyroid hormones in the rat. *Toxicol Appl Pharmacol* 1996, *136*, 29-48.

101. Luster, M.I., Portier, C., Simeonova, P. and Munson, A.E. Relationship between chemical-induced immunotoxicity and carcinogenesis. *Drung Information Journal* 1996, *30*, 281-286.

102. Melnick, R.L., Kohn, M.C. and Portier, C.J. Implications for risk assessment of suggested nongenotoxic mechanisms of chemical carcinogenesis. *Environ Health Perspect* 1996, *104 Suppl 1*, 123-34.

103. Portier, C.J. and Lyles, C.M. Practicing safe modeling: GLP for biologically based mechanistic models. *Environ Health Perspect* 1996, *104*, 806.

104. Portier, C.J., Kopp-Schneider, A. and Sherman, C.D. Calculating tumor incidence rates in stochastic models of carcinogenesis. *Math Biosci* 1996, *135*, 129-46.

105. Portier, C.J., Sherman, C.D., Kohn, M., Edler, L., Kopp-Schneider, A., Maronpot, R.M. and Lucier, G. Modeling the number and size of hepatic focal lesions following exposure to 2,3,7,8-TCDD. *Toxicol Appl Pharmacol* 1996, *138*, 20-30.

106. Sherman, C.D. and Portier, C.J. Stochastic simulation of a multistage model of carcinogenesis. *Math Biosci* 1996, *134*, 35-50.

107. Kopp-Schneider, A. and Portier, C.J. Carcinoma formation in NMRI mouse skin painting studies is a process suggesting greater than two stages. *Carcinogenesis* 1995, *16*, 53-9.

108. Sherman, C.D. and Portier, C.J. Quantitative analysis of multiple phenotype enzyme-altered foci in rat hepatocarcinogenesis experiments: the multipath/multistage model. *Carcinogenesis* 1995, *16*, 2499-506.

109. Hoel, D.G. and Portier, C.J. Nonlinearity of dose-response functions for carcinogenicity. *Environ Health Perspect* 1994, *102 Suppl 1*, 109-13.

110. Kohn, M.C., Lucier, G.W. and Portier, C.J. The importance of biological realism in dioxin risk assessment models. *Risk Anal* 1994, *14*, 993-1000.

111. Kohn, M.C. and Portier, C.J. A model of effects of TCDD on expression of rat liver proteins. *Prog Clin Biol Res* 1994, *387*, 211-22.

112. Kopp-Schneider, A., Portier, C.J. and Sherman, C.D. The exact formula for tumor incidence in the two-stage model. *Risk Anal* 1994, *14*, 1079-80.

113. Kopp-Schneider, A. and Portier, C.J. A stem cell model for carcinogenesis. *Math Biosci* 1994, *120*, 211-32.

114. Luster, M.I., Portier, C., Pait, D.G. and Germolec, D.R. Use of animal studies in risk assessment for immunotoxicology. *Toxicology* 1994, *92*, 229-43.[3]

115. Portier, C.J., Lucier, G.W. and Edler, L. Risk from low-dose exposures. *Science* 1994, *266*, 1141-2.

116. Portier, C.J. Biostatistical issues in the design and analysis of animal carcinogenicity experiments. *Environ Health Perspect* 1994, *102 Suppl 1*, 5-8.

117. Sherman, C.D., Portier, C.J. and Kopp-Schneider, A. Multistage models of carcinogenesis: an approximation for the size and number distribution of late-stage clones. *Risk Anal* 1994, *14*, 1039-48.

118. Bailer, A.J. and Portier, C.J. An index of tumorigenic potency. *Biometrics* 1993, *49*, 357-65.

119. Carr, G.J. and Portier, C.J. An evaluation of some methods for fitting dose-response models to quantal-response developmental toxicology data. *Biometrics* 1993, *49*, 779-91.

120. Fenger, J., Forslund, J., Grandjean, P., Gron, P., Jensen, F., Keiding, L., Kjaergaard, S., Larsen, J., Lewtas, J., Lynge, E., Moller, H., Moller, L., Nielsen, P., Ostenfeldt, N., Pilsgaard, H., Portier, C., Poulsen, E., Rastogi, S., Skov, T., Thomsen, A., Ulbak, K. and Osterlind, A. Report: The Working Group On Cancer And The Non-Occupational Environment. *Pharmacology and Toxicology* 1993, *72*, 167-171.

121. Kohn, M.C. and Portier, C.J. Effects of the mechanism of receptor-mediated gene expression on the shape of the dose-response curve. *Risk Anal* 1993, *13*, 565-72.

122. Kohn, M.C., Lucier, G.W., Clark, G.C., Sewall, C., Tritscher, A.M. and Portier, C.J. A mechanistic model of effects of dioxin on gene expression in the rat liver. *Toxicol Appl Pharmacol* 1993, *120*, 138-54.

---

[3] Board of Publications, Best Paper Award, Society of Toxicology, 1995

123. Lucier, G.W., Portier, C.J. and Gallo, M.A. Receptor mechanisms and dose-response models for the effects of dioxins. *Environ Health Perspect* 1993, *101*, 36-44.

124. Luster, M.I., Portier, C., Pait, D.G., Rosenthal, G.J., Germolec, D.R., Corsini, E., Blaylock, B.L., Pollock, P., Kouchi, Y., Craig, W. and et al. Risk assessment in immunotoxicology. II. Relationships between immune and host resistance tests. *Fundam Appl Toxicol* 1993, *21*, 71-82.

125. Maronpot, R.R., Foley, J.F., Takahashi, K., Goldsworthy, T., Clark, G., Tritscher, A., Portier, C. and Lucier, G. Dose response for TCDD promotion of hepatocarcinogenesis in rats initiated with DEN: histologic, biochemical, and cell proliferation endpoints. *Environ Health Perspect* 1993, *101*, 634-42.

126. Meier, K.L., Bailer, A.J. and Portier, C.J. A measure of tumorigenic potency incorporating dose-response shape. *Biometrics* 1993, *49*, 917-26.

127. Melnick, R.L., Huff, J., Barrett, J.C., Maronpot, R.R., Lucier, G. and Portier, C.J. Cell proliferation and chemical carcinogenesis: symposium overview. *Environ Health Perspect* 1993, *101 Suppl 5*, 3-7.

128. Melnick, R.L., Huff, J., Barrett, J.C., Maronpot, R.R., Lucier, G. and Portier, C.J. Cell proliferation and chemical carcinogenesis: a symposium overview. *Mol Carcinog* 1993, *7*, 135-8.

129. Portier, C.J., Kopp-Schneider, A. and Sherman, C.D. Using cell replication data in mathematical modeling in carcinogenesis. *Environ Health Perspect* 1993, *101 Suppl 5*, 79-86.

130. Portier, C., Tritscher, A., Kohn, M., Sewall, C., Clark, G., Edler, L., Hoel, D. and Lucier, G. Ligand/receptor binding for 2,3,7,8-TCDD: implications for risk assessment. *Fundam Appl Toxicol* 1993, *20*, 48-56.[4]

131. Kopp-Schneider, A. and Portier, C.J. Birth and death/differentiation rates of papillomas in mouse skin. *Carcinogenesis* 1992, *13*, 973-8.

132. Luster, M.I., Pait, D.G., Portier, C., Rosenthal, G.J., Germolec, D.R., Comment, C.E., Munson, A.E., White, K. and Pollock, P. Qualitative and quantitative experimental models to aid in risk assessment for immunotoxicology. *Toxicol Lett* 1992, *64-65 Spec No*, 71-8.

133. Luster, M.I., Portier, C., Pait, D.G., White, K.L., Jr., Gennings, C., Munson, A.E. and Rosenthal, G.J. Risk assessment in immunotoxicology. I. Sensitivity and predictability of immune tests. *Fundam Appl Toxicol* 1992, *18*, 200-10.

134. Piegorsch, W.W., Carr, G.J., Portier, C.J. and Hoel, D.G. Concordance of carcinogenic response between rodent species: potency dependence and potential underestimation. *Risk Anal* 1992, *12*, 115-21.

135. Tritscher, A.M., Goldstein, J.A., Portier, C.J., McCoy, Z., Clark, G.C. and Lucier, G.W. Dose-response relationships for chronic exposure to 2,3,7,8-tetrachlorodibenzo-p-dioxin in a rat tumor promotion model: quantification and immunolocalization of CYP1A1 and CYP1A2 in the liver. *Cancer Res* 1992, *52*, 3436-42.

[4] Best-applied statistics paper, Centers for Disease Control, 1993.

136. Williams, P. and Portier, C. Analytic expressions for maximum likelihood estimators in a non-parametric model of tumor incidence and death. *Communications in Statistics - Theory and Methods* 1992, *21*, 711-732.

137. Williams, P. and Portier, C. Explicit solutions for constrained maximum likelihood estimators in survival/sacrifice experiments. *Biometrika* 1992, *74*, 717-729.

138. Carr, G.J. and Portier, C.J. An evaluation of the Rai and Van Ryzin dose-response model in teratology. *Risk Anal* 1991, *11*, 111-20.

139. Kopp-Schneider, A. and Portier, C.J. Distinguishing between models of carcinogenesis: the role of clonal expansion. *Fundam Appl Toxicol* 1991, *17*, 601-13.

140. Kopp-Schneider, A., Portier, C.J. and Rippmann, F. The application of a multistage model that incorporates DNA damage and repair to the analysis of initiation/promotion experiments. *Math Biosci* 1991, *105*, 139-66.

141. Portier, C.J. and Kopp-Schneider, A. A multistage model of carcinogenesis incorporating DNA damage and repair. *Risk Anal* 1991, *11*, 535-43.

142. Rogan, W.J., Blanton, P.J., Portier, C.J. and Stallard, E. Should the presence of carcinogens in breast milk discourage breast feeding? *Regul Toxicol Pharmacol* 1991, *13*, 228-40.

143. Bailer, A.J. and Portier, C.J. A note on fitting one-compartment models: non-linear least squares versus linear least squares using transformed data. *J Appl Toxicol* 1990, *10*, 303-6.

144. Portier, C.J. and Edler, L. Two-stage models of carcinogenesis, classification of agents, and design of experiments. *Fundam Appl Toxicol* 1990, *14*, 444-60.

145. Kopp, A. and Portier, C.J. A note on approximating the cumulative distribution function of the time to tumor onset in multistage models. *Biometrics* 1989, *45*, 1259-63.

146. Portier, C.J. and Kaplan, N.L. Variability of safe dose estimates when using complicated models of the carcinogenic process. A case study: methylene chloride. *Fundam Appl Toxicol* 1989, *13*, 533-44.

147. Portier, C.J. and Bailer, A.J. Testing for increased carcinogenicity using a survival-adjusted quantal response test. *Fundam Appl Toxicol* 1989, *12*, 731-7.

148. Portier, C.J. and Bailer, A.J. Two-stage models of tumor incidence for historical control animals in the National Toxicology Program's carcinogenicity experiments. *J Toxicol Environ Health* 1989, *27*, 21-45.

149. Bailer, A.J. and Portier, C.J. Effects of treatment-induced mortality and tumor-induced mortality on tests for carcinogenicity in small samples. *Biometrics* 1988, *44*, 417-31.

150. Bailer, A.J. and Portier, C.J. An illustration of dangers of ignoring survival differences in carcinogenic data. *J Appl Toxicol* 1988, *8*, 185-9.

151. Portier, C.J. Species correlation of chemical carcinogens. *Risk Anal* 1988, *8*, 551-3.

152. Portier, C. Life table analysis of carcinogenicity experiments. *Journal of the American College of Toxicology* 1988, *7*, 575-582.

153. Portier, C.J. Statistical properties of a two-stage model of carcinogenesis. *Environ Health Perspect* 1987, *76*, 125-31.

154. Portier, C.J. and Hoel, D.G. Issues concerning the estimation of the TD50. *Risk Anal* 1987, *7*, 437-47.

155. Portier, C.J. and Bailer, A.J. Simulating failure times when the event of interest is unobservable with emphasis on animal carcinogenicity studies. *Comput Biomed Res* 1987, *20*, 458-66.

156. Portier, C.J. and Dinse, G.E. Semiparametric analysis of tumor incidence rates in survival/sacrifice experiments. *Biometrics* 1987, *43*, 107-14.

157. Kupper, L.L., Portier, C., Hogan, M.D. and Yamamoto, E. The impact of litter effects on dose-response modeling in teratology. *Biometrics* 1986, *42*, 85-98.

158. Portier, C.J., Hedges, J.C. and Hoel, D.G. Age-specific models of mortality and tumor onset for historical control animals in the National Toxicology Program's carcinogenicity experiments. *Cancer Res* 1986, *46*, 4372-8.

159. Portier, C.J. Type 1 error and power of the linear trend test in proportions under the National Toxicology Program's modified pathology protocol. *Fundam Appl Toxicol* 1986, *6*, 515-9.

160. Portier, C. Estimating the tumor onset distribution in animal carcinogenesis experiments. *Biometrika* 1986, *3*, 371-378.

161. Portier, C.J. and Hoel, D.G. Design of animal carcinogenicity studies for goodness-of-fit of multistage models. *Fundam Appl Toxicol* 1984, *4*, 949-59.

162. Krewski, D., Crump, K.S., Farmer, J., Gaylor, D.W., Howe, R., Portier, C., Salsburg, D., Sielken, R.L. and Van Ryzin, J. A comparison of statistical methods for low dose extrapolation utilizing time-to-tumor data. *Fundam Appl Toxicol* 1983, *3*, 140-60.

163. Portier, C. and Hoel, D. Low-dose-rate extrapolation using the multistage model. *Biometrics* 1983, *39*, 897-906.

164. Portier, C. and Hoel, D. Optimal design of the chronic animal bioassay. *J Toxicol Environ Health* 1983, *12*, 1-19.

Journal Articles: (reviews and other)

165. McPartland, J., Dantzker, H.C., Portier, C.J., Building a robust 21st century chemical testing program at the U.S. Environmental Protection Agency: recommendations for strengthening scientific engagement, Environ Health Perspect, 2015. 123(1): 1-5.

166. Portier, C.J., L.R. Goldman, and B.D. Goldstein, Inconclusive findings: now you see them, now you don't! Environ Health Perspect, 2014. 122(2): p. A36.

167. Portier, C., ATSDR in the 21st Century, Journal of Environmental Health, 74(7):30-1, 2011

168. Portier, C. Comprehensive Public Health. Public Health Rep. 2011 126: Supp 1:3-6

169. Glass, R., Rosenthal, J., Jessup, C., Birnbaum, L., Portier, C. Tackling the research challenges of health and climate change. Env. Health Perspectives 2009 117 (12), A534-A535

170. Woodruff TJ, Zeise L, Axelrad DA, Guyton KZ, Janssen S, Miller M, et al. 2008. Meeting report: moving upstream-evaluating adverse upstream end points for improved risk assessment and decision-making. Environmental health perspectives 116(11): 1568-1575.

171. Portier, C. Risk Factors for Childhood Leukemia: Discussion and Summary, *Radiation Protection Dosimetry* 2009 **132**(2), 273-274.

172. Gohlke, J. M., Hrynkow, S. H., Portier, C. J., Health, Economy and Environment: Sustainable Energy Choices for a Nation, *Environmental Health Perspectives 2008* **116** (6), 236-237

173. Kissling, G. E., C. J. Portier and J. Huff. MtBE and cancer in animals: Statistical issues with poly-3 survival adjustments for lifetime studies. *Regul Toxicol Pharmacol.* 2008 **50** (3) 428-429

174. Barton, H. A., W. A. Chiu, R. Woodrow Setzer, M. E. Andersen, A. J. Bailer, F. Y. Bois, R. S. Dewoskin, S. Hays, G. Johanson, N. Jones, G. Loizou, R. C. Macphail, C. J. Portier, M. Spendiff and Y. M. Tan. Characterizing uncertainty and variability in physiologically based pharmacokinetic models: state of the science and needs for research and implementation. Toxicol Sci 2007 **99**(2): 395-402.

175. Gohlke, J. M. and C. J. Portier (2007). The forest for the trees: a systems approach to human health research. Environ Health Perspect 2007**115**(9): 1261-3.

176. Portier, C. J., Suk, W. A., Schwartz, D. A., Filling the translation-policy gap. *Env. Health Perspectives* 2007 **115** (3) A125

177. Becker, R. A., C. J. Borgert, S. Webb, J. Ansell, S. Amundson, C. J. Portier, A. Goldberg, L. H. Bruner, A. Rowan, R. D. Curren and W. T. Stott (2006). "Report of an ISRTP workshop: progress and barriers to incorporating alternative toxicological methods in the U.S." Regul Toxicol Pharmacol 2006 **46**(1): 18-22.

178. TCDD in female Sprague-Dawley rats. *Organohalogen Compounds* 1996, *29*, 222-227.

179. Portier, C.J. and D.A. Schwartz, The NIEHS and the National Toxicology Program: an integrated scientific vision. Environ Health Perspect, 2005. **113**(7): p. A440.

180. Portier, C. J., and Schwartz, D. A.. The NIEHS and the National Toxicology Program: an integrated scientific vision. *Environ Health Perspect* 2005, **113**, A440.

181. Portier, C.J., Suk, W. A. and D.A. Schwartz, Filling the translation-policy gap. Environ Health Perspect, 2005. **115**(3): p. A125.

182. Portier, C.J. Comments on the International Symposium on Light, Endocrine Systems and Cancer. *Neuroendocrinol Lett* 2002, *23 Suppl 2*, 79-81.

183. Portier, C. Chipping away at environmental health risk assessment. *Risk Policy Report* 2001, *8*, 37-38.

184. Portier, C. Decisions about environmental health risks: What are the key questions and how does this apply to melatonin? *Zentralblatt fur Arbeitsmedizin* 2000, *50*, 312-314.

185. Portier, C. Risk ranges for various endpoints following exposure to 2,3,7,8-TCDD. *Food Addit Contam* 2000, *17*, 335-46.

186. Brooks, E., Kohn, M., Van Birgelen, A., Lucier, G. and Portier, C. Stochastic models for papilloma formation following exposure to TCDD. *Organohalogen Compounds* 1999, *41*, 522-524.

187. Portier, C., Edler, L., Jung, D., Needham, L., Masten, S.A., Parham, F. and Lucier, G. Half-lives and body burdens for dioxin and dioxin-like compounds in humans estimated from an occupational cohort in Germany. *Organohalogen Compounds* 1999, *42*, 129-138.

188. Tritscher, A., Mahler, J., Portier, C., Lucier, G. and Walker, N.J. TCDD-induced lesions in rat lung following oral exposure. *Organohalogen Compounds* 1999, *42*, 285-288.

189. Lucier, G. and Portier, C. New Cancer Risk Guidelines. *Issues in Science and Technology* 1996, *13*, 10.

190. Portier, C. and Kohn, M. A biologically-based model for the carcinogenic effects of 2378-

191. Portier, C., Lucier, G. and Damstra, T. Dioxin research in Vietnam. *Science* 1995, *270*, 901.

192. Portier, C. and Kohn, M. Receptor Mediated Carcinogenesis and Dioxin. *Organohalogen Compounds* 1995, *26*, 267-272.

193. Edler, L. and Portier, C. Pharmacokinetic modeling of receptor/enzyme pathways for the action of dioxins. *Chemosphere* 1994, *25*, 239-242.

194. Edler, L., Portier, C. and Kopp-Schneider, A. Zur existenz von schwellenwerten: Wissenschaftliche methode oder statistisches artefakt in der risikoabschatzung. *Zbl Arbeitsmed* 1994, *44*, 16-21.

195. Kohn, M., Lucier, G., Sewall, C. and Portier, C. Physiological modeling of the effects of TCDD on thyroid hormones in the rat. *Organohalogen Compounds* 1994, *21*, 223-226.

196. Portier, C., Kohn, M., Sherman, C.D. and Lucier, G. Modeling the number and size of hepatic focal lesions following exposure to 2378-TCDD. *Organohalogen Compounds* 1994, *21*, 393-397.

197. Sherman, C.D. and Portier, C. The multipath/multistage model. *Informatik und Epidemiologie in Medizin und Biologie* 1994, *25*, 250-254.

198. Sherman, C.D. and Portier, C. The multipath/multistage model of carcinogenesis. *Organohalogen Compounds* 1994, *21*, 451-455.

199. Portier, C.J. Mechanistic modeling and risk assessment. *Pharmacol Toxicol* 1993, *72 Suppl 1*, 28-32.

200. Portier, C. Quantitative Risk Assessment. *ChemTech* 1990, *20*, 484-487.

Books/Book Chapters:

201. International Agency for Research on Cancer. Shiftwork, painting and firefighting (C. Portier member). *IARC Monographs on the Evaluation of Carcinogenic Risks to Humans Volume 98*. 2008 In press.

202. Trong, L., Portier, C.J. (eds) Proceedings of the Viet Nam – United States Scientific Conferences on Human Health and Environmental Effects of Agent Orange/Dioxin, Part 1: Environmental Effects - 3-6 March, 2002. Ha Noi, Vietnam, 2004.

203. Trong, L., Portier, C.J. (eds) Proceedings of the Viet Nam – United States Scientific Conferences on Human Health and Environmental Effects of Agent Orange/Dioxin, Part 2: Human Health Effects - 3-6 March, 2002. Ha Noi, Vietnam, 2004.

204. DeVito, M., Kim, A.H., Walker, N., Parham, F. and Portier, C. Dose-response modeling for 2,3,7,8-tetrachlorodibenzo-p-dioxin. *Dioxins and Health*; 2 ed.; John Wiley and Sons: Hoboken, NJ, 2003.

205. World Health Organization. Establishing a dialogue on Risks from Electromagnetic Fields. Sahl, J., Bonner, P., Kemp, R., Kheifets, L., Repacholi, M., van Deventer, E., Vogel, E. and Portier, C. WHO Press, 2002

206. International Agency for Research on Cancer. Non-ionizing Radiation, Part 1: Static and Extremely Low Frequency Electric and Magnetic Fields (C. Portier member). I *IARC Monographs on the Evaluation of Carcinogenic Risks to Humans Volume* 80. 19-26 2002

207. Wolfe, M., Boorman, G., Olden, K. and Portier, C. Power-Line Frequency Electric and Magnetic Fields: Effects on Human Health. *Human Health Risks from Exposure to ELF-EMF*; ICNIRP: Maastricht, The Netherlands, 2000.

208. Joint Committee on Food Additives (including C. Portier), World Health Organization/Food and Agriculture Organization. (1999) Evaluation of certain food additives and contaminants: Forty-ninth report of the Joint FAO/WHO Expert Committee on Food Additives. JCFA. WHO Technical Series #884. 1999.

209. Piver, W., Ando, M., Ye, F. and Portier, C. Temperature and air pollution as risk factors for cerebral vascular diseases in Tokyo, July-August 1980-1995. Proceedings of the 10-th Global Warming International Conference 1999

210. National Institute of Environmental Health Sciences. NIEHS Report on Health Effects from Exposure to Power-Line Frequency Electric and Magnetic Fields. Portier, C.J., Wolfe, M.S., Boorman, G.A., Bernheim, N.J., Galvin, M.J., Newton, S.A., Parham, F.M. and Olden, K.O. NIEHS 1999

211. Willems, B., Portier, C. and Lucier, G. Mechanism-based carcinogenic risk assessment of estrogens and estrogen-like compounds. *The Handbook of Environmental Chemistry*; Springer-Verlag: Heidelberg, 1999.

212. Portier, C.J. and Germolec, D. Immunotoxicology. *Encyclopedia of Biostatistics*; Wiley and Sons: Sussex, England, 1998 pp 1998-2000.

213. Portier, C.J. and Wolfe, M.S. Risk Communication: The focus in the NIEHS RAPID Program's review of EMF health Hazards. *Risk Perception, Risk Communication and Its Application to EMF Exposure*; ICNIRP: Oberschleisheim, Germany, 1998; pp 295-302.

214. Portier, C.J. Risk ranges for various endpoints following exposure to 2,3,7,8-TCDD. *WHO-ECEH/IPCS Assessment of health risk of dioxins*; International Program for Chemical Safety: Geneva, 1998; pp 23.

215. Portier, C.J. and Wolfe, M.S. (Eds.) EMF Science Review Symposium Breakout Group Report for Epidemiology Research Findings; National Institute of Environmental Health Sciences: Research Triangle Park, North Carolina, 1998.

216. Portier, C.J. and Wolfe, M.S. (Eds.) EMF Science Review Symposium Breakout Group Report for Clinical and *In Vivo* Laboratory Findings. In *NIH Publication Number 98-4400*; National Institute of Environmental Health Sciences: Research Triangle Park, North Carolina, 1998.

217. Portier, C.J. and Wolfe, M.S. (Eds.) Assessment of Health Effects from Exposure to Power-Line Frequency Electric and Magnetic Fields. In *NIH Publication Number 98-3981*; National Institute of Environmental Health Sciences: Research Triangle Park, North Carolina, 1998; pp 508.

218. Sherman, C.D. and Portier, C.J. Multistage carcinogenesis Models. *Encyclopedia of Biostatistics*; Wiley and Sons: Sussex, England, 1998; pp 2808-2814.

219. Portier, C.J. and Wolfe, M.S. Linking science to decisions: A strategy for electric and magnetic fields. *Biological Effects of ELF Electric and Magnetic Fields*; ICNIRP: Oberschleisheim, Germany, 1997; pp 211-218.

220. Portier, C.J. and Wolfe, M.S. (Eds.) EMF Science Review Symposium Breakout Group Report for Theoretical Mechanisms and *In Vitro* Research Findings; National Institute of Environmental Health Sciences: Research Triangle Park, North Carolina, 1997.

221. Buchanan, R. and Portier, C.J. The use of data on biologically reactive intermediates in risk assessment. *Biological reactive intermediates V : basic mechanistic research in toxicology and human risk assessment*; 387 ed.; Plenum Press: New York, 1995.

222. Luster, M., Portier, C., Pait, D.G., Rosenthal, G. and Germolec, D. Immunotoxicology and Risk Assessment. *Methods in immunotoxicology*; Wiley-Liss: New York, 1995; pp 51-68.

223. Portier, C.J. Quantitative models for cancer dose-response relationships: parameter estimation. *Low-Dose Extrapolation of Cancer Risks*; ILSI Press: Washington, 1995; pp 123-134.

224. Bailer, A.J. and Portier, C.J. Modeling risks from water contaminants: the application of concentration-response models. *Water contamination and health : integration of exposure assessment, toxicology, and risk assessment*; Dekker: New York, 1994; pp 447-466.

225. Kohn, M.C. and Portier, C.P. A model of the effects of TCDD on expression of rat liver proteins. *Receptor Mediated Biological Processes: Implications for Evaluating Carcinogens*; Wiley-Liss: New York, 1994; pp 211-222.

226. Kohn, M.C., Lucier, G.W. and Portier, C.J. Receptor mechanisms and dioxin risk assessment. *Hazardous Waste and Public Health: International Congress on the Health Effects of Hazardous Waste*; Princeton Press: Princeton, NJ, 1994; pp 421-432.

227. Portier, C.J. and Sherman, C.D. The potential effects of chemical mixtures on the carcinogenic process within the context of the mathematical multistage model. *Risk Assessment of Chemical Mixtures: Biological and Toxicological Issues*; Academic Press: New York, 1994; pp 665-686.

228. Portier, C.J., Hoel, D.G., Kaplan, N.L. and Kopp, A. Biologically based models for risk assessment. *IARC Sci Publ* 1990, 20-8.

229. Portier, C. Quantitative risk assessment. *Carcinogenicity and Pesticides*; American Chemical Society: New York, 1989; pp 164-174.

230. Portier, C. Design of long-term animal carcinogenicity experiments: dose allocation, animal allocation and sacrifice times. *Statistical Methods in Toxicological Research*; Gordan and Breech: New York, 1989; pp 455-467.

231. Portier, C. Utilizing biologically-based models to estimate carcinogenic risk. *Scientific Issues in Quantitative Cancer Risk Assessment*; Birkhauser: New York, 1989; pp 252-266.

232. Kaplan, N., Hogan, M., Portier, C. and Hoel, D. An evaluation of the safety factor approach in risk assessment. *Developmental Toxicology: Mechanisms and Risk*; Cold Spring Harbor Laboratory: New York, 1988.

233. Portier, C. (1985) Optimal dose/animal allocation for terminal sacrifice studies. Proceedings of the ASA Conference on Long-Term Animal Carcinogenicity Studies, 42-50.

234. Portier, C.J., Hoel, D. and VanRyzin, J. Statistical analysis of the carcinogenesis bioassay data relating to the risks from exposure to 2,3,7,8-tertachlorodibenzo-p-dioxin. *Public Health Risks of the Dioxins*; W. Kaufmann: Los Altos, 1984; pp 99-120.

Reports and Agency Publications:

235. A Human Health Perspective on Climate Change, Report of the Interagency Working Group on Climate Change and Health (C. Portier, chair), NIEHS, RTP, NC 2009

236. IARC Report of the Advisory Working Group for Priorities for future IARC Monographs, International Agency for Research on Cancer, Lyon, France 2008

237. NIEHSCenters for Children's Environmental Health and Disease Prevention Research Program, Review Panel Report, NIEHS, RTP, NC 2007

238. WHO Environmental Health Criteria Monograph 238; Extremely Low Frequency Electric and Magnetic Fields, (C. Portier, Chair), pp 543, World Health Organization Press, Geneva, Switzerland – 2007 ISBN 978 92 4 157238 5

239. IARC Report of the Advisory Group to Plan Monograph Volume 100: A Review of Human Carcinogens, International Agency for Research on Cancer, Lyon, France 2006

240. NIEHS Report of the Expert Panel to the NIEHS on Thimerosal Exposure in Pediatric Vaccines: Feasibility of Studies Using the Vaccine Safety Datalink, NIEHS, RTP, NC 2006

241. IARC Report of the Advisory Group to Revise the IARC Monographs Preamble International Agency for Research on Cancer, Lyon, France 2005

242. NTP. A National Toxicology Program for the 21st Century: A roadmap to achieve the NTP vision, pp. 4. National Toxicology Program/National Institute of Environmental Health Sciences, Research Triangle Park, NC. 2004

243. NTP. NTP Current directions and evolving strategies, pp. 4. National Toxicology Program/National Institute of Environmental Health Sciences, Research Triangle Park, NC. 2004

244. NTP. NTP Vision: Toxicology in the 21st Century: The role of the National Toxicology Program, pp. 4. National Toxicology Program/National Institute of Environmental Health Sciences, Research Triangle Park, NC 2003

245. USEPA Science Advisory Panel (C. Portier, Chair). Review of Characterization of Epidemiology Data Relating to Prostate Cancer and Exposure to Atrazine. FIFRA-SAP. US EPA Science Advisory Panel Report No. 2003-02.

246. USEPA Science Advisory Panel (C. Portier, Member). Review of Proposed Science Policy: PPAR-alpha Agonist-Mediated Hepatocarcinogenesis in Rodents and Relevance to Human Health Risk Assessments. FIFRA-SAP. US EPA Science Advisory Panel Report No. 2003-04.

247. USEPA Science Advisory Panel (C. Portier, Chair). Review of Physiologically-Based Pharmacokinetic/Pharmacodynamic Modeling: Preliminary Evaluation and Case Study for the N-Methyl Carbamate Pesticides: A Consultation. FIFRA-SAP. US EPA Science Advisory Panel Report No. 2003-05.

248. USEPA Science Advisory Panel (C. Portier, Member). Comments on Determination of the Appropriate FQPA Safety Factor(s) in the Organophosphorous Pesticide Cumulative Risk Assessment: Susceptibility and Sensitivity to the Common Mechanism, Acetylcholinesterase Inhibition. FIFRA-SAP. US EPA Science Advisory Panel Report No. 2002-03.

249. USEPA Science Advisory Panel (C. Portier, Member). Review Of Draft Termite Bait Product Performance Testing Guideline. FIFRA-SAP. US EPA Science Advisory Panel Report No. 2002-04.

250. USEPA Science Advisory Panel (C. Portier, Chair). Review of Corn Rootworm Plant-Incorporated Protectant Insect Resistance Management and Non-Target Insect Issues. FIFRA-SAP. US EPA Science Advisory Panel Report No. 2002-05.

251. USEPA Science Advisory Panel (C. Portier, Member). Review of Stochastic Human Exposure and Dose Simulation Model (SHEDS). FIFRA-SAP. US EPA Science Advisory Panel Report No. 2002-06.

252. IARC Monographs Volume 80 (C. Portier member) Non-Ionizing Radiation, Part 1: Static and Extremely Low-Frequency (ELF) Electric and Magnetic Fields, 429 pages; 2002

253. USEPA Science Advisory Panel (C. Portier, Member). Comments on Methods to Conduct a Preliminary Cumulative Risk Assessment for Organophosphorous Chemicals. FIFRA-SAP. US EPA Science Advisory Panel Report No. 2002-01.

254. USEPA Science Advisory Panel (C. Portier, Chair). BT Plant-Pesticides Risk and Benefit Assessment. FIFRA-SAP. US EPA Science Advisory Panel Report No. 2001-07.

255. USEPA Science Advisory Panel (C. Portier, Chair). Review of dietary exposure evaluation model (DEEM) and MaxLIP pesticide residue decompositing procedures and software. FIFRA-SAP. Science Advisory Panel Report No. 2001-01B.

256. USEPA Science Advisory Panel (C. Portier, Chair). Review of dietary exposure evaluation model (DEEM). FIFRA-SAP. EPA Science Advisory Panel Report No. 2001-01C.

257. USEPA Science Advisory Panel (C. Portier, Chair). Consultation on Development and Use of Distributions of Pesticide Concentrations in Drinking Water for FQPA Assessments. FIFRA-SAP. EPA Science Advisory Panel Report No. 2001-01D.

258. USEPA Science Advisory Panel (C. Portier, Member). Review of Probabilistic models and methodologies: Advancing the ecological risk assessment process in the EPA Office of Pesticides Programs. FIFRA-SAP. US EPA Science Advisory Panel Report No. 2001-06.

259. USEPA Science Advisory Panel (C. Portier, Chair). Review of Common Mechanism of Action of Thiocarbamates and Dithiocarbamates. FIFRA-SAP. US EPA Science Advisory Panel Report No. 2001-12.

260. USEPA Science Advisory Panel (C. Portier, Member). Review of Preliminary Cumulative Hazard and Dose-Response Assessment for Organophosphorus Pesticides: Determination of Relative Potency and Points of Departure for Cholinesterase Inhibition. FIFRA-SAP. US EPA Science Advisory Panel Report No. 2001-11.

261. USEPA Science Advisory Panel (C. Portier, Chair). Review of Characterization and Non-Target Organism Data Requirements for Protein Plant-Pesticides and- Cumulative Risk Assessment Methodology Issues of Pesticide Substances that Have a Common Mechanism of Toxicity. FIFRA-SAP. EPA-SAP-99-06. February 4, 2000.

262. USEPA Science Advisory Panel (C Portier, Member). Review of food allergenicity of CRY9C Endotoxin and other non-digestible proteins. FIFRA-SAP. USEPA Science Advisory Panel Report 2000-01A.

263. USEPA Science Advisory Panel (C Portier, Member). Review of atrazine: hazard and dose-response assessment and characterization. FIFRA-SAP. USEPA Science Advisory Panel Report No. 2000-05.

264. USEPA Science Advisory Panel (C. Portier, Chairman). Review of Statistical Methods for Use of Composite Data in Acute Dietary Exposure Assessment. FIFRA-SAP. EPA-SAP 99-03B. May 26, 1999.

265. USEPA Science Advisory Panel (C. Portier, Chairman). Review of the Use of Watershed-derived Percent Crop Areas as a Refinement Tool in FQPA Drinking Water Exposure Assessments for Tolerance Reassurance. FIFRA-SAP. EPA-SAP 99-03C. May 26, 1999.

266. USEPA Science Advisory Panel (C. Portier, Member). *Review of Burkholderia cepacia*: Risk Assessment of a Biopesticide with Affinities to a Human Opportunistic Pathogen. FIFRA-SAP. EPA-SAP-99-04. September 30, 1999.

267. USEPA Science Advisory Panel (C. Portier, Member). A Consultation on Protocol Design to Assess Acute Neurotoxicity Studies following Oral Administration of Pesticides. FIFRA-SAP. EPA-SAP-99-04B. September 30, 1999.

268. USEPA Science Advisory Panel (C. Portier, Member). Review of Higher Tier Ecological Risk Assessment for Chlorfenapyr. FIFRA-SAP. EPA-SAP-99-04C. September 30, 1999.

269. USEPA Science Advisory Panel (C. Portier, Chair). Review of -Spray Drift: Review of Proposed Pesticide Deposition Curves to Adjacent Areas. FIFRA-SAP. EPA-SAP-99-04C. September 30, 1999.

270. USEPA Science Advisory Panel (C. Portier, Member). Review of Guidance Document for Small Scale Prospective Ground Water Monitoring Studies. FIFRA-SAP. SAP98-01. November 19, 1998.

271. USEPA Science Advisory Panel (C. Portier, Member). Review of Common Mechanism of Action of Organophosphates. FIFRA-SAP. March, 1998.

272. USEPA Science Advisory Panel (C. Portier, Member). Review of Suggested Probabilistic Risk Assessment Methodology for Evaluating Pesticides That Exhibit a Common Mechanism of Action. FIFRA-SAP. March, 1998.

273. USEPA Science Advisory Panel (C. Portier, Member). Review of the Use of FQPA 10x Safety Factor to Address Special Sensitivity of Infants and Children to Pesticides. FIFRA-SAP. March, 1998.

274. USEPA Science Advisory Panel (C. Portier, Member). Review of Post Application Exposure Guidelines: Series 875-Group B. FIFRA-SAP. March, 1998.

275. USEPA Science Advisory Panel (C. Portier, Member). Review of Proposed Methods for Basin-scale Estimation of Pesticide Concentrations in Flowing Water and Reservoirs for Tolerance Reassessment. FIFRA-SAP. July, 1998.

276. USEPA Science Advisory Panel (C. Portier, Member). Review of Linear Low Dose Extrapolation for Cancer Risk Decisions: Sources of Uncertainty and How They Affect the Precision of Risk Estimates. FIFRA-SAP. July, 1998.

277. USEPA Science Advisory Panel (C. Portier, Member). Review of DDVP (Dichlorvos) Risk Issues. FIFRA-SAP. July, 1998.

278. USEPA Science Advisory Panel (C. Portier, Member). Review of FQPA 10x Safety Factor: Status Report. FIFRA-SAP. July, 1998.

279. USEPA Science Advisory Panel (C. Portier, Member). Review of Chlorothalonil: Mechanism for the Formation of Renal and Forestomach Tumors. FIFRA-SAP. July, 1998.

280. Environmental Protection Agency. Dose Response Modeling for TCDD. Lucier, G., Gallo, M., Portier, C.J., Bayard, S., Cooper, K., Georgopolous, P., McGrath, L., Andersen, M., DeVito, M., White, P., Kedderis, L., Mills, J. and Silbergeld, E. EPA/600/BP-92/001b 1994

281. Portier, C.J. Optimal bioassay design under the Armitage-Doll multistage model of carcinogenesis. PhD, University of North Carolina. 1981.

APPENDICES

Document One

Differences in the carcinogenic evaluation of glyphosate between the International Agency for Research on Cancer (IARC) and the European Food Safety Authority (EFSA)

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The International Agency for Research on Cancer (IARC) Monographs Programme identifies chemicals, drugs, mixtures, occupational exposures, lifestyles and personal habits, and physical and biological agents that cause cancer in humans and has evaluated about 1000 agents since 1971. Monographs are written by ad hoc Working Groups (WGs) of international scientific experts over a period of about 12 months ending in an eight-day meeting. The WG evaluates all of the publicly available scientific information on each substance and, through a transparent and rigorous process,[1] decides on the degree to which the scientific evidence supports that substance's potential to cause or not cause cancer in humans.

For Monograph 112,[2] 17 expert scientists evaluated the carcinogenic hazard for four insecticides and the herbicide glyphosate.[3] The WG concluded that the data for glyphosate meet the criteria for classification as a *probable human carcinogen*.

The European Food Safety Authority (EFSA) is the primary agency of the European Union for risk assessments regarding food safety. In October 2015, EFSA reported[4] on their evaluation of the Renewal Assessment Report[5] (RAR) for glyphosate that was prepared by the Rapporteur Member State, the German Federal Institute for Risk Assessment (BfR). EFSA concluded that 'glyphosate is unlikely to pose a carcinogenic hazard to humans and the evidence does not support classification with regard to its carcinogenic potential'. Addendum 1 (the BfR Addendum) of the RAR[5] discusses the scientific rationale for differing from the IARC WG conclusion.

Serious flaws in the scientific evaluation in the RAR incorrectly characterise the potential for a carcinogenic hazard from exposure to glyphosate. Since the RAR is the basis for the European Food Safety Agency (EFSA) conclusion,[4] it is critical that these shortcomings are corrected.

THE HUMAN EVIDENCE

EFSA concluded 'that there is very limited evidence for an association between glyphosate-based formulations and non-Hodgkin lymphoma (NHL), overall inconclusive for a causal or clear associative relationship between glyphosate and cancer in human studies'. The BfR Addendum (p. ii) to the EFSA report explains that 'no consistent positive association was observed' and 'the most powerful study showed no effect'. The IARC WG concluded there is *limited evidence of carcinogenicity in humans* which means "A positive association has been observed between exposure to the agent and cancer for which a causal interpretation is considered by the Working Group to be credible, but chance, bias or confounding could not be ruled out with reasonable confidence."[1]

The finding of *limited evidence* by the IARC WG was for NHL, based on high-quality case–control studies, which are particularly valuable for determining the carcinogenicity of an agent because their design facilitates exposure assessment and reduces the potential for certain biases. The Agricultural Health Study[6] (AHS) was the only cohort study available providing information on the carcinogenicity

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of glyphosate. The study had a null finding for NHL (RR 1.1, 0.7–1.9) with no apparent exposure–response relationship in the results. Despite potential advantages of cohort versus case–control studies, the AHS had only 92 NHL cases in the unadjusted analysis as compared to 650 cases in a pooled case–control analysis from the USA.[7] In addition, the median follow-up time in the AHS was 6.7 years, which is unlikely to be long enough to account for cancer latency.[8]

The RAR classified all of the case–control studies as 'not reliable,' because, for example, information on glyphosate exposure, smoking status and/or previous diseases had not been assessed. In most cases, this is contrary to what is actually described in the publications. Well-designed case–control studies are recognised as strong evidence and routinely relied on for hazard evaluations.[9 10] The IARC WG carefully and thoroughly evaluated all available epidemiology data, considering the strengths and weaknesses of each study. This is key to determining that the positive associations seen in the case–control studies are a reliable indication of an association and not simply due to chance or methodological flaws. To provide a reasonable interpretation of the findings, an evaluation needs to properly weight studies according to quality rather than simply count the number of positives and negatives. The two meta-analyses cited in the IARC Monograph[11] are excellent examples of objective evaluations and show a consistent positive association between glyphosate and NHL.

The final conclusion[5] (Addendum 1, p.21) that "there was no unequivocal evidence for a clear and strong association of NHL with glyphosate" is misleading. IARC, like many other groups, uses three levels of evidence for human cancer data.[1] *Sufficient evidence* means 'that a causal relationship has been established' between glyphosate and NHL. BfR's conclusion is equivalent to deciding that there is not *sufficient evidence*. Legitimate public health concerns arise when 'causality is credible', that is, when there is *limited evidence of carcinogenicity.*

EVIDENCE FROM ANIMAL CARCINOGENICITY STUDIES

EFSA concluded 'No evidence of carcinogenicity was confirmed by the majority of the experts (with the exception of one minority view) in either rats or mice due to a lack of statistical significance in pairwise comparison tests, lack of consistency in multiple animal studies and slightly increased incidences only at dose levels at or above the limit dose/maximum tolerated dose (MTD), lack of preneoplastic lesions and/or being within historical control range'. The IARC WG review found a significant positive trend for renal tumours in male CD-1 mice,[12] a rare tumour, although no comparisons of any individual exposure group to the control group were statistically significant. The WG also identified a significant positive trend for hemangiosarcoma in male CD-1 mice,[13] again with no individual exposure group significantly different from controls. Finally, the WG also saw a significant increase in the incidence of pancreatic islet cell adenomas in two studies in male Sprague-Dawley rats.[14–16] In one of these rat studies, thyroid gland adenomas in females and liver adenomas in males were also increased. By the IARC review criteria,[1] this constitutes *sufficient evidence* in animals.

The IARC WG reached this conclusion using data that were publicly available in sufficient detail for independent scientific evaluation (a requirement of the IARC Preamble[1]). On the basis of the BfR Addendum, it seems there were three additional mouse studies and two additional rat studies that were unpublished and available to EFSA. Two of the additional studies were reported to have a significant trend for renal tumours, one in CD-1 mice (Sugimoto. *18-Month Oral Oncogenicity Study in Mice*. Unpublished, designated ASB2012–11493 in RAR. 1997), and one in Swiss-Webster mice (Unknown. *A chronic feeding study of glyphosate (roundup technical) in mice*. Unpublished, designated ABS2012–11491 in RAR. 2001). One of these studies (Sugimoto. Unpublished, 1997) also reported a significant trend for hemangiosarcoma. The RAR also reported two studies in CD-1 mice showing significant trends for malignant lymphoma (Sugimoto. Unpublished, 1997; Unknown. *Glyphosate Technical: Dietary Carcinogencity Study in the Mouse*. Unpublished, designated ABS2012–11492 in RAR. 2009).

The RAR dismissed the observed trends in tumour incidence because there are no individual treatment groups that are significantly different from controls and because the maximum observed response is reportedly within the range of the historical control data (Table 5.3–1, p.90). Care must be taken in using historical control data to evaluate animal carcinogenicity data. In virtually all guidelines,[1 17 18] scientific reports[19] and publications[20–23] on this issue, the recommended first choice is the use of concurrent controls and trend tests, even in the EC regulations cited in the RAR[18] (see p.375). Trend tests are more powerful than pairwise comparisons, particularly for rare tumours where data are sparse. Historical control data should be from studies in the same time frame, for the same animal strain, preferably from the same laboratory or the same supplier and preferably reviewed by the same pathologist.[17 18] While the EFSA final peer review[4] mentions the use of historical control data from the original laboratory, no specifics are provided and the only referenced historical control data[24] are in the BfR addendum.[5] One of the mouse studies[12] was clearly done before this historical control database was developed, one study (Sugimoto. Unpublished, 1997) used Crj:CD-1 mice rather than Crl:CD-1 mice, and one study[13] did not specify the substrain and was reported in 1993 (probably started prior to 1988). Hence, only a single study (Unknown. Unpublished, 2009) used the same mouse strain as the cited historical controls, but was reported more than 10 years after the historical control data set was developed.

The RAR dismissed the slightly increased tumour incidences in the studies considered because they occurred "only at dose levels at or above the limit dose/maximum tolerated dose (MTD)", and because there was a lack of preneoplastic lesions. Exceeding the MTD is demonstrated by an increase in mortality or other serious toxicological findings at the highest dose, not by a slight reduction in body weight. No serious toxicological findings were reported at the highest doses for the mouse studies in the RAR. While some would argue that these high doses could cause cellular disruption (eg, regenerative hyperplasia) leading to cancer, no evidence of this was reported in any study. Finally, a lack of preneoplastic lesions for a significant neoplastic finding is insufficient reason to discard the finding.

MECHANISTIC INFORMATION

The BfR Addendum dismisses the IARC WG finding that 'there is strong evidence that glyphosate causes genotoxicity' by suggesting that unpublished evidence not seen by the IARC WG was overwhelmingly negative and that, since the reviewed studies were not done under guideline principles, they should get less weight. To maintain transparency, IARC reviews only publicly available data. The use of confidential data submitted to the BfR makes it impossible for any scientist not associated with BfR to review this conclusion. Further weakening their interpretation,

the BfR did not include evidence of chromosomal damage from exposed humans or human cells that were highlighted in Tables 4.1 and 4.2 of the IARC Monograph [3]

The BfR confirms (p.79) that the studies evaluated by the IARC WG on oxidative stress were predominantly positive but does not agree that this is strong support for an oxidative stress mechanism. They minimise the significance of these findings predominantly because of a lack of positive controls in some studies and because many of the studies used glyphosate formulations and not pure glyphosate. In contrast, the WG concluded that (p.77) 'Strong evidence exists that glyphosate, AMPA and glyphosate-based formulations can induce oxidative stress'. From a scientific perspective, these types of mechanistic studies play a key role in distinguishing between the effects of mixtures, pure substances and metabolites.

Finally, we strongly disagree that data from studies published in the peer-reviewed literature should automatically receive less weight than guideline studies. Compliance with guidelines and Good Laboratory Practice does not guarantee validity and relevance of the study design, statistical rigour and attention to sources of bias.[25 26] The majority of research after the initial marketing approval, including epidemiology studies, will be conducted in research laboratories using various models to address specific issues related to toxicity, often with no testing guidelines available. Peer-reviewed and published findings have great value in understanding mechanisms of carcinogenicity and should be given appropriate weight in an evaluation based on study quality, not just on compliance with guideline rules.

GENERAL COMMENTS

Science moves forward on careful evaluations of data and a rigorous review of findings, interpretations and conclusions. An important aspect of this process is transparency and the ability to question or debate the findings of others. This ensures the validity of the results and provides a strong basis for decisions. Many of the elements of transparency do not exist for the RAR.[5] For example, citations for almost all references, even those from the open scientific literature, have been redacted. The ability to objectively evaluate the findings of a scientific report requires a complete list of cited supporting evidence. As another example, there are no authors or contributors listed for either document, a requirement for publication in virtually all scientific journals where financial support, conflicts of interest and affiliations of authors are fully disclosed. This is in direct contrast to the IARC WG evaluation listing all authors, all publications and public disclosure of pertinent conflicts of interest prior to the WG meeting.[27]

Several guidelines have been devised for conducting careful evaluation and analysis of carcinogenicity data, most after consultation with scientists from around the world. Two of the most widely used guidelines in Europe are the OECD guidance on the conduct and design of chronic toxicity and carcinogenicity studies[17] and the European Chemicals Agency Guidance on Commission Regulation (EU) No 286/2011;[18] both are cited in the RAR. The methods used for historical controls and trend analysis are inconsistent with these guidelines.

Owing to the potential public health impact of glyphosate, which is an extensively used pesticide, it is essential that all scientific evidence relating to its possible carcinogenicity is publicly accessible and reviewed transparently in accordance with established scientific criteria.

SUMMARY

The IARC WG concluded that glyphosate is a 'probable human carcinogen', putting it into IARC category 2A due to *sufficient evidence* of carcinogenicity in animals, *limited evidence* of carcinogenicity in humans and *strong* evidence for two carcinogenic mechanisms.

- The IARC WG found an association between NHL and glyphosate based on the available human evidence.
- The IARC WG found significant carcinogenic effects in laboratory animals for rare kidney tumours and hemangiosarcoma in two mouse studies and benign tumours in two rat studies.
- The IARC WG concluded that there was strong evidence of genotoxicity and oxidative stress for glyphosate, entirely from publicly available research, including findings of DNA damage in the peripheral blood of exposed humans.

The RAR concluded[5] (Vol. 1, p.160) that 'classification and labelling for carcinogenesis is not warranted' and 'glyphosate is devoid of genotoxic potential'.

- EFSA[4] classified the human evidence as 'very limited' and then dismissed any association of glyphosate with cancer without clear explanation or justification.
- Ignoring established guidelines cited in their report, EFSA dismissed evidence of renal tumours in three mouse studies, hemangiosarcoma in two mouse studies and malignant lymphoma in two mouse studies. Thus, EFSA incorrectly discarded all findings of glyphosate-induced cancer in animals as chance occurrences.
- EFSA ignored important laboratory and human mechanistic evidence of genotoxicity.
- EFSA confirmed that glyphosate induces oxidative stress but then, having dismissed all other findings of possible carcinogenicity, dismissed this finding on the grounds that oxidative stress alone is not sufficient for carcinogen labelling.

The most appropriate and scientifically based evaluation of the cancers reported in humans and laboratory animals as well as supportive mechanistic data is that glyphosate is a *probable human carcinogen*. On the basis of this conclusion and in the absence of evidence to the contrary, it is reasonable to conclude that glyphosate formulations should also be considered likely human carcinogens. The CLP Criteria[18] (Table 3.6.1, p.371) allow for a similar classification of Category 1B when there are 'studies showing limited evidence of carcinogenicity in humans together with limited evidence of carcinogenicity in experimental animals'.

In the RAR, almost no weight is given to studies from the published literature and there is an over-reliance on non-publicly available industry-provided studies using a limited set of assays that define the minimum data necessary for the marketing of a pesticide. The IARC WG evaluation of *probably carcinogenic to humans* accurately reflects the results of published scientific literature on glyphosate and, on the face of it, unpublished studies to which EFSA refers.

Most of the authors of this commentary previously expressed their concerns to EFSA and others regarding their review of glyphosate[28] to which EFSA has published a reply.[29] This commentary responds to the EFSA reply.

The views expressed in this editorial are the opinion of the authors and do not imply an endorsement or support for these opinions by any organisations to which they are affiliated.

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Competing interests CJP, MTS and DDW are providing advice to a US law firm involved in glyphosate litigation. CJP also works part-time for the Environmental Defense Fund on issues not related to pesticides.

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REFERENCES

1 IARC. *Preamble to the IARC Monographs*. 2006. http://monographs.iarc.fr/ENG/Preamble/CurrentPreamble.pdf
2 Guyton KZ, Loomis D, Grosse Y, *et al*. Carcinogenicity of tetrachlorvinphos, parathion, malathion, diazinon, and glyphosate. *Lancet Oncol* 2015;16:490–1.
3 IARC Working Group. Glyphosate. In: *Some organophosphate insecticides and herbicides: diazinon, glyphosate, malathion, parathion, and tetrachlorvinphos*. Vol 112. IARC Monogr Prog, 2015:1–92.
4 European Food Safety Authority. Conclusion on the peer review of the pesticide risk assessment of the active substance glyphosate. *EFSA J* 2015;13:4302.
5 European Food Safety Authority. *Final Addendum to the Renewal Assessment Report*. 2015. http://registerofquestions.efsa.europa.eu/roqFrontend/outputLoader?output=ON-4302
6 De Roos AJ, Blair A, Rusiecki JA, *et al*. Cancer incidence among glyphosate-exposed pesticide applicators in the Agricultural Health Study. *Environ Health Perspect* 2005;113:49–54.
7 De Roos AJ, Zahm SH, Cantor KP, *et al*. Integrative assessment of multiple pesticides as risk factors for non-Hodgkin's lymphoma among men. *Occup Environ Med* 2003;60:E11.
8 Weisenburger DD. Pathological classification of non-Hodgkin's lymphoma for epidemiological studies. *Cancer Res* 1992;52(19 Suppl):5456s–62s; discussion 5462s-5464s.
9 Checkoway H, Pearce N, Kriebel D. Research methods in occupational epidemiology. 2nd edn. *Monographs in epidemiology and biostatistics*. New York: Oxford University Press, 2004:xiv, 372 p.
10 Rothman KJ, Greenland S, Lash TL. *Modern epidemiology*. 3rd edn. Philadelphia: Wolters Kluwer Health/Lippincott Williams & Wilkins, 2008:x, 758 p.
11 Schinasi L, Leon ME. Non-Hodgkin lymphoma and occupational exposure to agricultural pesticide chemical groups and active ingredients: a systematic review and meta-analysis. *Int J Environ Res Public Health* 2014;11:4449–527.
12 EPA. *Glyphosate; EPA Reg. # 524–308; mouse oncogenicity study*. 1985. V. Accession No. 251007–014. Tox review 004370. p.
13 JMPR. FAO plant production and protection paper, 178, 2004—pesticide residues in food—2004. *Report of the Joint Meeting of the FAO Panel of Experts on Pesticide Residues in Food and the Environment and WHO the Core Assessment Group*; Vol 178. Geneva, Switzerland: World Health Organization/Food and Agriculture Organization, 2004:95–169.
14 EPA. *Second peer review of Glyphosate*. 19911–19.
15 EPA. *Glyphosate—EPA Registration No. 524–308–2-Year Chronic Feeding/Oncogenicity Study in Rats with Technical Glyphosate*, I. William Dykstra. Toxicology Branch, Editor. 1991. V. MRID 416438–01 Tox review 008897. p.
16 EPA. *Glyphosate; 2-Year Combined Chronic Toxicity/Carcinogenicity Study in Sprague-Dawley Rats—List A Pesticide for Reregistration*, B. William Dykstra. Toxicology, Editor. 1991. V. MRID 416438–01. Tox review 008390. 1–29.
17 OECD. *Guidance Document 116 on the Conduct and Design of Chronic Toxicity and Carcinogenicity Studies*, H.a.S.P. Environment, Editor. Paris: OECD, 2012
18 European Chemicals Agency. *Guidance on the Application of the CLP Criteria: Guidance to Regulation (EC) No 1272/2008 on classification, labelling and packaging (CLP) of substances and mixtures*. Helsinki, Finland: European Chemicals Agency, 2015.
19 NRC Committee to Review the Styrene Assessment in The National Toxicology Program 12th Report on Carcinogens. *Review of the Styrene Assessment in the National Toxicology Program 12th Report on Carcinogens: Workshop Summary*; Washington DC: National Academies Press, 2014.
20 Keenan C, Elmore S, Francke-Carroll S, *et al*. Best practices for use of historical control data of proliferative rodent lesions. *Toxicol Pathol* 2009;37:679–93.
21 Haseman JK, Boorman GA, Huff J. Value of historical control data and other issues related to the evaluation of long-term rodent carcinogenicity studies. *Toxicol Pathol* 1997;25:524–7.
22 Greim H, Gelbke HP, Reuter U, *et al*. Evaluation of historical control data in carcinogenicity studies. *Hum Exp Toxicol* 2003;22:541–9.
23 Haseman JK, Huff J, Boorman GA. Use of historical control data in carcinogenicity studies in rodents. *Toxicol Pathol* 1984;12:126–35.
24 Giknis M, Clifford C. *Spontaneous Neoplastic Lesions in the Crl:CD-1(ICR)BR Mouse*. Charles River Laboratories; 2000.
25 Myers JP, vom Saal FS, Akingbemi BT, *et al*. Why public health agencies cannot depend on good laboratory practices as a criterion for selecting data: the case of bisphenol A. *Environ Health Perspect* 2009;117:309–15.
26 Buonsante VA, Muilerman H, Santos T, *et al*. Risk assessment's insensitive toxicity testing may cause it to fail. *Environ Res* 2014;135:139–47.
27 IARC Monograph 112. *List of Working Group Participants*. IARC Monogr Eval Carcinog Risks Hum 2015 26 March, 2015 [cited 2015 24 November]. http://monographs.iarc.fr/ENG/Meetings/vol112-participants.pdf
28 Portier CJ, *et al*. *Open Letter: Review of the Carcinogenicity of Glyphosate by EFSA and BfR*. 2015 [cited 2016 1/18/2016]. http://www.efsa.europa.eu/sites/default/files/Prof_Portier_letter.pdf
29 Url B. Response to Open Letter: Review of the Carcinogenicity of Glyphosate by EFSA and BfR. 2016. 1/13/2016. http://www.efsa.europa.eu/sites/default/files/EFSA_response_Prof_Portier.pdf

Document Two

Comments of Christopher J. Portier, PhD.
USEPA (EPA-HQ-OPP-2016-0385-0094)
Glyphosate Issue Paper: Evaluation of Carcinogenic Potential
October 4, 2016

Disclaimer: This work was done with my own resources and on my own time. I have received no reimbursement for any of these comments and no other party has contributed to the drafting of these comments. These comments are solely my opinion and my responsibility.

General Comments and Overall Summary

My comments on the glyphosate review by the USEPA (EPA-HQ-OPP-2016-0385-0094) is rather long and detailed. Realizing that the time and energies of the Science Advisory Panel (SAP) are limited, I will summarize my findings here. Each summarized finding is linked to the line(s) in my more technical review for those who wish to see more details. Because of my own limited time, I have chosen to focus my comments on the human evidence and the animal carcinogenicity evidence, foregoing the review of the other evidence presented. However, I will note that after reading the review on the mechanistic evidence relating to genotoxicity and oxidative stress, I still agree with the findings from the IARC Working Group that there is *strong evidence* that these mechanisms are operable.

Human Evidence Findings

1. The meta-analyses are improperly characterized by the EPA (lines 21-33)
2. The exposure-response relationship in the Agricultural Health Study (AHS) has greater weight than in the other studies, but has problems of its own (lines 39-45)
3. It is not clear in which direction possible confounding would alter the relative risks (lines 61-66) although possible confounding is an issue (line 68).
4. Recall bias is a concern, especially with the case-control studies (lines 70-72)
5. The EPA speculates without data that the more positive studies should have had lower relative risks than other studies (lines 77-80)
6. The follow-up time in the AHS study is likely to be too short to have seen an impact of the magnitude seen in the case-control studies and EPA does a poor job of characterizing the data they used to reach an opposite conclusion (lines 85-109)
7. The EPA speculates that earlier years of exposure prior to the start of the AHS would have effectively expanded the time on study in the AHS without any solid basis (lines 111-114)
8. The Bradford-Hill criteria outlined in the 1997 Guidelines for Carcinogenic Risk Assessment (GCRA) support a conclusion that a causal association in the epidemiology data is credible, but that chance, bias and or confounding could possibly explain the results. (lines 116-127)
9. EPA's interpretation that "*the association between glyphosate exposure and risk of NHL cannot be determined based on the available data*" does not correctly

characterize the human data presented. A better interpretation is that *"a positive association has been observed between exposure to the agent and cancer for which a causal interpretation is considered to be credible, but chance, bias or confounding could not be ruled out with reasonable confidence"*. This is the interpretation given these data by the IARC Working Group (lines 129-139)

Animal Carcinogenicity Data Findings

1. EPA's QCAR sets clear guidelines on evaluating animal cancer data with regard to when a high dose is exceeded (lines 151-159), how to interpret trend tests and pairwise comparisons (lines 163-169), how to use historical control data (lines 173-176) and what constitutes a valid historical control data set (180-182)
2. EPA has misinterpreted the language in OCSPP 870.4200 and OCSPP 870.4300 by assuming that an optional highest dose in an animal carcinogenicty study is also a threshold for inclusion of doses in their evaluation. In other words, 1000 mg/kg/day is not an upper bound, 5% in diet is the upper bound (lines 184-210)
3. I have individual comments on every rat study evaluated by the EPA (lines 212-287).
4. EPA consistently dismisses significant findings in rat studies because of a lack of a preneoplastic finding (studies listed starting a lines 217, 229, 277). This presumes that all mechanisms by which chemicals induce tumors in animals will involve enough stages that there would be a histologically identifiable preneoplastic lesion from which final tumors are formed. This simply is not the case and this criteria is applied without any concern for its validity by the EPA.
5. EPA consistently dismisses significant findings in rat studies because of a lack of a significant pairwise comparison even though there is a significant trend in violation of the GCRA (studies listed starting a lines 229, 255, 277).
6. EPA gives less weight to responses seen at doses above 1000 mg/kg/day in all rat studies, even though no dose exceeds 5% of feed. Considering that these findings are in studies with only 50-60 animals per group, that no study appears to have exceeded a maximum tolerated dose (as defined by the EPA and others), it is not clear why EPA does not accept these findings and then do an appropriate margin-of-exposure evaluation or linear extrapolation from these data to show a lack of risk in humans.
7. EPA's summary, which states that *"In 5 of the 9 rat studies conducted with glyphosate, no tumors were identified for detailed evaluation."* is misleading and fails to properly characterize the broad array of findings in these data (lines 291-322). In short, three of these studies were inadequate leaving 2 studies in Sprague-Dawley rats (1 positive) and four studies in Wistar rats (2 positive).
8. With only two studies in Sprague-Dawley rats, the strong positive response seen for thyroid c-cell carcinomas in female rats in one of these studies should be considered positive and due to exposure to glyphosate (lines 324-330)

9. I have individual comments on every mouse study evaluated by the EPA (lines 337-460).
10. EPA consistently dismisses significant findings in mouse studies because of a lack of a preneoplastic finding (studies listed starting a lines 342, 380). This presumes that all mechanisms by which chemicals induce tumors in animals will involve enough stages that there would be a histologically identifiable preneoplastic leasion from which final tumors are formed. This simply is not the case and this criteria is applied without any concern for its validity by the EPA.
11. EPA dismisses significant findings in ALL mouse studies because of a lack of a significant pairwise comparison even though there is a significant trend in violation of the GCRA (lines 337-460).
12. EPA gives less weight to responses seen at doses above 1000 mg/kg/day in all mouse studies, even though no dose exceeds 5% of feed. Considering that these findings are in studies with only 50-60 animals per group, that no study appears to have exceeded a maximum tolerated dose (as defined by the EPA and others), it is not clear why EPA does not accept these findings and then do an appropriate margin-of-exposure evaluation or linear extrapolation from these data to show a lack of risk in humans.
13. EPA uses an outside historical control dataset in one study (start line 380) to dismiss findings and fails to use an equally valid historical control data set identified by the IARC to assess the importance of renal tumors in another study (start line 342). A full evaluation of this second study using the historical control data identified by the IARC supports a strong positive finding in this study (lines 350-365).
14. EPA relies on two-sided p-values for trend tests when one-sided p-values would be more appropriate for identifying adverse effects (lines 367-370; 410-413; Tables 2,4,6)
15. EPA has serious errors in the use of a historical control population that uses data from animals that lived 24 months to compare to response in a study that only went 18 months (lines 388-408). When properly applied, the finding is significant compared to the historical control rate.
16. EPA excludes three positive findings in one study, identified by the European Food Safety Agency for which I sent them data prior to this current EPA review being released (lines 425-434)
17. EPA excludes positive results in a study in Swiss Albino mice because there is an infection in the animals that are not seen in any of the data evaluated by others and for which no documentation is provided (lines 445-460)
18. EPA summarizes the mouse data incorrectly (as they did with rats) when they state that *"No tumors were identified for detailed evaluation in 2 of the 6 mouse carcinogenicity studies."* One study had inadequate dosing and should have been excluded, and one study used Glyphosate trimesium salt rather than pure glyphosate. The remaining four mouse studies all had at least one positive finding (lines 464-475)
19. EPA did not analyze the consistency across mouse studies on the findings relating to renal tumors. I did (Tables 1-3, lines 498-532). Note, all studies were adjusted to an estimated 24 month response using the poly-3 adjustment (lines 487-517)

20. EPA did not analyze the consistency across mouse studies on the findings relating to malignant lymphomas. I did (Tables 1, 4-5, lines 536-541). Note, all studies were adjusted to an estimated 24 month response using the poly-3 adjustment (lines 487-517)
21. EPA did not analyze the consistency across mouse studies on the findings relating to heamngiosarcomas. I did (Tables 1, 6-7, lines 566-599). Note, all studies were adjusted to an estimated 24 month response using the poly-3 adjustment (lines 487-517)
22. Trends in male mice for malignant lymphomas and hemangiosarcomas remained even after doses above 1000 mg/kg/day were excluded (Tables 4-7, lines 536-599).
23. My conclusion is that the mouse data clearly indicates that glyphosate can induce malignant lymphomas and hemangiosarcomas in male CD-1 mice, even when doses above 1000 mg/kg/day are eliminated. There is also a suggestion that glyphosate can induce hemangiomas in female CD-1 mice. The mouse data also demonstrate that glyphosate can induce malignant lymphomas in male CD-1 mice and male Swiss Albino mice. Finally, the renal tumors seen in the CD-1 mice also appear in the Swiss Albino mice, supporting the role glyphosate plays in inducing these tumors. This is clearly sufficient evidence of the carcinogenicity of glyphosate in mice. (lines 573-600)

In summary, these data demonstrate an association in humans to NHL, evidence in rats for thyroid tumors, and very strong evidence in mice for renal tumors, hemangiosarcomas and malignant lymphomas. EPA's exclusion of doses above 1000 mg/kg/day is unscientific and their argument of a lack of significance above this dose is unsupported.

In every case where EPA could choose between a public health protective choice where slight weaknesses in a study or a lack of a very strong finding could raise concerns versus a choice where every study must be perfect and definitive otherwise it is not used, EPA has chosen to discard positive findings leaving them to finally conclude there is no concern. These data simply do not support a finding that glyphosate is *"not likely to be carcinogenic to humans"*.

EPA should declare glyphosate a probable human carcinogen and go on to do a risk assessment to determine if human exposure is sufficient to warrant concern. That resulting risk assessment should be reviewed by the Science Advisory Panel.

DETAILED TECHNICAL REVIEW

Human Evidence

The EPA's final conclusion on the evidence from human exposures to glyphosate and the risk of NHL is as follows:

Page 68: *"Based on the weight-of-evidence, the agency cannot exclude chance and/or bias as an explanation for observed associations in the database. Due to study limitations and contradictory results across studies of at least equal quality, a conclusion regarding the association between glyphosate exposure and risk of NHL cannot be determined based on the available data. The agency will continue to monitor the literature for studies and any updates to the AHS will be considered when available."*

The Agency provides many reasons for this finding. I would summarize them as follows:

1. *"All meta-analysis estimates reported were non-statistically significant except the meta-risk ratio reported by IARC (2015), which was borderline significant with the lower limit of the 95% CI at 1.03"*

Comment: In fact, there were three groups that did meta-analyses. Two were reported as significant (Schinasi and Leon, 2014 and IARC, 2015), although the IARC (2015) corrected an issue they saw with the Schinasi and Leon analysis. The IARC study showed a meta-RR of 1.3 with a confidence bound of (1.03-1.65). The other group (Chang and Delzel, 2016) provided four separate meta-analyses, all of which are reported as having a meta-RR of 1.3 with associated confidence bounds ranging from (1.0-1.6) to (1.0-1.8). Chang and Delzell presented only 1 significant digit for the lower confidence bounds and since their model 1 is exactly the same as the IARC model, they also had at least one significant finding. In fact they characterize their findings as *"we found marginally significant positive meta-RRs for the association between glyphosate use and risk of NHL"*. Thus, the data across all studies, when combined, point to a positive association between glyphosate and NHL in humans.

2. The exposure-response relationship seen in Eriksson et al. (2008) and McDuffie et al. (2001), even though significant, contradicted the exposure-response seen in the Agricultural Health Study (AHS).

Comment: There were 92 cases of NHL in the AHS, with 77.2% (71 cases) having some exposure, whereas the analysis of the tertiles to investigate exposure response relationships, used only 61 cases. Thus, 14% of the exposed cases were excluded. In comparison, both Eriksson et al. (a highly rated study by EPA) and McDuffie et al. were able to characterize all exposed individuals into their exposure groupings with zero loss. To characterize the exposure-response relationship in the AHS as superior to the other two studies is inappropriate.

3. Control for confounding varied across studies and there is a strong potential for

confounding by co-exposures to other pesticides.

Comment: This is correct with some studies doing better than others. However, the magnitude of the impact of this confounding differs by study as well. They cite the one case, Eriksson where the effect estimate went from 2.02 (1.10-3.71) unadjusted to 1.51 (0.77-2.94) adjusted. Others included in the meta-analysis are as follows: DeRoos et al. (2005), 1.2 (0.7-1.9) unadjusted, 1.1 (0.7-1.9) adjusted; DeRoos et al., 2003, 2.1 (1.1-4.0) unadjusted, 1.6 (0.9-2.8) adjusted; Hardell et al., 2002, 3.01 (1.08-8.52) unadjusted, 1.85 (0.55-6.20) adjusted). Orsi et al. (RR 1.0 (0.5-2.2)) and McDuffie et al. (RR 1.2 (0.83-1.74)) did not do analyses adjusting for other pesticides. EPA could remove these studies from the meta-analysis and redo it, but it is unlikely to dramatically change the overall results.

The EPA also expressed concern that what they see as a reduction when you correct for other pesticide exposures would carry over for other confounders. This is highly speculative since many of the NHL patients had no exposure to glyphosate and there are likely truck operators and mechanics (diesel exhaust fumes), factory workers (solvents) and other outdoor workers (UV radiation) in the cases and controls and the result of correcting for the confounders could go either way.

However, it is fair to say that confounding could not be ruled out in these studies.

4. Recall bias is a concern, especially in the case-control studies.

Comment: I agree.

5. The highest risk measures are coming from studies that would likely have lower exposures to glyphosate.

Comment: This is entirely speculative and is based upon an ecological assessment (glyphosate use has increased dramatically over time) and not upon actual data pertaining to the studies at hand. Nor does it fully account for the time since first exposure for the studies done with earlier cohorts.

6. The follow-up time in the DeRoos et al. (2005) study is sufficient that it should be given more weight than the other studies.

Comment: As noted by Portier et al., the median follow-up time in the AHS study was 6.7 years (not 7) and there is a question of whether this is long enough. EPA actually provides a solid argument for why there is concern. EPA gave three publications that they suggest puts the latency period for NHL between 1 and 25 years. Kato et al. (2005) in a high quality population-based, incidence case-control study looking at the relationships between organic solvent exposure and NHL in women found statistical significance only for women occupationally exposed prior to 1970 (cases and controls were recruited between 1995 and 1998) and cited two other studies with similar results (no reference given). They concluded this long latency was either due to higher exposures prior to 1970 or "*at least a 25 year*

latency period is required for NHL induction by these exposures". Weisenburger (1992), in discussing the problems with pathological identification of NHL and the known mechanisms in 1992 states that "The latency for NHL following an environmental exposure is largely unknown" then goes on to say that following chemotherapy for Hodgkin's disease, "the median latency is 5-6 years" based upon 44 case reports from two publications. I was unable to get a copy of one publication, but the publication by Jacquillat et al (1991) showed 24 patients, 17 of whom received radiation therapy along with chemotherapy, 5 radiation alone, three chemotherapy alone and one unknown. The latency ranged from 1 to 11 years in this paper (median 5.5 years) and up to 16 years in the other (abstract review only) These are rather extreme exposures relative to those from glyphosate and it would not be surprising for the glyphosate lag time to be longer than that from chemotherapy and radiation treatment, as suggested by Weisenberger et al. I was unable to obtain a copy of the third paper (Fontana et al., 1998) and the abstract provides no information on lag times.

The rest of the arguments are speculative dealing mostly with years of exposure prior to the beginning of the AHS. Without an analysis including this prior information on exposure with concurrent exposure, it is unclear that the resulting relative risks would go down or up.

Summary: The conclusion by the EPA that "*the association between glyphosate exposure and risk of NHL cannot be determined based on the available data*" fails to account for the overall strength of this evidence and the nature of that evidence. Using the Bradford-Hill criteria for causality described in the 2005 Guidelines for Carcinogenic Risk Assessment (GCRA), I would note that the observations are consistent (relative risks are positive, meta-analyses are positive), significant (in the meta-analysis), not specific (and as noted in the GCRA "*although the presence of specificity may support causality, its absence does not exclude it*"), temporally observed, shows a biological gradient, is coherent with the animal evidence (discussed later), has no experimental evidence from humans, and has no support from structure-activity relationships. So, is causality plausible here? Yes, absolutely. Is it demonstrated? No, clearly not. Are the findings possibly the result of chance, bias and or confounding? Yes, but more unlikely than likely.

The IARC Working Group concluded that there was "*limited evidence of carcinogenicity in humans*" from exposure to glyphosate where, as defined in the IARC Preamble, limited evidence means "*a positive association has been observed between exposure to the agent and cancer for which a causal interpretation is considered to be credible, but chance, bias or confounding could not be ruled out with reasonable confidence.*" This is a more accurate description of these data than that used by the EPA. If chance, bias and confounding could be ruled out, the IARC Working Group would have classified this as a "*known human carcinogen*", a much stronger finding. By arguing that "*the association between glyphosate exposure and risk of NHL cannot be determined based on the available data*", the EPA has given no weight to the human evidence in their final evaluation.

Animal Carcinogenicity Studies

According to the EPA, of the 9 available rat studies, 4 showed treatment related effects in various organs and of the 6 mouse studies they evaluated, 4 showed treatment effects in three tumors. In all cases, the EPA considers these findings to be not treatment related. I will first address the interpretations of individual studies, then discuss the entire package of studies.

Let's begin by repeating guidance from the GCRA :

"Other signs of treatment-related toxicity associated with an excessive high dose may include (a) significant reduction of body weight gain (e.g., greater than 10%), (b) significant increases in abnormal behavioral and clinical signs, (c) significant changes in hematology or clinical chemistry, (d) saturation of absorption and detoxification mechanisms, or (e) marked changes in organ weight, morphology, and histopathology. It should be noted that practical upper limits have been established to avoid the use of excessively high doses in long-term carcinogenicity studies of environmental chemicals (e.g., 5% of the test substance in the feed for dietary studies or 1 g/kg body weight for oral gavage studies [OECD, 1981])."

and

"A trend test such as the Cochran-Armitage test (Snedecor and Cochran, 1967) asks whether the results in all dose groups together increase as dose increases. A pairwise comparison test such as the Fisher exact test (Fisher, 1950) asks whether an incidence in one dose group is increased over that of the control group. By convention, for both tests a statistically significant comparison is one for which p is less than 0.05 that the increased incidence is due to chance. Significance in either kind of test is sufficient to reject the hypothesis that chance accounts for the result."

and

"Generally speaking, statistically significant increases in tumors should not be discounted simply because incidence rates in the treated groups are within the range of historical controls or because incidence rates in the concurrent controls are somewhat lower than average."

and

"The most relevant historical data come from the same laboratory and the same supplier and are gathered within 2 or 3 years one way or the other of the study under review; other data should be used only with extreme caution."

These guidelines are critical in the discussions that follow. I will note that the EPA assessment cites OCSPP 870.4200 and OCSPP 870.4300 several times referring to an upper limit for evaluating the high dose in a carcinogenicity study. Theses guidelines give multiple guidance for how to select the appropriate dose. Here are the first two:

(i) For risk assessment purposes, at least three dose levels should be used, in

> *addition to the concurrent control group. Dose levels should be spaced to produce a gradation of effects. A rationale for the doses selected must be provided.*
>
> *(ii) The highest-dose level should elicit signs of toxicity without substantially altering the normal life span due to effects other than tumors. The highest dose should be determined based on the findings from a 90–day study to ensure that the dose used is adequate to assess the carcinogenic potential of the test substance. Thus, the selection of the highest dose to be tested is dependent upon changes observed in several toxicological parameters in subchronic studies. The highest dose tested need not exceed 1,000 mg/kg/day.*

Nowhere in this guidance does it state that the high dose **cannot exceed** 1,000 mg/kg/day; just that it **does not need to exceed** that number. The EPA notes this fact on Page 69 of the Report, but then later interprets it as a hard limit for excluding doses. Because other data are used to justify the high dose that have not been presented here, we must assume that the highest doses used in the Guideline studies were at or near the maximum-tolerated dose (MTD) and wholly appropriate for the overall evaluation. Thus, 1,000 mg/kg/day is not a threshold for determining where to cut off the data. The only document discussing excessive doses is the QCRA which uses >5% in feed for feeding studies and all doses used here are below that threshold.

Rat Studies

Burnett et al., 1979 (MRID 00105164): As noted by EPA, this study is inadequate due to insufficiently high dose. This study should not be considered negative.

Lankas, 1981 (MRID 00093879): This study in Sprague-Dawley rats was considered inadequate due to the highest dose being far below the MTD. However, the study did see an increase in testicular tumors. These tumors were dismissed because of a non-monotonic dose-response (0%, 6%, 2%, 12% in increasing dose), a lack of pre-neoplastic findings and a range of historical controls (mean 4.5%, range 3.4% to 6.7%) that was higher than seen in the controls, inflating the p-value (as noted in the GCRA, this argument is not an acceptable argument). Nonetheless, the finding in the high exposure group is clearly significant against concurrent controls and, had they presented all of the historical control evidence, might have been significant there as well. Since no data for this tumor is presented for any other study, it is hard to determine if this finding is unique among the studies.

Stout and Ruecker, 1990 (MRID 41643801): The Sprague-Dawley rats in this study were given doses considerably higher (max 1183 mg/kg/day) than those in the Lankas study and was considered adequate by the EPA for evaluation, although they warn that tumor doses in the highest group will be given less weight because it is so high. They found a statistically significant increase in adenomas of the liver and the pancreatic islet cells in males. For pancreatic tumors, EPA points to a lack of clear dose-response (2%, 18%, 10% 15%) and unusually low background response (historical controls provided were 5% mean, 2.9%, 8.5%, 5.8%, 1.8%, 8.3%, 5.0% and 5.1%, all in control groups larger than the concurrent control in this study; since 2% is near 1.8% and only 7 controls are given, this is not an unusually low

response). For liver adenomas (5%, 4%, 6%, 15%), EPA cites a lack of pairwise significance, a plateau of dose-response in the middle dose groups and no preneoplastic lesions as reasons to reject these findings. No historical control data is presented.

In female rats, thyroid C-cell adenomas and combined adenomas and carcinomas were significantly elevated by trend test but not by pairwise comparison. Because of this, they concluded *"although there may be an indication of a dose-response in females, the increases observed in the glyphosate treated groups were not considered to be different than those observed in the concurrent controls"* ignoring their Guidelines regarding *"Significance in either kind of test is sufficient to reject the hypothesis that chance accounts for the result."* Here, preneoplastic lesions were observed, but no monotonic dose-response so they were ignored. Thyroid tumors in male rats were marginally significant (p~0.08).

Atkinson et al., 1993a (MRID 496317023): No adverse effects reported in Sprague-Dawley rats given doses in the same range as the Stout and Ruecker study. No data provided.

Brammer, 2001 (MRID 49704601): This is a two-year study in Wistar rats which showed a statistically significant trend in liver adenomas in male rats (0%, 4%, 0%, 10%) with a maximum dose of 1498 mg/kg/day. EPA provides three reasons for dismissing these findings: non-monotonic dose-response, higher survival in the controls, and multiple comparisons p-value adjustment.

Pavkov and Wyand 1987 (MRIDs 40214007, 41209905, 41209907): This is again a study in Sprague-Dawley rats (substrain given for this study). This study showed no significant findings. The EPA did not comment on the dosing used, however, the maximum dose used in this study was 55.7 mg/kg/day, not much difference from the doses used in Burnett (30 mg/kg/day) and Lankas (34 mg/kg/day) and far lower than doses showing no toxicity in Sprague-Dawley rats. This study should be considered inadequate by the EPA.

Suresh, 1996 (MRID 49987401): This two-year study in Wistar rats using a maximum dose of 886 mg/kg/day saw no significant increases in any tumors. Again, no details are given on tumors appearing in other studies.

Enemoto, 1997 (MRID 50017103-50017105): Also conducted in Wistar rats, but with a maximum dose of 1247 mg/kg/day, demonstrated no increases in tumors. Again, no details are given on tumors appearing in other studies.

Wood et al., 2009a (MRID 49957404): In a last study performed in Wistar rats with a maximum dose of 1229.7 mg/kg/day, a significant increase in female rat mammary tumors (adenomas and carcinomas combined) was observed (4%, 6%, 2%, 16%). EPA dismissed these findings based upon multiple comparisons and no pre-neoplastic lesions.

Excel, 1997: Excluded by the EPA because they had insufficient information on the study and an industry-sponsored review of the literature (Greim et al., 2015) stated it was *"unreliable"*. Greim et al. had multiple errors and considerable missing data (pointed out to EPA in a previous mailing) making it an unreliable source for this decision. No information

is given on this study in any available documents I was able to find including the review by EFSA.

Summary and Comments on the Rat Studies

All told, there are 9 rat studies presented, four in Wistar rats and 5 in Sprague-Dawley rats.

EPA states that *"In 5 of the 9 rat studies conducted with glyphosate, no tumors were identified for detailed evaluation."*, but two of these studies have inadequate dosing to identify.

They also state *"Some of the tumor incidences at the highest dose tested (approaching or exceeding 1,000 mg/kg/day for almost all studies) were statistically significant from concurrent controls using raw (unadjusted) p-values; however, none of the pairwise comparisons were found to be statistically significant following adjustment for multiple comparisons, except the testicular tumors seen in a single study. Furthermore, these high-dose tumors were given less weight."* However, as noted below in my calculation of the limit of 5% of compound in diet, the dose can easily go over 2000 mg/kg/day before reaching this value. They have confused the maximum gavage dose with the maximum dietary dose. These findings should carry equal weight as all other doses.

Three of the Sprague-Dawley rat studies used doses so low that the statistical power to detect an effect was compromised. Even still, one of these studies saw an increase in testicular tumors that was not noted in any other study and could be disregarded (provided there really is no response for this tumor in the other studies). In the remaining two studies (Stout/Ruecker and Atkinson), the EPA argues the highest dose *"exceeds the highest dose recommended in the test guidelines on how to conduct carcinogenicity studies"*. According to Laaksonen et al. (, Lab. Anim. 47(4) 245-56, 2013), Sprague-Dawley rats eat, on average, about 600g/kg/week at study start and about half that at 2 years. Based on the guidelines, 5% in diet is acceptable and on a daily basis would be between 2.1 g/kg/day to 4.3 g/kg/day; thus the <2 g/kg/day used in these two studies should be acceptable. This argument is not supported for these studies. These two studies differed on their findings of cancer with Atkinson negative for all cancers and Stout/Ruecker positive for two cancers, one in females and one in males. The remaining reasons for dismissing these findings include a lack of preneoplastic findings and a non-monotonic dose-response.

The thyroid tumors in female rats Stout/Ruecker) should be considered a positive finding. The dose-response is clear and the marginal findings in males should increase the concern for this tumor. There is no reason to believe that adenomas and carcinomas MUST arise from preneoplastic lesions in thyroid C-cell tumors. The rates from the other study for these tumors are no presented, but even if they had been, how do you judge one positive study against one negative study? The public protective decision in this case should be to conclude these tumors arose as a function of exposure to glyphosate.

The remaining tumors can be debated; in all cases where a decision could go either way, EPA dismisses findings rather than accepts them.

Mouse Studies

Reyna and Gordon, 1973 (MRID 00061113): This study is new to the EPA assessment. This study used doses as high as 50 mg/kg/day, far below the maximum doses used in the other studies that were below the maximum tolerated dose. In essence, this study is inadequate and should not be used for making a decision.

Knezevich and Hogan, 1983 (MRID 00130406): This is the first 24 month study in CD-1 mice at dietary doses of up to 6069 mg/kg/day according to EPA. EPA does not show their conversion from ppm in feed to mg/kg/day so it is unknown why this number is different from the European Food Safety Authority (EFSA) which lists the highest dose as 5874 mg/kg/day. The actual high dose used, according to EFSA, was 30,000 ppm or 3% in feed, below the EPA threshold given in the GCRA. According to EPA, *"No effect on survival was observed"* suggesting this high dose did not exceed the MTD.

This study saw an increase in kidney tubular cell adenomas and carcinomas (2%, 0%, 0%, 6%), a very rare tumor in these mice. Four reasons were given for discounting this finding: *"1) renal tubular cell tumors are spontaneous lesions for which there is a paucity of historical control data for this mouse stock; 2) there was no statistical significance in a pairwise comparison of treated groups with the concurrent controls and there was no evidence of a statistically significant linear trend; 3) multiple renal tumors were not found in any animal; and 4) compound-related nephrotoxic lesions, including pre-neoplastic changes, were not present in male mice in this study"*. In fact, the one-sided p-value (alternative is an increased risk) for this study was 0.03. In 1986, the EPA did have an adequate historical control population for these tumors and found they were highly statistically significant. The IARC also identified an adequate historical control population (Chandra and Frith, 1994) who reported only 1 tumor in 725 CD-1 mice also supporting a highly significant finding. As noted earlier, the second reason violates the QCRA if the one-sided test is applied. The third argument is not supported with such a small number of affected animals and a very rare tumor and the fourth reason, while arguable, presumes there would be a preneoplastic lesion rather than a unique mutational event to begin the cancer process.

Note: The raw p-value presented in Table 4.12 is for a two-sided test, a one-sided test is more appropriate here and has a raw p-value of 0.034. If the true control rate is 0.0014 as noted by Chandra and Frith (1994), the probability of seeing a finding more extreme than the one noted here is 0.0017. Even if the background is as high as 1%, the p-value would be 0.026.

Atkinson, 1993b (MRID 49631702): This 24 month study in CD-1 mice showed an increase in hemangiosarcomas (0%, 0%, 0%, 9%) which was statistically significant (p=0.003) with a marginally significant comparison between control and high dose of 0.053. The only negative comment given by the EPA on this study was *"however, the incidence of hemangiosarcomas at the high-dose was not statistically significant when compared to the concurrent controls"*, thus excluding the finding from the trend test because of a non-significant pairwise test, in violation of the QCRA.

Wood et al., 2009b (MRID 49957402): This study, also in CD-1 mice, was for 80 weeks (approximately 18 months) with a high dose in males of 810 mg/kg/day (again, not exceeding the 5% dose in feed). There was no effect on survival suggesting the study did not exceed the MTD. There was a monotonic increase in lung adenocarcinomas (10%, 10%, 14%, 22%) and a monotonic increase in malignant lymphomas (0%, 2%, 4%, 10%). For the lung cancers, the EPA again argued a lack of significance for pairwise comparisons (in violation

of their QCRA) and there was no evidence of progression from adenomas to carcinomas.

For the malignant lymphomas, the EPA noted that *"For this strain of mouse, the mean incidence for untreated animals is approximately 4.5% (range: 1.5%-21.7%) based on historical control data from Charles River (59 studies performed from 1987-2000; Giknis and Clifford, 2005) and Huntingdon Laboratories (20 studies from 1990-2002; Son and Gopinath, 2004)."* These controls are not from the same laboratory at the same time, but EPA did paraphrase the QCRA noting that these data *"should be used with caution"* whereas the GCRA states *"other data should be used only with extreme caution"*. In this case they did neither. The paper by Son and Gopinath documents the numbers of tumors seen in animals that die prior to 80 weeks out of 1453 males in 20 control groups. They saw a total of 36 animals with lymphomas, for a raw rate of 2.4%; however this is a lower bound on the rate since they did not look at all animals at 80 weeks to get obtain the number that are alive and having a tumor. It is not clear how EPA interpreted these numbers in their presentation. The study by Giknis and Clifford (2005) had 52 studies (not 59) and only 26 of them were for 18 months; the rest were for 2 years and these last 26 would be inappropriate as a historical control. The numbers cited by the EPA *("4.5% (range: 1.5%-21.7%)"*) are directly out of Giknis and Clifford for all 52 studies and the range fails to include the 11 studies with no tumors (lower end of range is 0). In the 26 studies ending at 18 months, Giknis and Clifford saw tumor incidence as follows (0/60, 0/50, 0/50, 0/50, 0/50, 0/50, 0/50, 0/50, 1/69, 1/50, 1/50, 1/50, 1/50, 1/50, 1/50, 1/50, 2/60, 2/59, 2/53, 2/50, 2/47, 2/46, 3/60, 3/59, 4/49, 7/50) thus ranging from 0% to 14% with a weighted mean of 2.5%.

NOTE: The p-value sited by EPA for the trend test is the two-sided p-value; a one sided p-value is more appropriate and the correct value is 0.0043. If you assume that 2.5% is the historical control rate, the probability of seeing a more significant finding that the one seen in this study is 0.0079.

Sugimoto, 1997 (MRID 50017108 - 50017109): In another study in CD-1 mice (with sub strain noted), mice were given, for 18 months, a maximum dose of 40,000 ppm of glyphosate which is 4% in the diet, again below the 5% in feed set by the QCRA. The second highest dose was 0.8% in diet. This study demonstrated a clear dose-response for hemangioma in female mice (0%, 0%, 4%, 10%) with a p-value for trend of p=0.002 by EPA's calculation. There were no treatment effects on survival suggesting this dose did not exceed the MTD. This tumor was not considered treatment related by the EPA because of no pairwise significance with the high dose versus control using a multiple comparisons analysis (the uncorrected p-value is 0.028 and the corrected p-value is 0.055).

What is not mentioned by the EPA but was evaluated by the EFSA, was the dose-response trend for hemangiosarcoma in male mice for which the one-sided p-value for trend is 0.008. Here the responses are 0%, 0%, 0% and 4%, a very low response rate. However, this is only an 18 month study, so low rates of tumors are to be expected.

What is also not mentioned are the malignant lymphomas and kidney tumors also found in males in this study (EFSA, 2015). The renal tumors had rates of 0%, 0%, 0%, 4% (the same as the hemangiosarcomas in males) with a p-value for trend of 0.008. The malignant lymphomas had rates of 4%, 4%, 0%, 12% with a p-value for trend of 0.008. I will compare these rates to those seen in the other studies later.

Pavkov and Turnier, 1987 (MRIDs 40214006, 41209907): This is a two-year chronic toxicity study in CD-1 mice with a maximum dose of 991 mg/kg/day. They list this study as completely negative for any cancer findings. However, this study evaluated Glyphosate trimesium salt (52.6% pure). No details on this study are provided by the EPA and I could find no other regulatory body that has reviewed this study nor is it listed in the Greim et al. (2015) manuscript. It is also the only carcinogenicity study with such a low percentage of pure glyphosate.

Kumar, 2001: This 18-month chronic carcinogenicity study in Swiss Albino mice with high-dose exposures of 10,000 ppm (1% diet) was excluded by the EPA *"due to the presence of a viral infection within the colony, which confounded the interpretation of the study findings"*. No information on this viral infection is given in the EPA Assessment. It is not possible to determine where this information on a viral infection came from. In the most recent draft classification document on glyphosate by the European Chemical Agency, they state that *"in the study report itself, there was no evidence of health deterioration due to suspected viral infection and, thus, the actual basis of EPA's decision is not known"* when referring to this study. The only reference I can find is from the paper by Greim et al. who down-rated the study *"based on speculation of a viral infection within the colony"*.

This study is important as they saw increases in kidney tumors (0%, 0%, 2%, 4%) and malignant lymphomas (20%, 30%, 32%, 38%) with one-sided p-values for trend of 0.04 and 0.05 respectively. While these are not strikingly strong p-values, they show a consistency in the male mouse data for these tumors.

Summary and Comments on the Mouse Studies

EPA concluded that *"No tumors were identified for detailed evaluation in 2 of the 6 mouse carcinogenicity studies."* One of these mouse studies should have been excluded because of the low doses used in the study. The other study has no details provided by the EPA or any other regulatory body and uses Glyphosate trimesium salt (52.6% pure).

EPA then concluded *"In the remaining 4 mouse studies, 3 observed a statistically significant trend in tumor incidences in the hemangiosarcomas, lung adenomas, malignant lymphomas or hemangiomas; however, the agency determined that none of the tumors observed in the mouse are treatment related."* In fact, there were 5 additional studies since they excluded the one study in Swiss Albino mice because of an infection in the study animals that appears to be speculative. Let's consider these 5 remaining studies. Since the hemangiomas only occurred in one study in female mice, I will not discuss it further.

Table 1 provides a summary of the findings in the 5 studies for which I could find sufficient data to make a comparison across the three main tumor findings in male mice: renal tumors, hemangiosarcoms and malignant lymphomas. A review of all of the studies in one simple picture illustrates the consistency of the findings across the various studies. Now, let's compare the actual tumor rates to see how they compare.

Table 1: Cancer findings in studies of glyphosate in male mice

Year	Strain	Length[1]	Top Dose[2]	Renal Tumors	Hemangio-sarcomas	Malignant Lymphoma
1983[5]	Crl:CD-1	24	4,841	+[3]		
1993[5]	?:CD-1	24	1,000		+	+/-[4]
1997	CrJ:CD-1	18	4,843	+[5]	+[5]	+[5]
2001	Swiss	18	1,460	+[5]	**No Data**	+[5]
2009	Crl:CD-1	18	810			+

Cancer increases in risk generally as a power of length of exposure (1). This relationship was used to develop a means to adjust the length of time an animal is on a study, enabling a scientist to determine risk at the end of two-years, the typical time used for animal bioassays (2, 3). This is called the Poly-3 adjustment. The US National Toxicology Program uses the Poly-3 test to evaluate significance in their animal bioassays. Now you will note that three of the mouse studies were only conducted for 18 months. (Comparing 18 month studies with 24 month studies without making an adjustment for the differences in length of exposure is like comparing cancer rates in 40 year-olds exposed for 20 years to cancer rates in 65 year-olds exposed for 45 years and concluding they are not consistent with each other; the conclusion is meaningless because the correct evaluation was not done.) Thus, in order to compare all 5 studies, we must use the Poly-3 adjustment to extrapolate the 18 month studies to estimate what we think the cancer risk would have looked like at 24 months. The adjustment decreases the number of animals without tumors in all groups in the 18 month studies by $(18/24)^3$. The one-sided p-values for both the unadjusted trend test and the poly-3 adjusted trend test are given in Table 2 for male mouse renal tumors.

[1] months
[2] mg/kg/day
[3] indicates p-value for trend <0.05
[4] p=0.08
[5] not evaluated by the EPA

Table 2: Analysis of Male Mouse Renal Tumors From the Individual Studies

Year	Strain	Length	Doses (mg/kg/d)	Response	p-Trend (p-poly3)
1983	Crl:CD-1	24	157, 814, 4841	1/50, 0/49, 1/50, 3/50	0.03 (0.03)
1993	?:CD-1	24	100, 300, 1000	2/50, 2/50, 0/50, 0/50	0.94 (0.94)
1997	CrJ:CD-1	18	165, 838, 4348	0/50, 0/50, 0/50, 2/50	0.008 (0.009)
2001	SW	18	15, 151, 1460	0/49, 0/49, 1/50, 2/50	0.04 (0.04)
2009	Crl:CD-1	18	71, 234, 810	0/51, 0/51, 0/51, 0/51	-

As an example of how the Poly-3 adjustments work, consider a comparison of the high-dose renal tumor response in the 1983 study (3/50=6%) to the high-dose response in the 1997 study (2/50=4%). In the 1997 study, 48 animals had no tumors at 18 months; the poly-3 adjustment reduces this to 20.25 leading to an incidence estimate of 2/22.25=9%. Because the Poly3 test effectively reduces the number of animals on study, even though the incidence estimate goes up, the p-value for the trend test goes down. Numerous evaluations of the validity of the poly-3 adjustment have been published in the peer-reviewed literature and it seems to work very well.

Now that the lengths of the studies have been adjusted, the next question to ask is whether this dose-response is consistent across all of the studies or whether there are anomalies. Combining all of the studies into one analysis can help us to evaluate this question; if the pooled data are no longer significant or less significant, the studies are not consistent and do not complement each other. Combining all of the studies into one pooled analysis and performing a trend analysis on the pooled data yields highly significant findings (Table 3, Line 1). Excluding the Swiss Albino mouse study and only using the CD-1 mice also yields a significant trend (Table 3, Line 2). Repeating these analyses with the Poly-3 adjusted data does not alter the significant findings. Since EPA is concerned about doses above 1000 mg/kg/day, I excluded doses above this dose and re-analyzed the data. The results of the restricted analysis are shown in Table 3, Lines 3-4. Without the doses above 1000 mg/kg/day, the effect disappears.

Table 3: Pooled Analysis of Male Mouse Renal Tumors

Year	Strain	p-Trend (p-poly3)
All Combined	CD-1 and Swiss	0.0004 (0.001)
CD-1 Combined	CD-1	0.001 (0.001)
All Combined, doses>1000 dropped	CD-1 and Swiss	0.80 (0.84)
CD-1 Combined, doses>1000 dropped	CD-1	0.85 (0.86)

Tables 4 and 5 repeat these analyses for malignant lymphomas. Because of the different backgrounds between the Swiss mice and the CD-1 mice, when they are all combined, the joint analysis is not significant (Table 5, line 1). Removing the Swiss mouse study and only evaluating the CD-1 mice leads to highly significant trends in all analyses (Table 5, lines 2). A significant trend remains in CD-1 mice even after removing the doses>1000 mg/kg/day (Table 5, line 4) suggesting this is not a high-dose only effect.

Table 4: Analysis of Male Mouse Malignant Lymphoma From the Individual Studies

Year	Strain	Length	Doses (mg/kg/d)	Response	p-Trend (p-poly3)
1983	Crl:CD-1	24	157, 814, 4841	2/50, 5/49, 4/50, 2/50	0.51 (0.51)
1993	?:CD-1	24	100, 300, 1000	4/50, 2/50, 1/50, 6/50	0.08 (0.08)
1997	CrJ:CD-1	18	165, 838, 4348	2/50, 2/50, 0/50, 6/50	0.008 (0.012)
2001	SW	18	15, 151, 1460	10/49, 15/49, 16/49, 19/49	0.05 (0.09)
2009	Crl:CD-1	18	71, 234, 810	0/51, 1/51, 2/51, 5/51	0.004 (0.005)

552
553 **Table 5: Pooled Analysis of Male Mouse Malignant Lymphoma**
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Year	Strain	p-Trend (p-poly3)
All Combined	CD-1 and Swiss	0.17 (0.19)
CD-1 Combined	CD-1	0.02 (0.01)
All Combined, doses>1000 dropped	CD-1 and Swiss	0.86 (0.93)
CD-1 Combined, doses>1000 dropped	CD-1	0.03 (0.05)

555
556 Tables 6 and 7 repeat these analyses for hemangiosarcomas. The findings in the Swiss
557 mouse were unavailable so Tables 6 and 7 only contain analyses of the CD-1 mouse data.
558 All pooled analyses are highly significant (Table 7) and they remain significant if
559 doses>1000 are excluded (Table 7, line 2). So again, this is not a high dose-only effect.
560
561 **Table 6: Analysis of Male Mouse Hemangiosarcomas From the Individual Studies**
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Year	Strain	Length	Doses (mg/kg/d)	Response	p-Trend (p-poly3)
1983	Crl:CD-1	24	157, 814, 4841	0/50, 0/49, 1/50, 0/50	0.63 (0.63)
1993	?:CD-1	24	100, 300, 1000	0/50, 0/50, 0/50, 4/50	0.0004 (0.0004)
1997	CrJ:CD-1	18	165, 838, 4348	0/50, 0/50, 0/50, 2/50	0.008 (0.009)
2001	SW	18	15, 151, 1460	No Data	-
2009	Crl:CD-1	18	71, 234, 810	0/51, 0/51, 0/51, 0/51	-

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Table 7: Pooled Analysis of Male Mouse Hemangiosarcomas

Year	Strain	p-Trend (p-poly3)
CD-1 Combined	CD-1	0.02 (0.03)
CD-1 Combined and Doses Pooled[1]	CD-1	0.02 (0.02)
CD-1 Combined, doses>1000 dropped	CD-1	<0.0001 (<0.0001)
CD-1 Combined, doses>1000 dropped and Doses Pooled[2]	CD-1	0.0003 (0.0003)

In summary, the results seen for renal tumors, malignant lymphomas and hemangiosarcomas in male mice in the 4 CD-1 studies for which the data were available are consistent and have a much stronger trend when all of the data are combined. The trend tests for malignant lymphomas and hemangiosarcomas in these studies remain significant when doses above 1000 mg/kg/day are eliminated.

EPA's approach has been to eliminate each study separately, generally by arguing the dose is too high (even though no signs of exceeding the MTD are apparent and their guidelines do not support the cut-off they are using), that there are no precursor lesions (suggesting cancer cannot arise without precursor lesions which is not a scientific necessity), and that the pairwise comparisons are not significant so the trend test should be ignored (in violation of their own guidelines). In addition, EPA has failed to present all of the positive tumor sites seen in these mouse studies, they have incorrectly used (probably inappropriate) historical controls and when these are used correctly a significant finding remains, they have included studies that should have been dismissed due to power issues, have included a study for which there is almost no available information other than the one paragraph they have presented, and have not evaluated the data across the studies to look for consistency in the response for tumors that appear in multiple studies. In essence, this is a very weak scientific evaluation of the available mouse carcinogenicity data.

My conclusion is that the mouse data clearly indicates that glyphosate can induce malignant lymphomas and hemangiosarcomas in male CD-1 mice, even when doses above 1000 mg/kg/day are eliminated. There is also a suggestion that glyphosate can induce hemangiomas in female CD-1 mice. The mouse data also demonstrate that glyphosate can induce malignant lymphomas in male CD-1 mice and male Swiss Albino mice. Finally, the renal tumors seen in the CD-1 mice also appear in the Swiss Albino mice supporting the role glyphosate plays in inducing these tumors. This is clearly sufficient evidence of the carcinogenicity of glyphosate in mice.

1. Portier CJ, Hedges JC, Hoel DG. Age-specific models of mortality and tumor onset for historical control animals in the National Toxicology Program's carcinogenicity experiments. *Cancer Res* (1986) **46**(9):4372-8. PubMed PMID: 3731095.

2. Bailer AJ, Portier CJ. Effects of treatment-induced mortality and tumor-induced mortality on tests for carcinogenicity in small samples. *Biometrics* (1988) **44**(2):417-31. PubMed PMID: 3390507.

3. Portier CJ, Bailer AJ. Testing for increased carcinogenicity using a survival-adjusted quantal response test. *Fundam Appl Toxicol* (1989) **12**(4):731-7. PubMed PMID: 2744275.

Document Three

Table 1: Human Epidemiology Studies

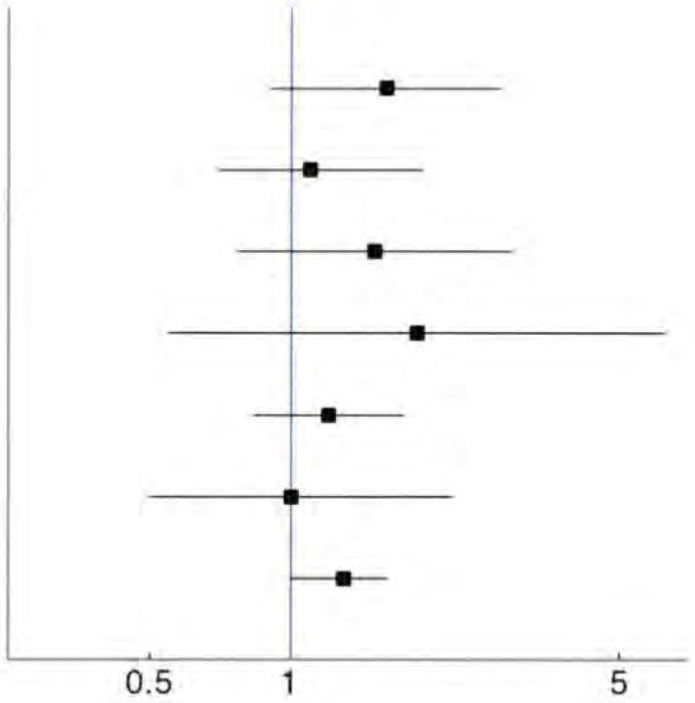
Study	Type	Size	Findings	Exposed Cases
Agricultural Health Study *(De Rooset al., 2005)*	Cohort – licensed pesticide applicators	52 395 (+32 347 spouses), 92 cases, 4-8 years follow-up	1.1 (0.7-1.9) C 0.7 (0.4-1.4) 21-56% tertile compared to <20% tertile 0.9 (0.5-1.6) 21-56% tertile compared to >57% tertile (31 cases no quantification of exposure)	73
US Midwest *(De Roos et al., 2003)*	Pooled analysis 3 case-control studies	NHL: 650 cases, 1933 controls	2.1 (1.1-4) U 1.6 (0.9-2.8) C	36 36
Cross-Canada *(McDuffie et al., 2001)*	Population-based case-control study	517 cases, 1506 controls	1.2 (0.83-1.74) U 1.0 (0.63-1.57) ≤2 d/Y 2.12 (1.2-3.73) >2 d/Y	51 28 23
Swedish Case-Control Study *(Eriksson et al., 2008)*	Population-based case-control study	910 cases, 1016 control	2.02 (1.1-3.71) U 1.51 (0.77-2.94) C 1.69 (0.7-4.07) ≤10 d/Y 2.36 (1.04-5.37) >10 d/Y 1.11 (0.24-5.08) ≤10 Y 2.26 (1.16-4.4) >10 Y	29 29 12 17 NR NR
Swedish Case-Control Study *(Hardell et al., 1999)*	Population-based case-control study	404 cases, 741 control (limited power)	2.3 (0.4-1.3) U 5.8 (0.6-5.4) C (not specified)	4 NR
France Case-Control *(Orsi et al, 2009)*	Hospital-based case-control study	244 cases, 456 controls	1.0 (0.5-2.2) U	12
Swedish Case-Control Study *(Hardell et al., 2002)*	Population-based case-control study	515 cases, 1141 controls	3.04 (1.08-8.5) U 1.85 (0.55-6.2) C (notspecified)	8 8
US Case-Control Study *(Lee et al., 2004)*	Population-based case-control study	872 cases, 2381controls	1.4 (0.98-2.1) U – no asthma 1.2 (0.4-3.3) U - asthma	53 6

Table 2: Meta Analyses

Study	Included Studies	Findings
Schinasi and Leon, 2014	McDuffie et al., 2001; Hardell et al., 2002; De Roos et al., 2003 and 2005; Eriksson et al., 2008; Orsi et al., 2009)	1.5 (1.1-2.0)
IARC Monograph Working Group	McDuffie et al., 2001; Hardell et al., 2002; De Roos et al., 2003 and 2005; Eriksson et al., 2008; Orsi et al., 2009)	1.3 (1.103-1.65) – used adjusted risk estimates from Hardell et al., 2003 and Eriksson et al., 2008
Chang and Delzell, 2016	McDuffie et al., 2001; Hardell et al., 2002; De Roos et al., 2003 and 2005; Eriksson et al., 2008; Orsi et al., 2009)	1.3 (1.0-1.6)

Figure 1: Tree Plot of Epidemiology Studies

Study	RR	Lower	Upper	Weight
De Roos et al. (2003)	1.600	0.900	2.800	16.2
De Roos et al. (2003)	1.100	0.700	1.900	21.0
Eriksson et al., (2008)	1.510	0.770	2.940	11.6
Hardell et al. (2002)	1.850	0.550	6.200	3.6
McDuffie et al. (2001)	1.200	0.830	1.740	38.1
Oris et al. (2009)	1.000	0.500	2.200	9.5
Meta-Analysis	1.300	1.000	1.600	100.0



Chang and Delzell (2016)

Table 3: Carcinogenicity Studies in Male Mice

Year	Strain	Length[1]	Top Dose[2]	Renal Tumors	Hemangio-sarcomas	Malignant Lymphoma
1983[5]	Crl:CD-1	24	4,841	+[3]		
1993[5]	?:CD-1	24	1,000		+	+/-[4]
1997	CrJ:CD-1	18	4,843	+	+	+
2001	SW	18	1,460	+	**Data Not Available**	+
2009	Crl:CD-1	18	810			+

1 – months; 2 – mg/kg bw/day; 3 - + indicates a p-value of <0.05 as calculated by BfR using the Armitage linear trend test in proportions; 4 – p=0.054; 5 – studies evaluated in IARC review; p=0.08

Table 4: Analysis of Male Mouse Renal Tumors From the Individual Studies

Year	Strain	Length	Doses (mg/kg/d)	Response	p-Trend (p-poly3)
1983	Crl:CD-1	24	157, 814, 4841	1/50, 0/49, 1/50, 3/50	0.03 (0.03)
1993	?:CD-1	24	100, 300, 1000	2/50, 2/50, 0/50, 0/50	0.94 (0.94)
1997	CrJ:CD-1	18	165, 838, 4348	0/50, 0/50, 0/50, 2/50	0.008 (0.009)
2001	SW	18	15, 151, 1460	0/49, 0/49, 1/50, 2/50	0.04 (0.04)
2009	Crl:CD-1	18	71, 234, 810	0/51, 0/51, 0/51, 0/51	-

Table 5: Pooled Analysis of Male Mouse Renal Tumors

Year	Strain	p-Trend (p-poly3)
All Combined	CD-1 and Swiss	0.0004 (0.001)
All Combined and Doses Pooled[1]	CD-1 and Swiss	0.0008 (0.002)
CD-1 Combined	CD-1	0.001 (0.001)
CD-1 Combined and Doses Pooled[1]	CD-1	0.001(0.001)
All Combined, doses>1000 dropped	CD-1 and Swiss	0.80 (0.84)
All Combined, doses>1000 dropped and Doses Pooled[2]	CD-1 and Swiss	0.39 (0.40)
CD-1 Combined, doses>1000 dropped	CD-1	0.85 (0.86)
CD-1 Combined, doses>1000 dropped and Doses Pooled[2]	CD-1	0.80 (0.80)

[1] – Doses were combined as follows: all controls, doses between 0 and 310 mg/kg/day, doses between 310 and 1500 mg/kg/day, and doses greater than 1500 mg/kg/day. Average doses in each pooled group were used in the analysis. [2] - Doses were combined as follows: all controls, doses between 0 and 310 mg/kg/day, and doses between 310 and 1500 mg/kg/day. Average doses in each pooled group were used in the analysis.

Figure 2: Renal tumors in male mice poly-3 adjusted showing individual dose groups

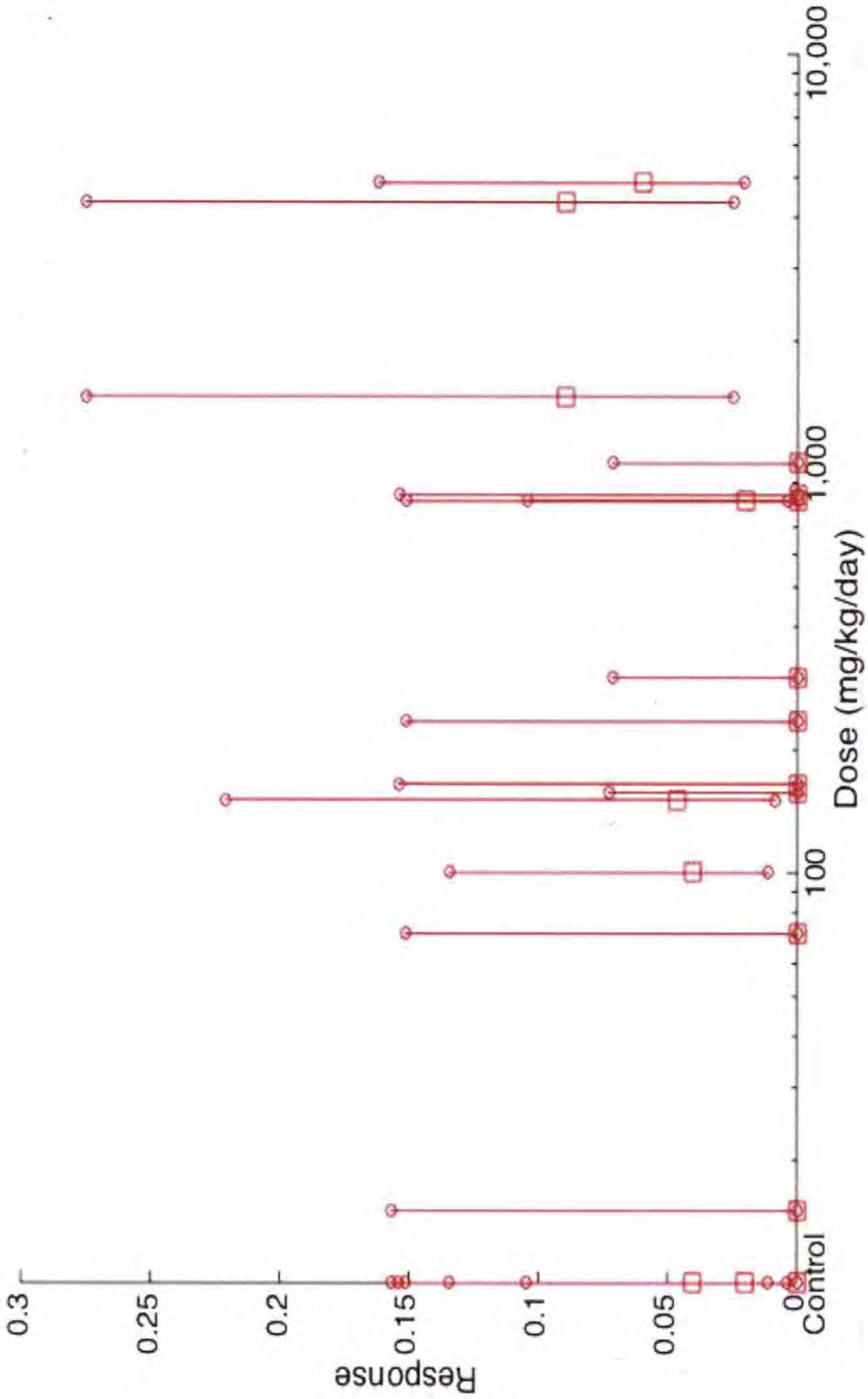


Figure 3: Renal tumors in male mice poly-3 adjusted and clustered by similar doses

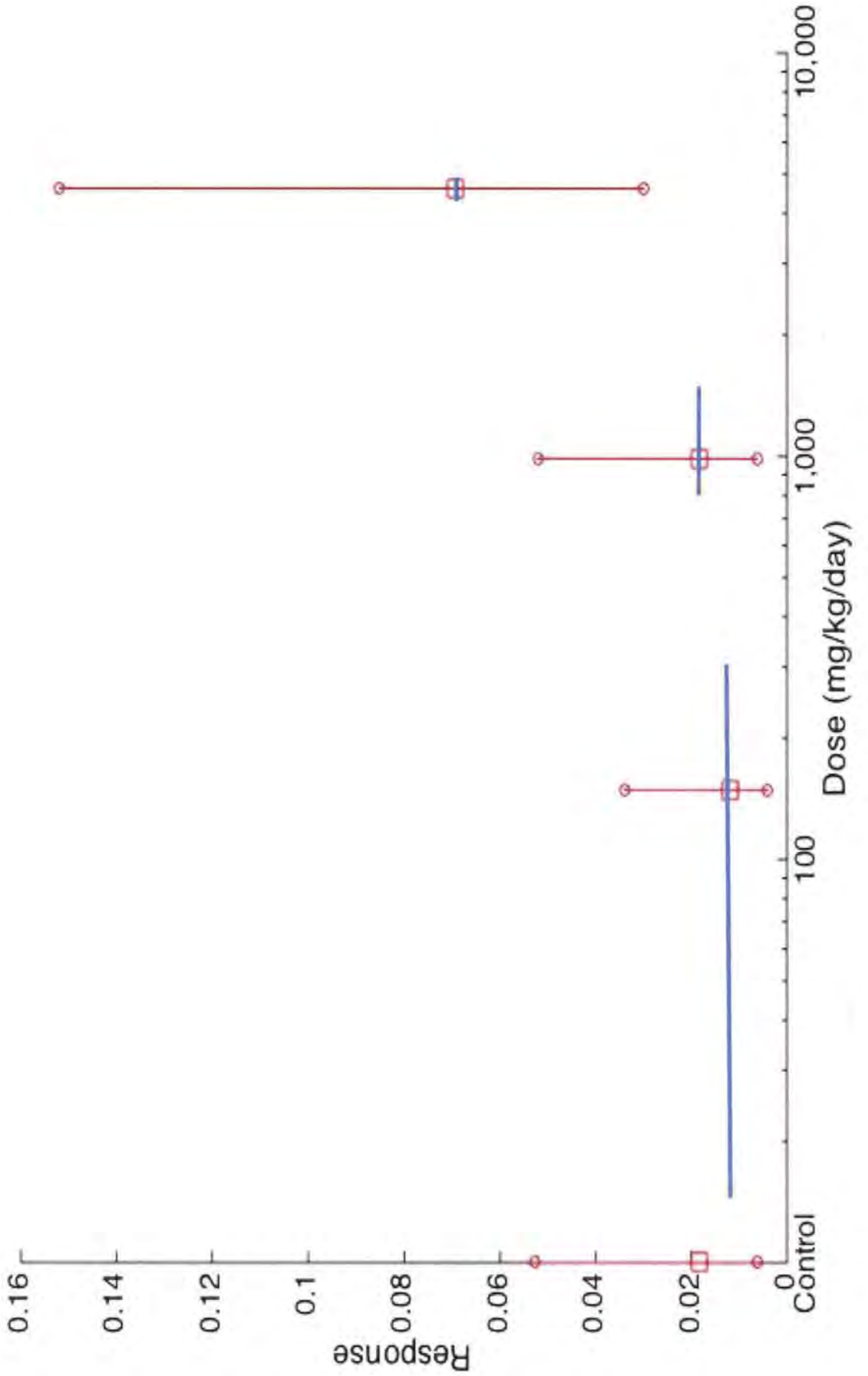


Table 6: Analysis of Male Mouse Malignant Lymphoma From the Individual Studies

Year	Strain	Length	Doses (mg/kg/d)	Response	p-Trend (p-poly3)
1983	Crl:CD-1	24	157, 814, 4841	2/50, 5/49, 4/50, 2/50	0.51 (0.51)
1993	?:CD-1	24	100, 300, 1000	4/50, 2/50, 1/50, 6/50	0.08 (0.08)
1997	CrJ:CD-1	18	165, 838, 4348	2/50, 2/50, 0/50, 6/50	0.008 (0.012)
2001	SW	18	15, 151, 1460	10/49, 15/49, 16/49, 19/49	0.05 (0.09)
2009	Crl:CD-1	18	71, 234, 810	0/51, 1/51, 2/51, 5/51	0.004 (0.005)

Table 7: Pooled Analysis of Male Mouse Malignant Lymphoma

Year	Strain	p-Trend (p-poly3)
All Combined	CD-1 and Swiss	0.17 (0.19)
All Combined and Doses Pooled[1]	CD-1 and Swiss	0.32 (0.31)
CD-1 Combined	CD-1	0.02 (0.01)
CD-1 Combined and Doses Pooled[1]	CD-1	0.01(0.009)
All Combined, doses>1000 dropped	CD-1 and Swiss	0.86 (0.93)
All Combined, doses>1000 dropped and Doses Pooled[2]	CD-1 and Swiss	0.02 (0.03)
CD-1 Combined, doses>1000 dropped	CD-1	0.03 (0.05)
CD-1 Combined, doses>1000 dropped and Doses Pooled[2]	CD-1	0.04 (0.04)

[1] – Doses were combined as follows: all controls, doses between 0 and 310 mg/kg/day, doses between 310 and 1500 mg/kg/day, and doses greater than 1500 mg/kg/day. Average doses in each pooled group were used in the analysis. [2] - Doses were combined as follows: all controls, doses between 0 and 310 mg/kg/day, and doses between 310 and 1500 mg/kg/day. Average doses in each pooled group were used in the analysis.

Figure 4: Malignant lymphomas in male mice poly-3 adjusted showing individual dose groups

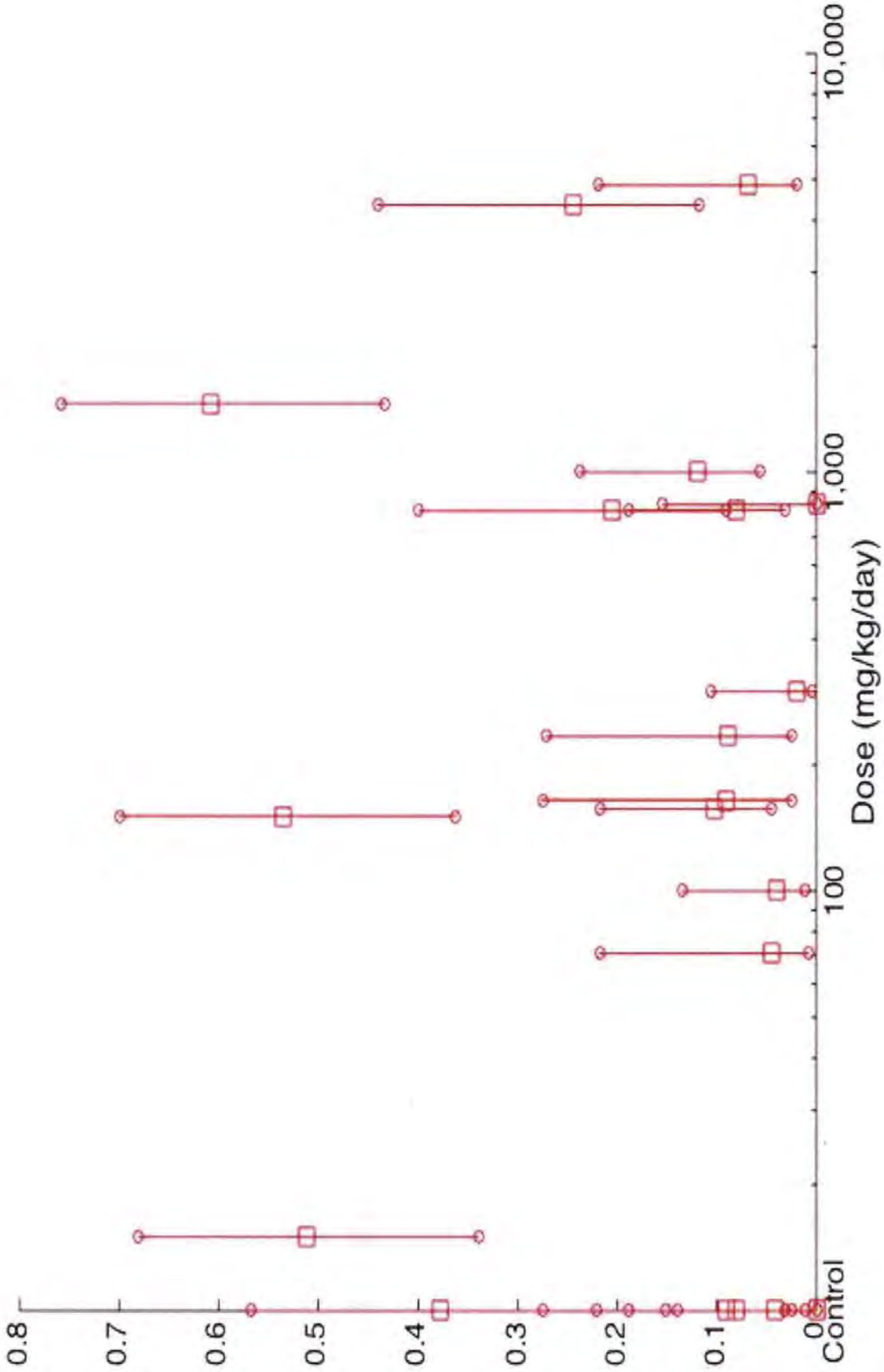


Figure 5: Malignant lymphomas in male CD-1 mice poly-3 adjusted showing individual dose groups

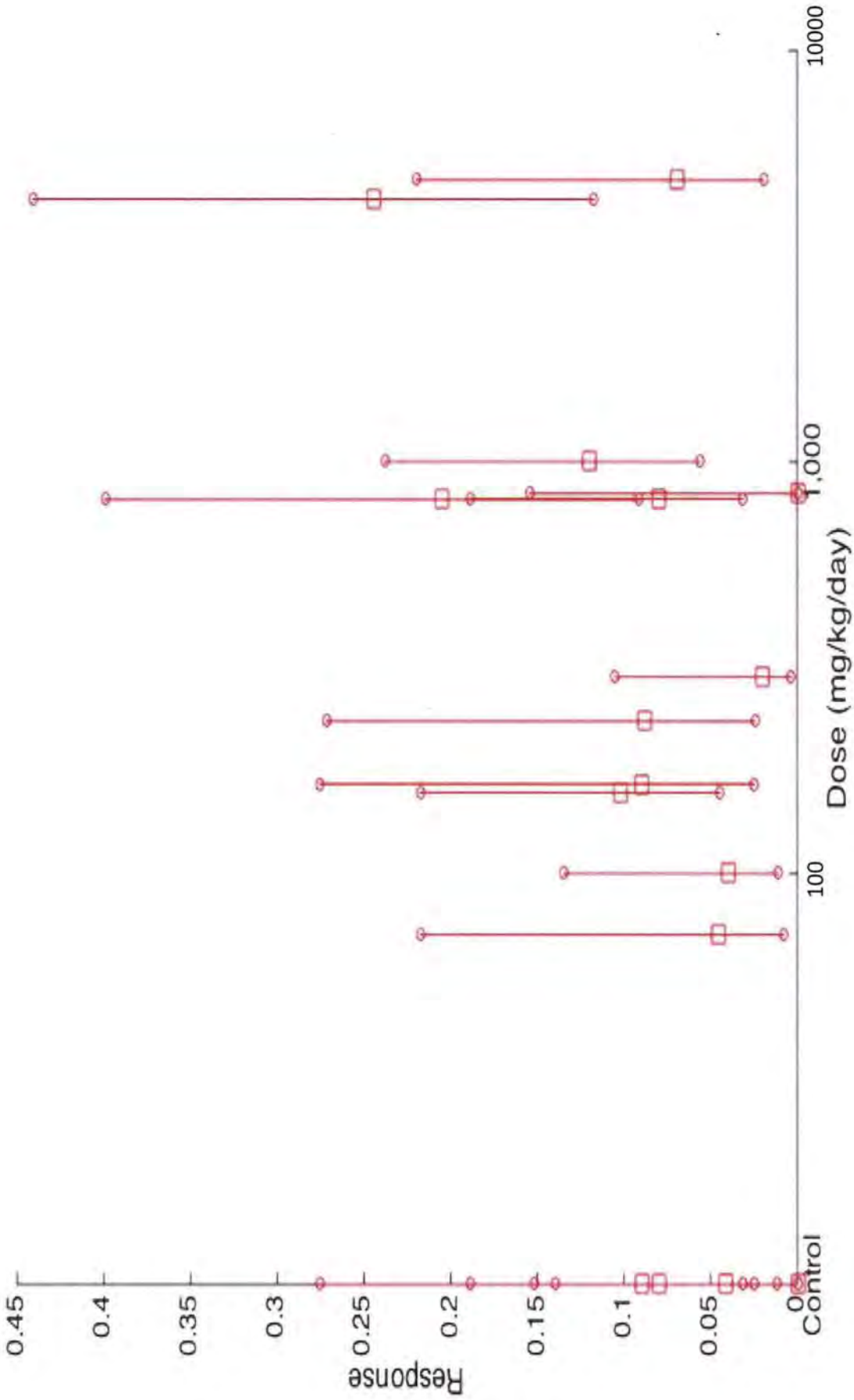


Figure 6: Malignant lymphomas in male CD-1mice poly-3 adjusted and clustered by similar doses

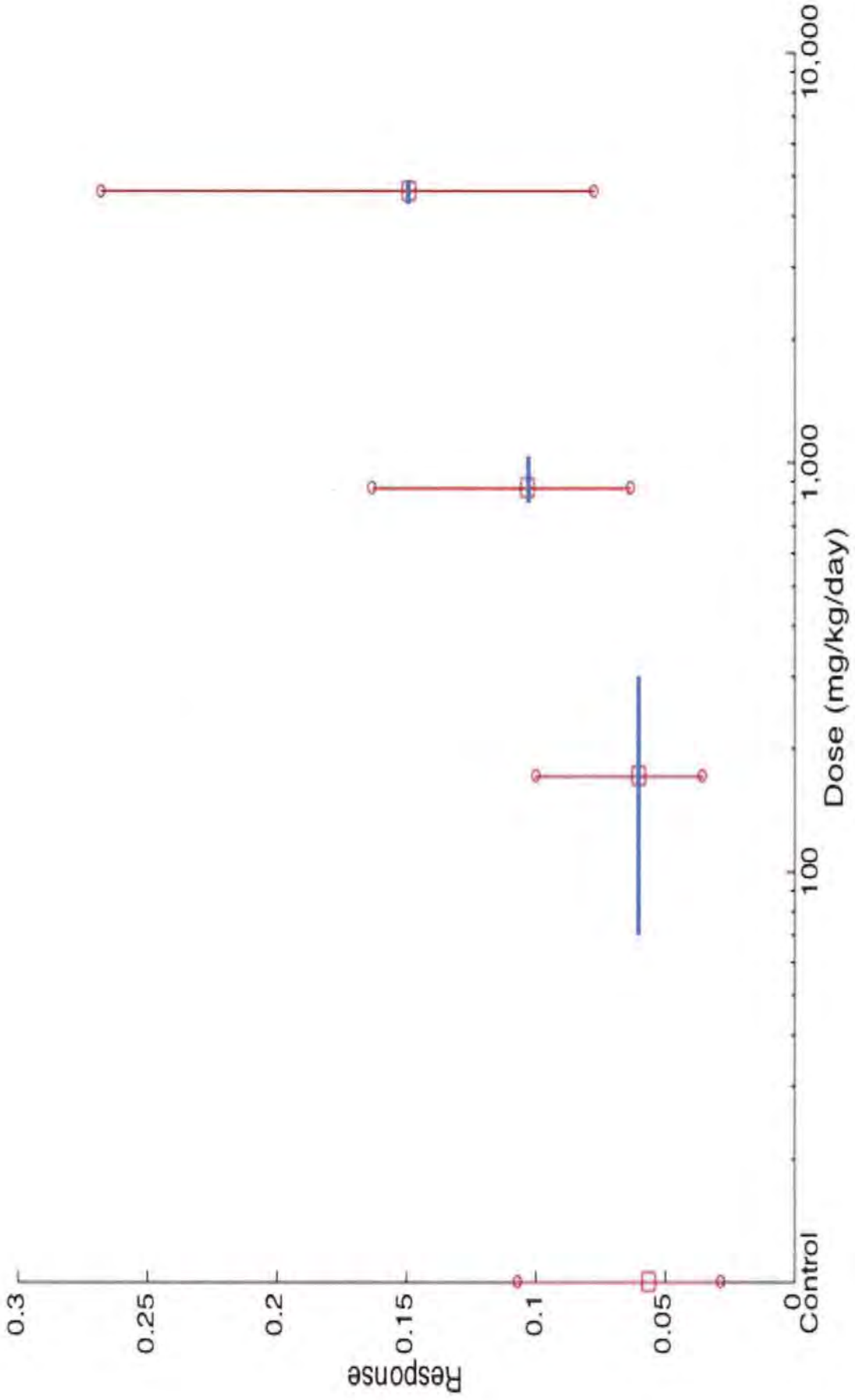


Table 8: Analysis of Male Mouse Hemangiosarcomas From the Individual Studies

Year	Strain	Length	Doses (mg/kg/d)	Response	p-Trend (p-poly3)
1983	Crl:CD-1	24	157, 814, 4841	0/50, 0/49, 1/50, 0/50	0.63 (0.63)
1993	?:CD-1	24	100, 300, 1000	0/50, 0/50, 0/50, 4/50	0.0004 (0.0004)
1997	CrJ:CD-1	18	165, 838, 4348	0/50, 0/50, 0/50, 2/50	0.008 (0.009)
2001	SW	18	15, 151, 1460	No Data	-
2009	Crl:CD-1	18	71, 234, 810	0/51, 0/51, 0/51, 0/51	-

Table 9: Pooled Analysis of Male Mouse Hemangiosarcomas

Year	Strain	p-Trend (p-poly3)
CD-1 Combined	CD-1	0.02 (0.03)
CD-1 Combined and Doses Pooled[1]	CD-1	0.02 (0.02)
CD-1 Combined, doses>1000 dropped	CD-1	<0.0001 (<0.0001)
CD-1 Combined, doses>1000 dropped and Doses Pooled[2]	CD-1	0.0003 (0.0003)

[1] – Doses were combined as follows: all controls, doses between 0 and 310 mg/kg/day, doses between 310 and 1500 mg/kg/day, and doses greater than 1500 mg/kg/day. Average doses in each pooled group were used in the analysis. [2] - Doses were combined as follows: all controls, doses between 0 and 310 mg/kg/day, and doses between 310 and 1500 mg/kg/day. Average doses in each pooled group were used in the analysis.

Figure 7: Hemangiosarcomas in male CD-1 mice poly-3 adjusted showing individual dose groups

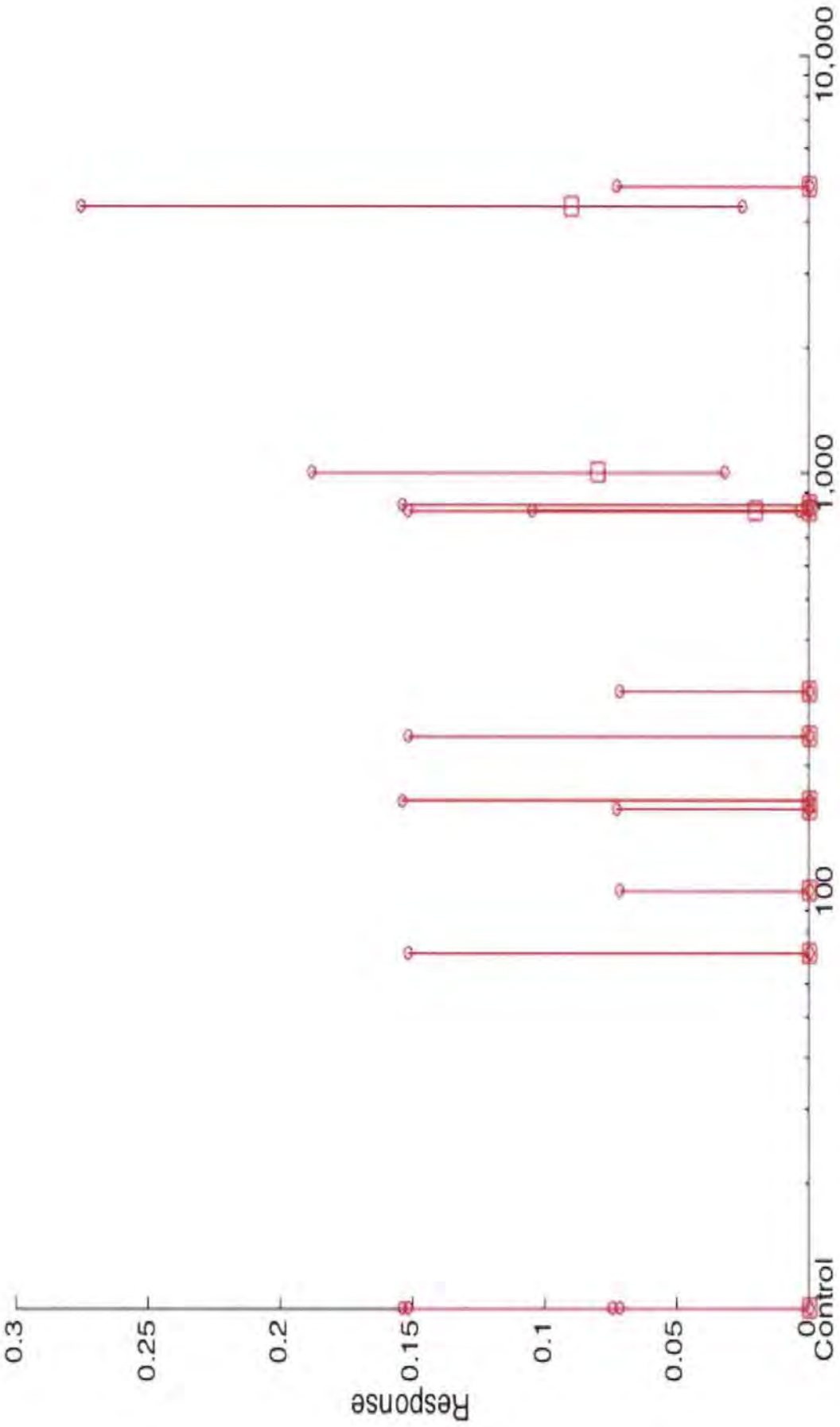
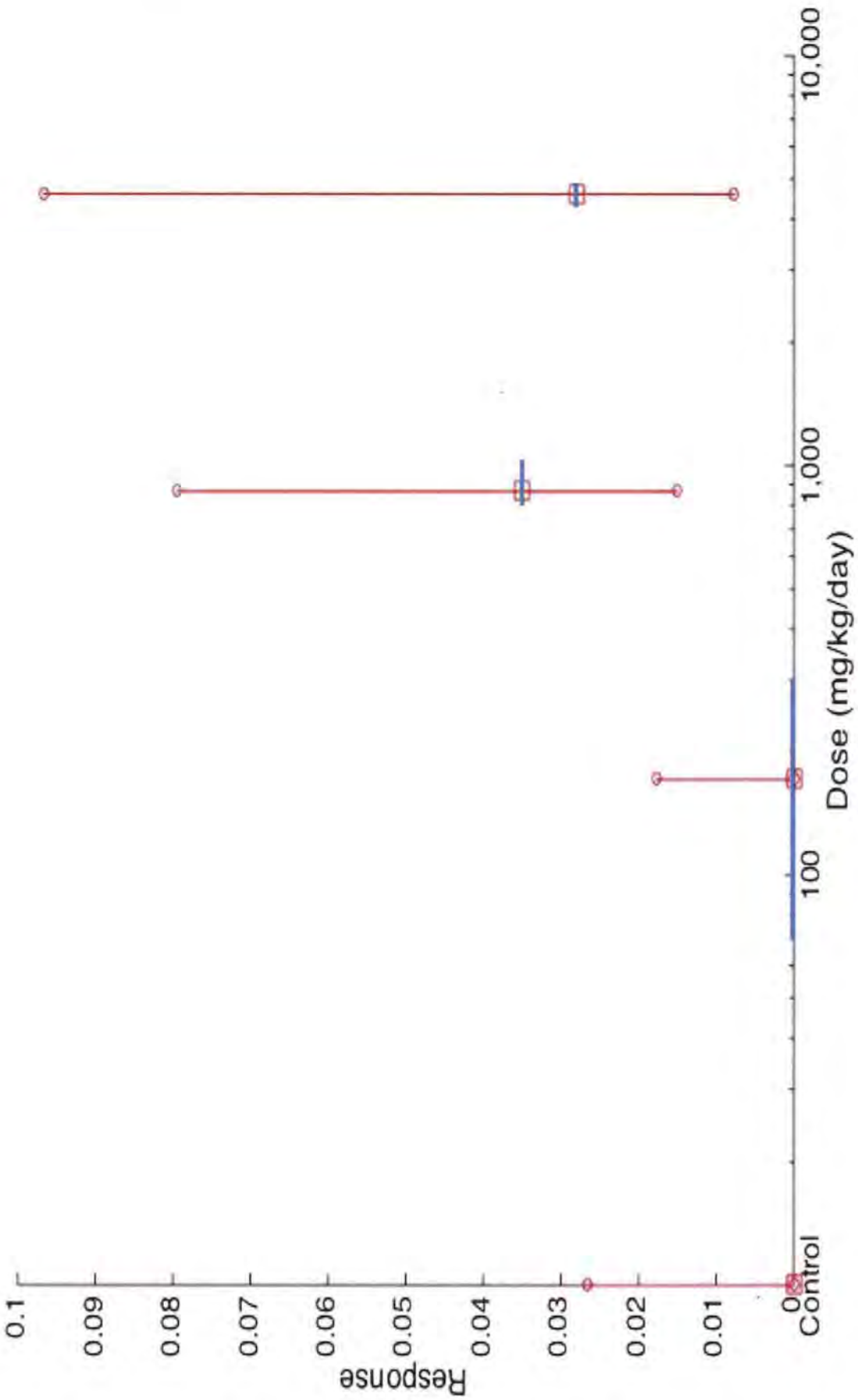


Figure 8: Hemangiosarcomas in male CD-1 mice poly-3 adjusted and clustered by similar doses



Document Four

BAuA
Federal Institute for Occupational Safety and Health
Federal Office for Chemicals
Friedrich-Henkel-Weg 1-25 44149
Dortmund, Germany

RE: CLH Report for Glyphosate, EC Number 213-997-4

Dear Sirs,

Below are my comments on the evaluation of carcinogenicity in the CLH Report for Glyphosate (the Report), EC Number 213-997-4, prepared by the Federal Institute for Occupational Safety and Health (BAuA). In my comments, you will see that I disagree with the conclusions on the human epidemiological data and that I find serious flaws in the evaluation of the animal carcinogenicity data. I have also prepared a pooled analysis of the animal carcinogenicity data that clearly indicates the hemangiosarcomas and malignant lymphomas show statistically significant trends even when excluding doses above 1000 mg/kg/day.

I am also including several supplemental files with this submission including all cited papers, the computer code I used to produce the pooled analysis, and the computer code I used to calculate statistical significance for testing the observed data sets against the historical controls. I have also included a manuscript by Ghisi et al. (2016) that does a meta-analysis on the ability of glyphosate to induce micronuclei.

What I found most disturbing with this submission is that, despite our previous concerns about the EFSA conclusions on carcinogenicity, the review continues to disregard guidance set forth by ECHA, OECD, IARC and others on how to evaluate carcinogenicity data, especially regarding the use of the *limited evidence* category for the human data, the appropriate use of historical controls and the proper use of findings of a positive trend in an animal cancer study.

In my opinion, having reviewed a large number of compounds for carcinogenicity and having read both the Report and the ECHA Guidelines, glyphosate should be classified into Group 1b.

Sincerely,

Prof. Christopher J. Portier
Thun, Switzerland
[redacted]
July 8, 2016

Human Evidence

On page 93 of the Report, the human evidence regarding glyphosate carcinogenicity is summarized as follows:

"Epidemiological studies revealed partly contradictory results. However, in most studies, no association with an exposure to glyphosate could be established. In particular, the largest study, i.e., the AHS (see above), was negative. Taken together, the epidemiological data does not provide convincing evidence that glyphosate exposure in humans might be related to any cancer type. Epidemiological studies are of limited value for detecting the carcinogenic potential of an active substance in plant protection products since humans are never exposed to a single compound alone. Thus, the results of the studies are associated to different formulations containing glyphosate or mixtures of different active substances."

The first sentence claims the results are contradictory. This is only true if classify each study is classified as significant or non-significant. Examining the numerical findings presents a different picture. Table 1 lists the 8 studies (of sufficient quality to be utilized) that evaluated the relationship between non-Hodgkin lymphoma (NHL) and exposure to glyphosate. Simply looking to see if the studies tend to have a relative risk above or below 1 shows the studies to be consistently positive across the board with the exception of the AHS exposure-response analysis (that had problems with classifying the exposure) and the Orsi *et al* study (that had a relative risk of exactly 1). This is quite clearly illustrated using the tree plot in Figure 1.

The sentence '*Taken together, the epidemiological data does not provide convincing evidence that glyphosate exposure in humans might be related to any cancer type.*' is difficult to accept given that the three meta-analyses, all including the AHS study, show a statistically significant association between use of glyphosate pesticides and NHL in humans (Table 2). Finally, the statement that "*the results of the studies are*

associated to different formulations containing glyphosate or mixtures of different active substances." is not supported by actual data so this is speculation and not fact.

In "Guidance on the application of the CLP criteria – Version 4.1", Annex I: 3.6.2.2.3 states that "*The terms 'sufficient' and 'limited' have been used here as they have been defined by the International Agency for Research on Cancer (IARC) and read as follows: ... limited evidence of carcinogenicity: a positive association has been observed between exposure to the agent and cancer for which a causal interpretation is considered to be credible, but chance, bias or confounding could not be ruled out with reasonable confidence.*" The meta-analyses indicate that a positive association has been observed so the only reason you would have for not classifying the human evidence as limited is that you believe the causal relationship is not credible or that the bias and/or confounding is so bad as to make these studies worthless. This is clearly not the case. It is likely that the decision is being skewed by placing too much emphasis on the AHS study; the meta-analysis is designed to avoid this problem.

Finally, this paragraph also implies that human epidemiology data will never be of importance in evaluating a pesticide because the pure compound is not used on humans. Such a statement is not scientifically sound and fails to use the science to address the safety of the public.

Mouse Carcinogenicity Data

Also on page 93 of the Report, the data on the carcinogenicity of glyphosate in mice is summarized.

"*In the mouse, the incidences in malignant lymphoma, in renal tumours and haemangiosarcoma in male animals were considered in detail. Slightly higher incidences when compared with concurrent controls were confined to very high dose levels above the OECD-recommended limit dose of 1000 mg/kg bw/day and exceeding the MTD. In addition, the outcome of statistical tests was contradictory. Mostly, but not always, trend tests revealed statistical significance but pairwise comparisons failed to detect a significant difference relative to the control group. The reported incidences of all three tumour types fell within their historical control range which were, however, of variable reliability. If the four studies in CD-1 mice are considered together, it becomes apparent that all tumours were observed also in the control groups and in some groups receiving lower doses in at least one concurrent study. Furthermore, the results were not consistent with regard to dose responses. To conclude, there is not enough evidence to consider the tumours in mice as treatment-related.*"

It is unusual to have four studies in the same species and strain for an evaluation. It is possible to make direct comparisons between the studies and even pool the data for a combined analysis. Table 3 quickly summarizes the findings from the four studies in CD-1 mice and the one study in Swiss mice. One thing that stands out in Table 3 is that the studies were conducted for either 18 months or 24 months. This is a critical difference that does not get much discussion in the Report.

Cancer increases in risk generally as a power of length of exposure (Portier, Hedges and Hoel, 1986). This relationship was used to develop a means to adjust the length of time an animal is on a study, enabling a scientist to determine risk at the end of two-years, the typical time used for animal bioassays (Bailer and Portier (1988) and Portier and Bailer (1988)). This is called the Poly-3 adjustment. The US National Toxicology Program uses the Poly-3 test to evaluate significance in their animal bioassays. Now you will note that three of the mouse studies were only conducted for 18 months. (Comparing 18 month studies with 24 month studies without making an adjustment for the differences in length of exposure is like comparing cancer rates in 40 year-olds exposed for 25 years to cancer rates in 65 year-olds exposed for 50 years and concluding they are not consistent with each other; the conclusion is meaningless because the correct evaluation was not done.) Thus, in order to compare all 5 studies, we must use the Poly-3 adjustment to extrapolate the 18 month studies to estimate what we think the cancer risk would have looked like at 24 months. The adjustment decreases the number of animals without tumors in all groups by $(18/24)^3$. The p-values for both the unadjusted trend test and the poly-3 adjusted trend test are given in Table 4 for male mouse renal tumors.

As an example of how the Poly-3 adjustments work, consider a comparison of the high-dose renal tumor response in the 1983 study (3/50=6%) to the high-dose response in the 1997 study (2/50=4%). In the 1997 study, 48 animals had no tumors at 18 months; the poly-3 adjustment reduces this to 20.25 leading to an incidence estimate of 2/22.25=9%. Because the Poly3 test effectively reduces the number of animals on study, even though the incidence estimate goes up, the p-value for the trend test could go down. Numerous evaluations of the validity of the poly-3 adjustment have been published in the peer-reviewed literature and it seems to work very well.

Now that the lengths of the studies have been adjusted, the next question to ask is whether this dose-response is consistent across all of the studies or whether there are anomalies. Combining all of the studies into one pooled analysis (Table 5, Line 1) and performing a trend analysis on the pooled data yields highly significant findings (Table 5, Line 1). Excluding the Swiss Albino mouse study (2001) and only using the CD-1 mice also yields a significant trend (Table 5, Line 3). Repeating these analyses with the Poly-3 adjusted data does not alter the significant findings. Poly-3 adjusted dose-response for renal tumors in the entire set of mouse studies is shown in Figure 2. Here, each dose-response point from each study is plotted along with the 95% confidence bound around the response. It is somewhat hard to see that there is a pattern here that is consistent. To make it easier to see, I pooled all the controls into one group, pooled the animals given doses between $0<\text{dose}\leq 300$ in a second group, and similarly for animals given doses between $300<\text{dose}\leq 1500$ and $\text{dose}>1500$. These results are plotted against the mean dose in each set of pooled doses in Figure 3 (the horizontal blue lines show the range of the doses that were combined). The trend in the data is more evident in Figure 3 than in Figure 2. The pooled data sets were also analyzed by the unadjusted and poly-3 adjusted trend

tests and shown to be significant (Table 5, Lines 2 and 4). Finally, as noted in the Report, it seems that all of the response is in doses above 1000 mg/kg/day. After removing all doses above 1000 mg/kg/day and repeating all of the analyses, the results of the analysis are shown in Table 5, Lines 5-8. Without the doses above 1000 mg/kg/day, the effect disappears.

Tables 6 and 7 repeat these analyses for malignant lymphomas and Figures 4, 5, and 6 show the resulting plots of the data. In Figure 4, it is easily seen that the Swiss mice had a very different background tumor rate compared to the CD-1 mice so for the remaining two Figures (5 and 6), only CD-1 mice are plotted. Because of the different backgrounds between the Swiss mice and the CD-1 mice, when they are all combined, the joint analysis is not significant (Table 7, lines 1 and 2). Removing the Swiss mouse study and only evaluating the CD-1 mice leads to highly significant trends in all analyses (Table 7, lines 3-8). A significant trend remains even after removing the doses>1000 (Table 7, lines 5-8) suggesting this is not a high-dose only effect. This is very clear when you examine Figure 7.

Tables 8 and 9 repeat these analyses for hemangiosarcomas and Figures 7 and 8 show the resulting plots of the data. The findings in the Swiss mouse were unclear in the reporting so these tables only contain analyses of the CD-1 mouse data. All analyses are highly significant (Table 9) and they remain significant if doses>1000 are excluded (Table 9, lines 3 and 4). So again, this is not a high dose-only effect.

With these analyses, certain things are clear. The statement "*If the four studies in CD-1 mice are considered together, it becomes apparent that all tumours were observed also in the control groups and in some groups receiving lower doses in at least one concurrent study.*" is highly misleading. Combining all four studies in CD-1 mice leads to very strong statistical significance in the data. Also, "*Furthermore, the results were not consistent with regard to dose responses.*" is also incorrect and not actually supported by the data. Finally, the statement "*Slightly higher incidences when compared with concurrent controls were confined to very high dose levels above the OECD-recommended limit dose of 1000 mg/kg bw/day and exceeding the MTD.*" while partially correct is also very misleading. When doses above 1000 mg/kg/day are excluded, the pooled data from the four CD-1 mouse studies remain significant for both the malignant lymphomas and the hemangiosarcomas. Also, the OECD-recommended limit is not the MTD (maximum tolerated dose) and showing exceedance of an MTD requires more information than simply that the dose was large.

Given a careful, objective evaluation of these data, I strongly suggest you change your conclusion from the mouse studies from "*To conclude, there is not enough evidence to consider the tumours in mice as treatment-related.*" to "***To conclude, there is enough evidence to consider the tumours in mice as treatment-related.***"

Finally, a few comments on the reviews of the individual studies starting on page 67 of the Report.

Page 68 - "*Obviously, the carcinogenicity study in Swiss albino mice by Kumar (2001, ASB2012-11491) revealed an increase in malignant lymphoma incidence over the control at the top dose level of around 1460 mg/kg bw/day in both sexes but the background (control) incidence was also quite high. In fact, at least in males, the number of affected animals in the control groups was markedly higher in this strain than in three studies in CD-1 mice. It must be emphasised that this tumour is quite common in ageing mice and that Swiss mice are frequently affected (for details, see below). In this study, malignant lymphoma accounted for 54.6% of the total number of tumours when all groups are considered together.*" Without actually using historical controls, an attempt is made here to downplay the significance of this finding by saying the concurrent control was high. And then it is not clear at all why the 54.6% figure is put into this paragraph. Is this study positive? Yes. Are there flaws in this study? No. Why does this Report then downplay this finding? Especially when you see similar findings in the other studies?

Page 68 -"*In the most recent study in CD-1 mice by Wood et al. (2009, ASB2012-11490), there was a higher incidence of the same tumour type in high dose males (5/51 vs. 0/51 in the control group). Likewise, in the study by Sugimoto (1997, ASB2012-11493), there were a higher number of male mice affected at the exaggerated dose level of 40000 ppm (approx. 4350 mg/kg bw/day) than in the control group (6/50 vs. 2/50). In the study by Atkinson et al. (1993, TOX9552382), in contrast, there was no dose response and the incidence in the control group was similar to that at the top dose level.*" Regardless, this entire paragraph is attempting to compare control animals ranging over 16 years with differing terminal sacrifice times and from different laboratories. Such a comparison is inappropriate because of the known drift in strains over time and increasing tumor risk with age. The OECD guidelines make this very clear.

Page 69 – "*The trend test also provided a p-value above the significance level of 0.05, most probably because of the high control incidence (see Table 33).*" The p-value for trend is 0.0535122, technically above 0.05, but it is misleading when trying to compare across studies not to mention that this is almost significant.

Page 69 - "*In contrast, re-analysis of the studies by Wood et al. (2009, ASB2012-11490) and Sugimoto (1997, ASB2012-11493) showed statistically significant increases with dose for male CD-1 mice in the trend test (Table 34 and Table 35) but a rather low or even "zero" incidence in the control groups might be behind this finding.*" Where are the historical controls to support the speculation in the last part of this sentence? And of course the formal statistical analysis to go with it. Finally, as noted in the Report, OECD guidelines, IARC guidelines, NTP guidelines and others, the concurrent control is the best to use for evaluating a study.

Page 69 – "*This result was confirmed by the chi-square test. Also for this comparison, the very low control incidence (0/51) should be taken into consideration.*" Again, where are the historical controls to support this statement?

Page 71 – "*It may be concluded that the statistical significance of the suspected increase in malignant lymphoma in the various studies depends very much on the statistical method that is used for data analysis.*" This is usually the case; that is why the OECD guidelines make it clear that if either the trend test or the pairwise comparison is positive, the findings should be considered positive.

Page 71 – "*When the trend test is applied, the studies by Wood et al. (2009, ASB2012-11490) and Sugimoto (1997, ASB2012-11493) provide evidence of an effect which was not the case when pairwise comparison was performed. In contrast, the increase in the study of Kumar (2001, ASB2012-11491) was not confirmed neither by the trend test nor by a different pairwise test than the Z-test that had been used first.*" From my Table 6, there are two significantly positive studies, two studies with a marginal p-value and one study that would be positive if not for the highest dose dropping down. As noted in the Report, there was a drop in weight gain in the 1993 which could explain the drop in tumors at the highest exposure group (animals with reduced caloric intake are less likely to get tumors).

Page 71 – "*In the studies by Wood et al. (2009, ASB2012-11490) and by Atkinson et al. (1993, TOX9552382) in CD-1 mice, comparable top doses of 810 or 1000 mg/kg bw/day were administered and a similar incidence of malignant lymphoma was noted in high dose males (5/51 or 6/50, respectively). However, the control group incidences were clearly different (0/51 vs. 4/50) resulting in a positive trend test in the study by Wood et al. (2009, ASB2012-11490) only.*" The 1993 study was 24 months whereas the 2009 study was 18 months; it is not surprising the control tumor counts are higher in the 1993 study. What is surprising (and statistically significant) are the 6 tumors at the high dose in the 2009 study after only 18 months. And of course, this is another inappropriate comparison of control incidence over a 16 year timeframe. And finally, none of this is statistically significant.

Page 71 – "*Thus, if all four studies in CD-1 mice are taken together, there is no consistent dose response.*" See my formal analysis of this question.

Page 71 – "*Nonetheless, it seems well in line with information that was found in the literature providing confirmation that Swiss mice are prone to developing lymphoreticular tumours. According to older articles, control incidences in male mice of Swiss or Swiss-derived strains may reach 18–27.5% and exceed 36% in females (Sher, 1974, Z22020; Roe and Tucker, 1974, ASB2015-2534; Tucker, 1979, Z83266). In a more recent publication, Tadesse-Heath et al. (2000, ASB2015-2535) even mentioned a nearly 50% lymphoma (mostly of B cell origin) incidence in a colony of CFW Swiss mice but also emphasised the contribution of widespread infections with murine oncogenic viruses to the high but remarkably variable incidence of tumours of the lymphoreticular system in this species.*" Why are there guidelines if they are not used? Again, an argument is being made about historical controls using data which does not match OECD guidance (even bringing in Swiss-derived strains). And, if there are had historical control values from the lab, giving all five numbers and

some description of the studies (18 months or 24 months?) would seem to be in order.

Page 72 – "*However, in the study report itself, there was no evidence of health deterioration due to suspected viral infection and, thus, the actual basis of EPA's decision is not known.*" The entire discussion about infections is, at best, absurd if there is no evidence. Inclusion of this text is simply an attempt to discredit the study.

Page 72 – "*It ranged from 3.85% to 19.23% in the control groups from 12 studies that had been performed between 1992 and 1998 (Kitazawa, 2013, ASB2014-9146). Thus, the 12% incidence at the top dose level in the study with glyphosate was well covered by the range even though it was above the mean value of 6.33%.*" 12 studies with a mean of 6.33% and a range of 3.85 to 19.23 is an extremely skewed population. One study had 3.85% and one had 19.23 %; 12 x 6.33%=75.96 so the remaining 10 studies, in order to get an average of 6.33% would need to add up to 52.54 or 5.25% per study on average. Just from the math, it appears the 19.23% control is an outlier. Regardless, for sake of transparency, the actual rates should be given and assurances be given that they are all from studies of 18 months and not 24 months. And finally, a formal statistical analysis against the historical controls should be conducted. To illustrate; if the historical background is 6.33% and is based upon 50 animals in each control group and the controls are binomially distributed, then the probability of randomly seeing an outcome with a trend statistic equal to or larger than the one observed in this study is p=0.02. MATLAB code is provided that makes this calculation.

Page 72 – "*Unfortunately, for the study of Wood et al. (2009, ASB2012-11492), the submitted historical control data was not particularly useful for the assessment.*" Stop with this statement; everything else written is an inappropriate use of historical control data and should be ignored.

Page 73 – "*On balance, based on uncertainties with regard to partly contradictory study outcomes depending on the statistical method applied, inconsistent dose response in the individual studies, and a highly variable tumour incidence as suggested by historical control data, it is not likely that glyphosate has induced malignant lymphoma in mice. A possible role of oncogenic viruses should not be ignored. Moreover, human relevance of such an effect, if occurring only as a high-dose phenomenon as it was the case here, is considered equivocal.*" On balance, this entire paragraph is a wrong. The study outcomes are not contradictory (follow OECD guidance and it is simple), does response is not inconsistent (see my analysis), tumor incidence is not highly variable when properly adjusted for time on study differences and the entire historical control discussion is either inappropriate or inadequately applied.

Page 74 -"*Even though no historical control data from the performing laboratories was provided, a simple comparison of the control groups in the individual studies with*

glyphosate suggests that renal tumours may occur in untreated control males at a similar incidence than in the groups receiving very high doses." This is a misleading comment. First, no formal analysis of historical control data has been undertaken and, as we stated in our paper (Portier *et al.*, 2016), your own guidelines provide guidance on how to obtain and use historical control data; this has not been done here. I am also surprised to see the statement that "*no historical control data from the performing laboratories was provided*" when in response to a letter sent to Commissioner Andriukaitis, the EFSA Executive Director, Professor Url, wrote "*The Peer Review Report (EFSA, 2015b) confirms that EFSA conducted a specific check regarding the use of historical control data, requested additional information during the clock-stop procedure and only considered valid the historical control data from the performing laboratory in line with the international recommendations*". Which is it? Does the Report rely on valid historical control data from the performing laboratories or not?

Page 75 - "*Even if not fully comparable because of the strain differences, it should be remembered that the top dose incidence of 2/50 in this study was the same as seen in CD-1 mice in the study by Atkinson et al. (1993, TOX9552382) in the control and low dose groups.*" Why even include this sentence? They are not comparable.

Page 76 – "*Even though there was no clear dose response, it may be assumed that glyphosate (acid) when administered at high doses might produce mucosal irritation.*" So, if I am reading this right, statistically significant positive cancer results are being dismissed based on non-statistically significant non-cancer results that have a questionable linkage to the cancer results. Does this seem reasonable? I guess not since this appears in the next papagraph "*However, it is questionable if irritation would sufficiently explain tumour formation in the kidney.*".

Page 76 – "*The top dose finding of 2/50 in the study by Sugimoto (1997, ASB2012-11493) is at the upper edge of adenoma frequency. In the study by Knezevich and Hogan (1983, TOX9552381) which is not actually covered by the timeframe of the historical database, the adenoma incidence (2%) at the top dose level would be inside the historical range whereas a carcinoma incidence of 4% was above.*" Again, an improper use of historical controls. These are not appropriate for the 1983 study but are used anyway. For the 1997 study, only controls in mice sacrificed at 18 months should be used, mice sacrificed at 24 months will likely have greater incidence. This is quite evident when one looks at hemangiosarcomas in male mice in the Giknis and Clifford report (attached). Exactly half of the studies went 18 months, 24 went 2 years and the remaining two went 97 and 100 weeks. Hence this historical control dataset is inappropriate for this comparison. However, even if it were, the findings would still be significant. The paper gives a mean background level for adenomas of 0.24% and for adenocarcinoma of 0.14% for a combined background of 0.38%. The probability of seeing a dose-response trend equal to or larger than what was seen in the 1997 study is 0.01, a significant finding. The p-value for the 1983 study would be even smaller.

Page 77 – "*Even the incidences of affected animals at exaggerated doses exceeding the OECD-recommended limit of 1000 mg/kg bw/day and also the MTD were not statistically significantly increased when compared with the concurrent controls.*" As mentioned earlier in this document, if either test is positive, the findings should be considered positive so the second half of this sentence is inappropriate. How did "*there is some evidence that the MTD was exceeded in both studies at the highest dose level*" (Page 76) become absolute certainty about exceeding the MTD?

Page 77 – "*Even the incidences at exaggerated doses are covered by the historical control range.*" As noted earlier, this finding is not supported.

Page 77 – "*No pre-neoplastic kidney lesions have been observed in treated animals.*" Following this logic, the high dose animals got tumors by some unknown mechanism related to exceeding the MTD and that unknown mechanism did not damage the kidneys in any other animals enough to show preneoplastic effects. What is this mechanism and where is the evidence suggesting such a mechanism exists? And how does this statement "*However, it is questionable if irritation would sufficiently explain tumour formation in the kidney.*" fit in to this theory?

Page 77 – "*There is no plausible mechanism*" Following the logic again, some unknown mechanism related to exceeding the MTD caused the tumors at the highest doses and because there is no mechanism, the results should be dismissed.

Page 78 – "*According to Atkinson et al. (1993, TOX9552382), the historical control incidence in the performing laboratory ranged from 0/50 to 4/50 and, thus, would cover the incidence at the top dose level.*" Inadequate documentation of the historical control data makes it impossible to address this statement. The actual counts and ages at terminal sacrifice for the historical controls should be provided. As shown earlier, range is an inappropriate way to utilize historical controls. This is a clear example of a lack of transparency.

Page 78 – "*Historical control data provided by Charles River indicate a very variable incidence of haemangiosarcoma. On different sites of the body, tumours of this type were seen in untreated control animals in 8 of 52 studies.*" In this case, Giknis and Clifford give the actual values for each of their control groups. For hemangiosarcomas, there were zero tumors in all 26 studies terminated at 18 months, and only 8 of the remaining 26 studies that went two years had hemangiosarcomas. Thus, the 18 month 1997 study is well outside the range of the historical controls.

Page 78 – "*Furthermore, since Sugimoto (1997, ASB2012-11493) employed a more than four times higher top dose than Atkinson et al. (1993, TOX9552382), a markedly higher haemangiosarcoma incidence would have been expected if this tumour was in fact treatment-related.*" Again, this is a comparison of an 18 month study to a 24 month study. The finding that the 24 month 1993 study has an 8% response at a dose of 1000 mg/kg/day while the 18 month 1997 study has a 4% response at a 4-

fold higher dose is not unexpected.

Document Five

Table 1: Human Epidemiology Studies

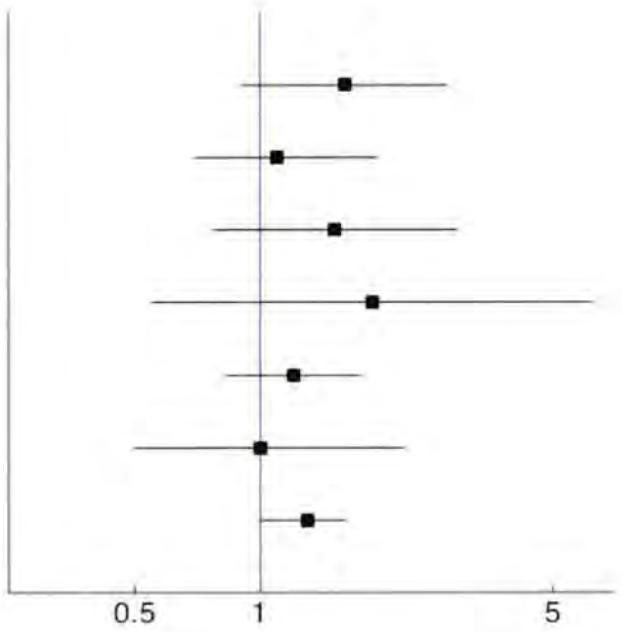
Study	Type	Size	Findings	Exposed Cases
Agricultural Health Study *(De Rooset al., 2005)*	Cohort – licensed pesticide applicators	52 395 (+32 347 spouses), 92 cases, 4-8 years follow-up	1.1 (0.7-1.9) C 0.7 (0.4-1.4) 21-56% tertile compared to <20% tertile 0.9 (0.5-1.6) 21-56% tertile compared to >57% tertile (31 cases no quantification of exposure)	73
US Midwest *(De Roos et al., 2003)*	Pooled analysis 3 case-control studies	NHL: 650 cases, 1933 controls	2.1 (1.1-4) U 1.6 (0.9-2.8) C	36 36
Cross-Canada *(McDuffie et al., 2001)*	Population-based case-control study	517 cases, 1506 controls	1.2 (0.83-1.74) U 1.0 (0.63-1.57) ≤2 d/Y 2.12 (1.2-3.73) >2 d/Y	51 28 23
Swedish Case-Control Study *(Eriksson et al., 2008)*	Population-based case-control study	910 cases, 1016 control	2.02 (1.1-3.71) U 1.51 (0.77-2.94) C 1.69 (0.7-4.07) ≤10 d/Y 2.36 (1.04-5.37) >10 d/Y 1.11 (0.24-5.08) ≤10 Y 2.26 (1.16-4.4) >10 Y	29 29 12 17 NR NR
Swedish Case-Control Study *(Hardell et al., 1999)*	Population-based case-control study	404 cases, 741 control (limited power)	2.3 (0.4-1.3) U 5.8 (0.6-5.4) C (not specified)	4 NR
France Case-Control *(Orsi et al, 2009)*	Hospital-based case-control study	244 cases, 456 controls	1.0 (0.5-2.2) U	12
Swedish Case-Control Study *(Hardell et al., 2002)*	Population-based case-control study	515 cases, 1141 controls	3.04 (1.08-8.5) U 1.85 (0.55-6.2) C (notspecified)	8 8
US Case-Control Study *(Lee et al., 2004)*	Population-based case-control study	872 cases, 2381controls	1.4 (0.98-2.1) U – no asthma 1.2 (0.4-3.3) U - asthma	53 6

Table 2: Meta Analyses

Study	Included Studies	Findings
Schinasi and Leon, 2014	McDuffie et al., 2001; Hardell et al., 2002; De Roos et al., 2003 and 2005; Eriksson et al., 2008; Orsi et al., 2009)	1.5 (1.1-2.0)
IARC Monograph Working Group	McDuffie et al., 2001; Hardell et al., 2002; De Roos et al., 2003 and 2005; Eriksson et al., 2008; Orsi et al., 2009)	1.3 (1.103-1.65) – used adjusted risk estimates from Hardell et al., 2003 and Eriksson et al., 2008
Chang and Delzell, 2016	McDuffie et al., 2001; Hardell et al., 2002; De Roos et al., 2003 and 2005; Eriksson et al., 2008; Orsi et al., 2009)	1.3 (1.0-1.6)

Figure 1: Tree Plot of Epidemiology Studies

Study	RR	Lower	Upper	Weight
De Roos et al. (2003)	1.600	0.900	2.800	16.2
De Roos et al. (2003)	1.100	0.700	1.900	21.0
Eriksson et al., (2008)	1.510	0.770	2.940	11.6
Hardell et al. (2002)	1.850	0.550	6.200	3.6
McDuffie et al. (2001)	1.200	0.830	1.740	38.1
Oris et al. (2009)	1.000	0.500	2.200	9.5
Meta-Analysis	1.300	1.000	1.600	100.0



Chang and Delzell (2016)

Table 3: Carcinogenicity Studies in Male Mice

Year	Strain	Length[1]	Top Dose[2]	Renal Tumors	Hemangio-sarcomas	Malignant Lymphoma
1983[5]	Crl:CD-1	24	4,841	+[3]		
1993[5]	?:CD-1	24	1,000		+	+/-[4]
1997	CrJ:CD-1	18	4,843	+	+	+
2001	SW	18	1,460	+	**Data Not Available**	+
2009	Crl:CD-1	18	810			+

1 – months; 2 – mg/kg bw/day; 3 - + indicates a p-value of <0.05 as calculated by BfR using the Armitage linear trend test in proportions; 4 – p=0.054; 5 – studies evaluated in IARC review; p=0.08

Table 4: Analysis of Male Mouse Renal Tumors From the Individual Studies

Year	Strain	Length	Doses (mg/kg/d)	Response	p-Trend (p-poly3)
1983	Crl:CD-1	24	157, 814, 4841	1/50, 0/49, 1/50, 3/50	0.03 (0.03)
1993	?:CD-1	24	100, 300, 1000	2/50, 2/50, 0/50, 0/50	0.94 (0.94)
1997	CrJ:CD-1	18	165, 838, 4348	0/50, 0/50, 0/50, 2/50	0.008 (0.009)
2001	SW	18	15, 151, 1460	0/49, 0/49, 1/50, 2/50	0.04 (0.04)
2009	Crl:CD-1	18	71, 234, 810	0/51, 0/51, 0/51, 0/51	-

Table 5: Pooled Analysis of Male Mouse Renal Tumors

Year	Strain	p-Trend (p-poly3)
All Combined	CD-1 and Swiss	0.0004 (0.001)
All Combined and Doses Pooled[1]	CD-1 and Swiss	0.0008 (0.002)
CD-1 Combined	CD-1	0.001 (0.001)
CD-1 Combined and Doses Pooled[1]	CD-1	0.001(0.001)
All Combined, doses>1000 dropped	CD-1 and Swiss	0.80 (0.84)
All Combined, doses>1000 dropped and Doses Pooled[2]	CD-1 and Swiss	0.39 (0.40)
CD-1 Combined, doses>1000 dropped	CD-1	0.85 (0.86)
CD-1 Combined, doses>1000 dropped and Doses Pooled[2]	CD-1	0.80 (0.80)

[1] – Doses were combined as follows: all controls, doses between 0 and 310 mg/kg/day, doses between 310 and 1500 mg/kg/day, and doses greater than 1500 mg/kg/day. Average doses in each pooled group were used in the analysis. [2] - Doses were combined as follows: all controls, doses between 0 and 310 mg/kg/day, and doses between 310 and 1500 mg/kg/day. Average doses in each pooled group were used in the analysis.

Figure 2: Renal tumors in male mice poly-3 adjusted showing individual dose groups

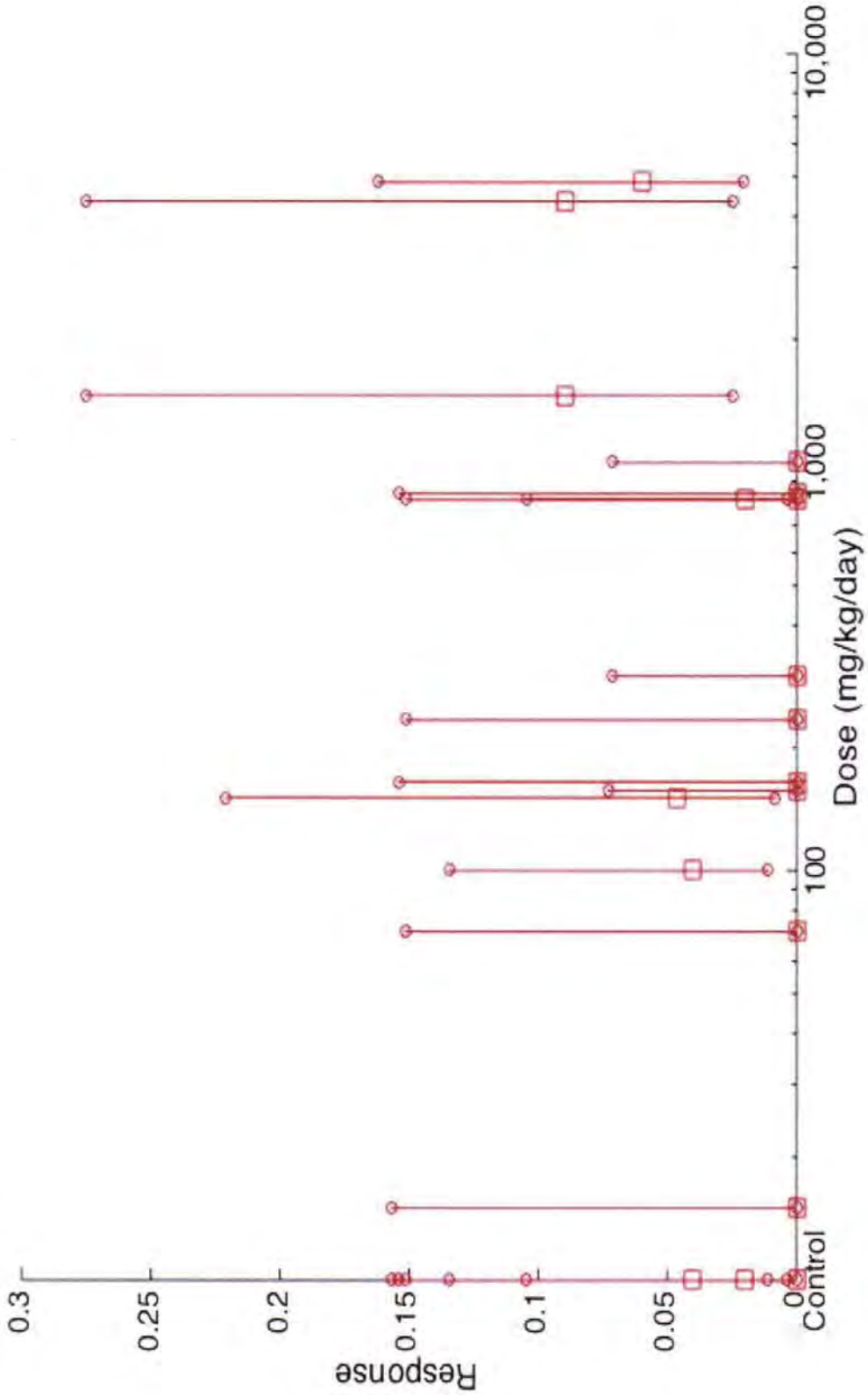


Figure 3: Renal tumors in male mice poly-3 adjusted and clustered by similar doses

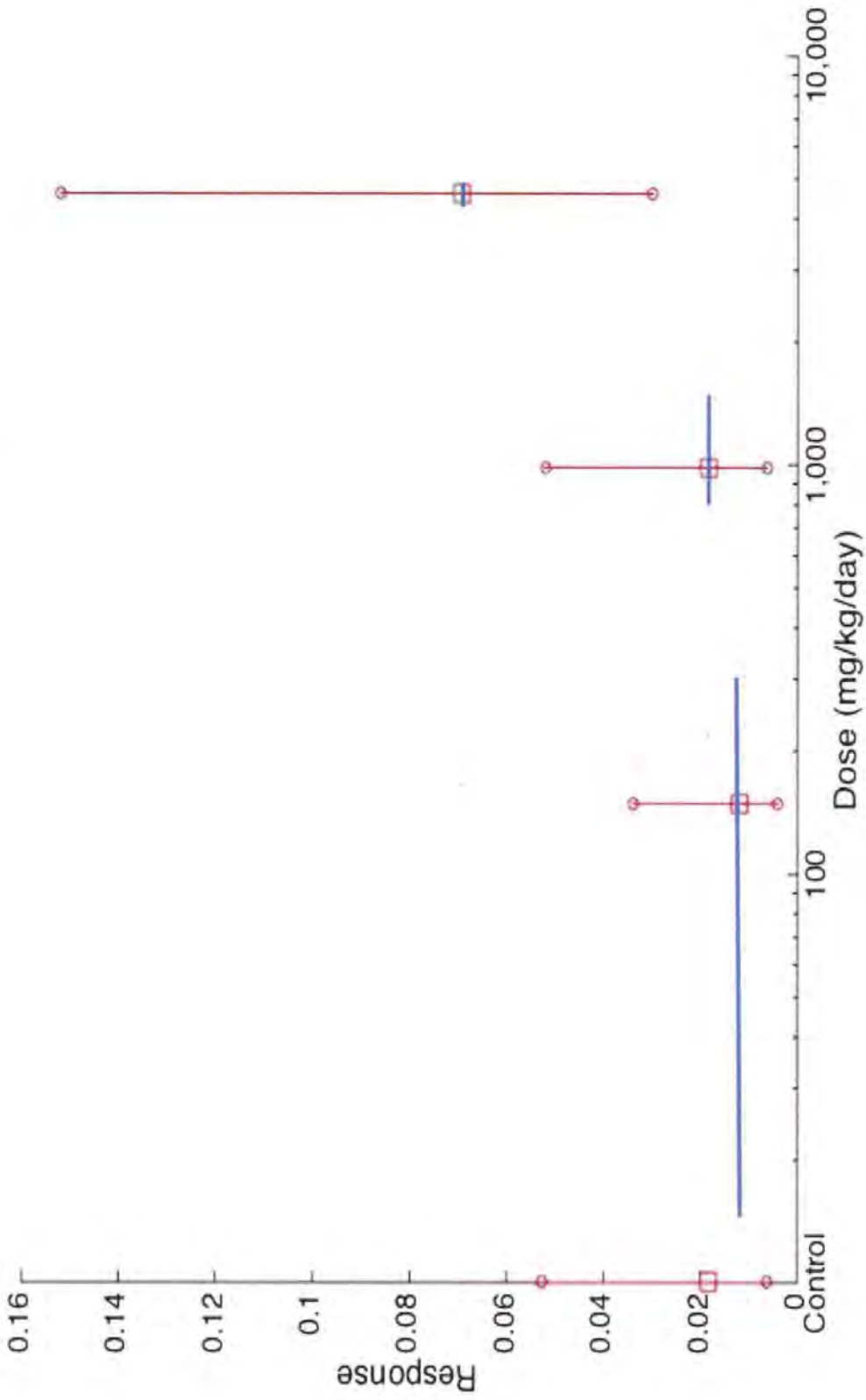


Table 6: Analysis of Male Mouse Malignant Lymphoma From the Individual Studies

Year	Strain	Length	Doses (mg/kg/d)	Response	p-Trend (p-poly3)
1983	Crl:CD-1	24	157, 814, 4841	2/50, 5/49, 4/50, 2/50	0.51 (0.51)
1993	?:CD-1	24	100, 300, 1000	4/50, 2/50, 1/50, 6/50	0.08 (0.08)
1997	CrJ:CD-1	18	165, 838, 4348	2/50, 2/50, 0/50, 6/50	0.008 (0.012)
2001	SW	18	15, 151, 1460	10/49, 15/49, 16/49, 19/49	0.05 (0.09)
2009	Crl:CD-1	18	71, 234, 810	0/51, 1/51, 2/51, 5/51	0.004 (0.005)

Table 7: Pooled Analysis of Male Mouse Malignant Lymphoma

Year	Strain	p-Trend (p-poly3)
All Combined	CD-1 and Swiss	0.17 (0.19)
All Combined and Doses Pooled[1]	CD-1 and Swiss	0.32 (0.31)
CD-1 Combined	CD-1	0.02 (0.01)
CD-1 Combined and Doses Pooled[1]	CD-1	0.01(0.009)
All Combined, doses>1000 dropped	CD-1 and Swiss	0.86 (0.93)
All Combined, doses>1000 dropped and Doses Pooled[2]	CD-1 and Swiss	0.02 (0.03)
CD-1 Combined, doses>1000 dropped	CD-1	0.03 (0.05)
CD-1 Combined, doses>1000 dropped and Doses Pooled[2]	CD-1	0.04 (0.04)

[1] – Doses were combined as follows: all controls, doses between 0 and 310 mg/kg/day, doses between 310 and 1500 mg/kg/day, and doses greater than 1500 mg/kg/day. Average doses in each pooled group were used in the analysis. [2] – Doses were combined as follows: all controls, doses between 0 and 310 mg/kg/day, and doses between 310 and 1500 mg/kg/day. Average doses in each pooled group were used in the analysis.

Figure 4: Malignant lymphomas in male mice poly-3 adjusted showing individual dose groups

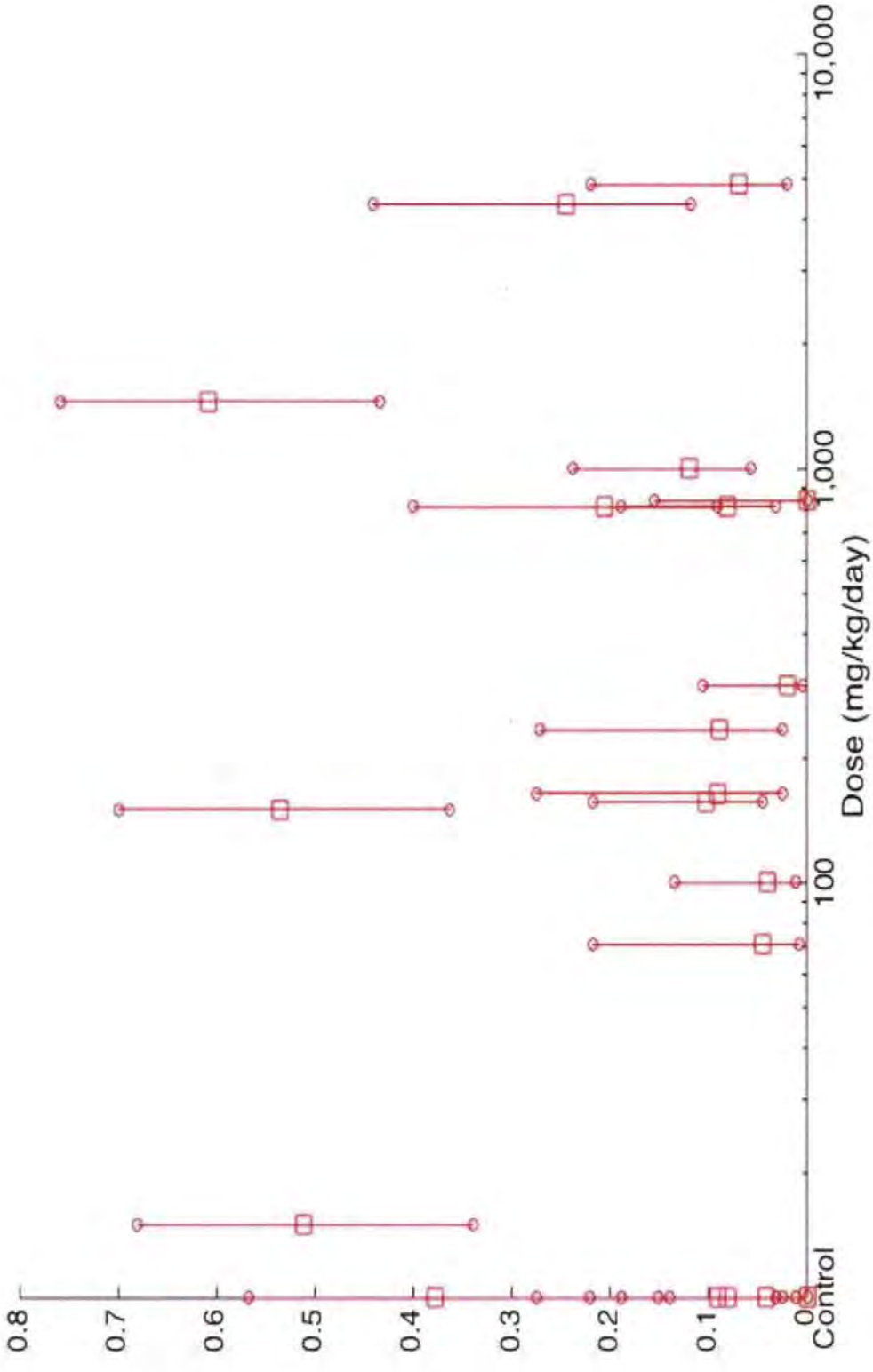


Figure 5: Malignant lymphomas in male CD-1 mice poly-3 adjusted showing individual dose groups

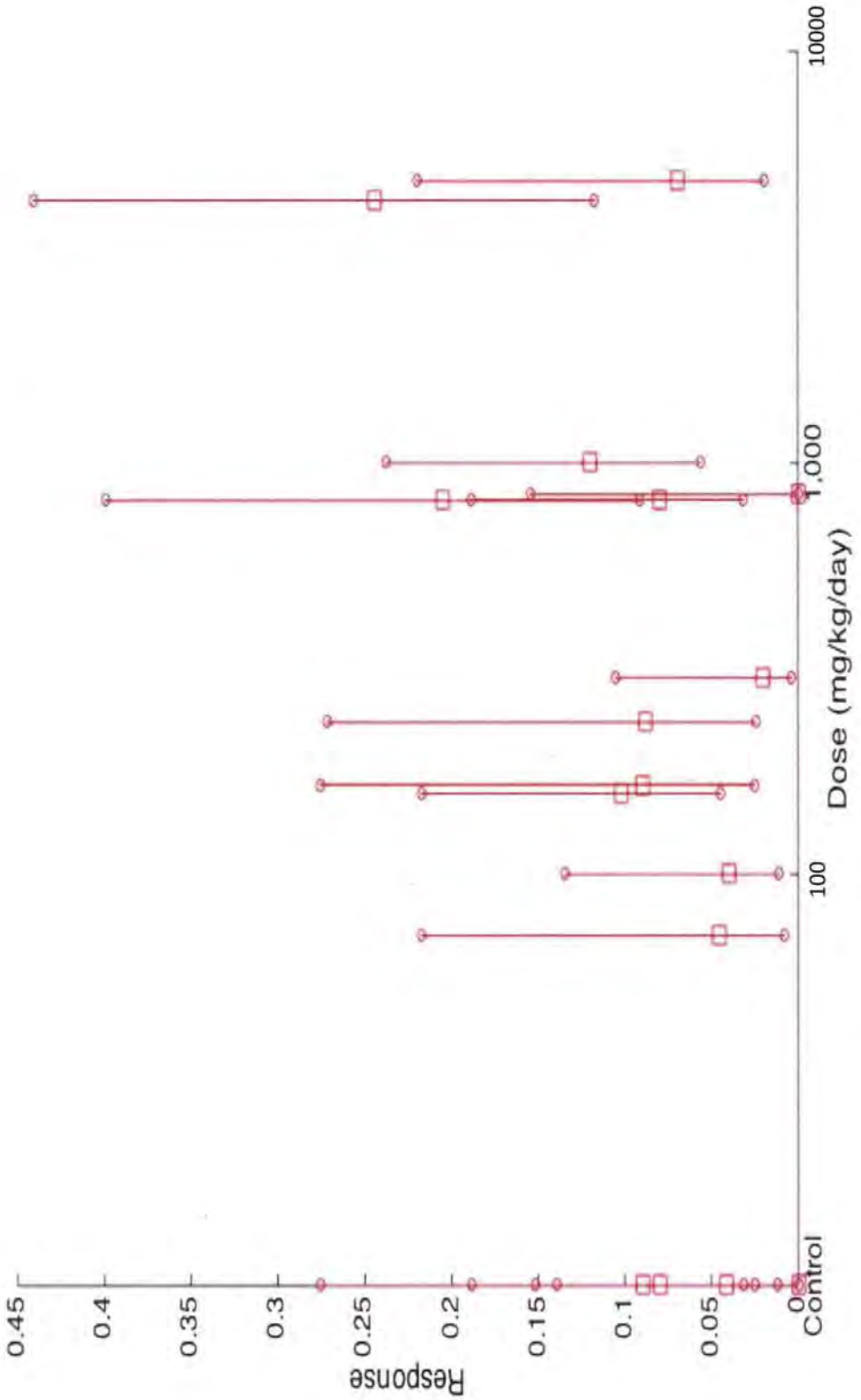


Figure 6: Malignant lymphomas in male CD-1mice poly-3 adjusted and clustered by similar doses

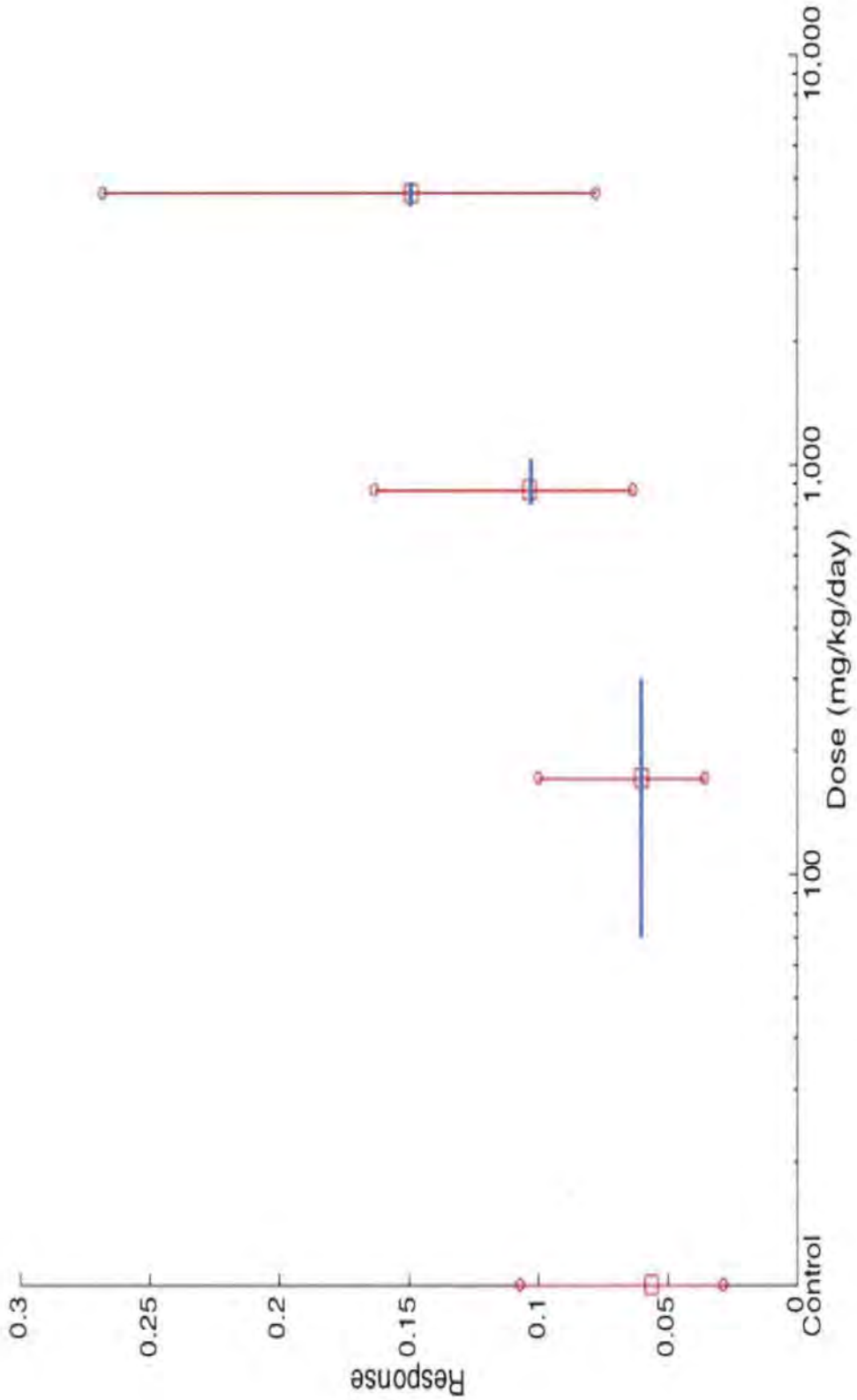


Table 8: Analysis of Male Mouse Hemangiosarcomas From the Individual Studies

Year	Strain	Length	Doses (mg/kg/d)	Response	p-Trend (p-poly3)
1983	Crl:CD-1	24	157, 814, 4841	0/50, 0/49, 1/50, 0/50	0.63 (0.63)
1993	?:CD-1	24	100, 300, 1000	0/50, 0/50, 0/50, 4/50	0.0004 (0.0004)
1997	CrJ:CD-1	18	165, 838, 4348	0/50, 0/50, 0/50, 2/50	0.008 (0.009)
2001	SW	18	15, 151, 1460	No Data	-
2009	Crl:CD-1	18	71, 234, 810	0/51, 0/51, 0/51, 0/51	-

Table 9: Pooled Analysis of Male Mouse Hemangiosarcomas

Year	Strain	p-Trend (p-poly3)
CD-1 Combined	CD-1	0.02 (0.03)
CD-1 Combined and Doses Pooled[1]	CD-1	0.02 (0.02)
CD-1 Combined, doses>1000 dropped	CD-1	<0.0001 (<0.0001)
CD-1 Combined, doses>1000 dropped and Doses Pooled[2]	CD-1	0.0003 (0.0003)

[1] – Doses were combined as follows: all controls, doses between 0 and 310 mg/kg/day, doses between 310 and 1500 mg/kg/day, and doses greater than 1500 mg/kg/day. Average doses in each pooled group were used in the analysis. [2] - Doses were combined as follows: all controls, doses between 0 and 310 mg/kg/day, and doses between 310 and 1500 mg/kg/day. Average doses in each pooled group were used in the analysis.

Figure 7: Hemangiosarcomas in male CD-1 mice poly-3 adjusted showing individual dose groups

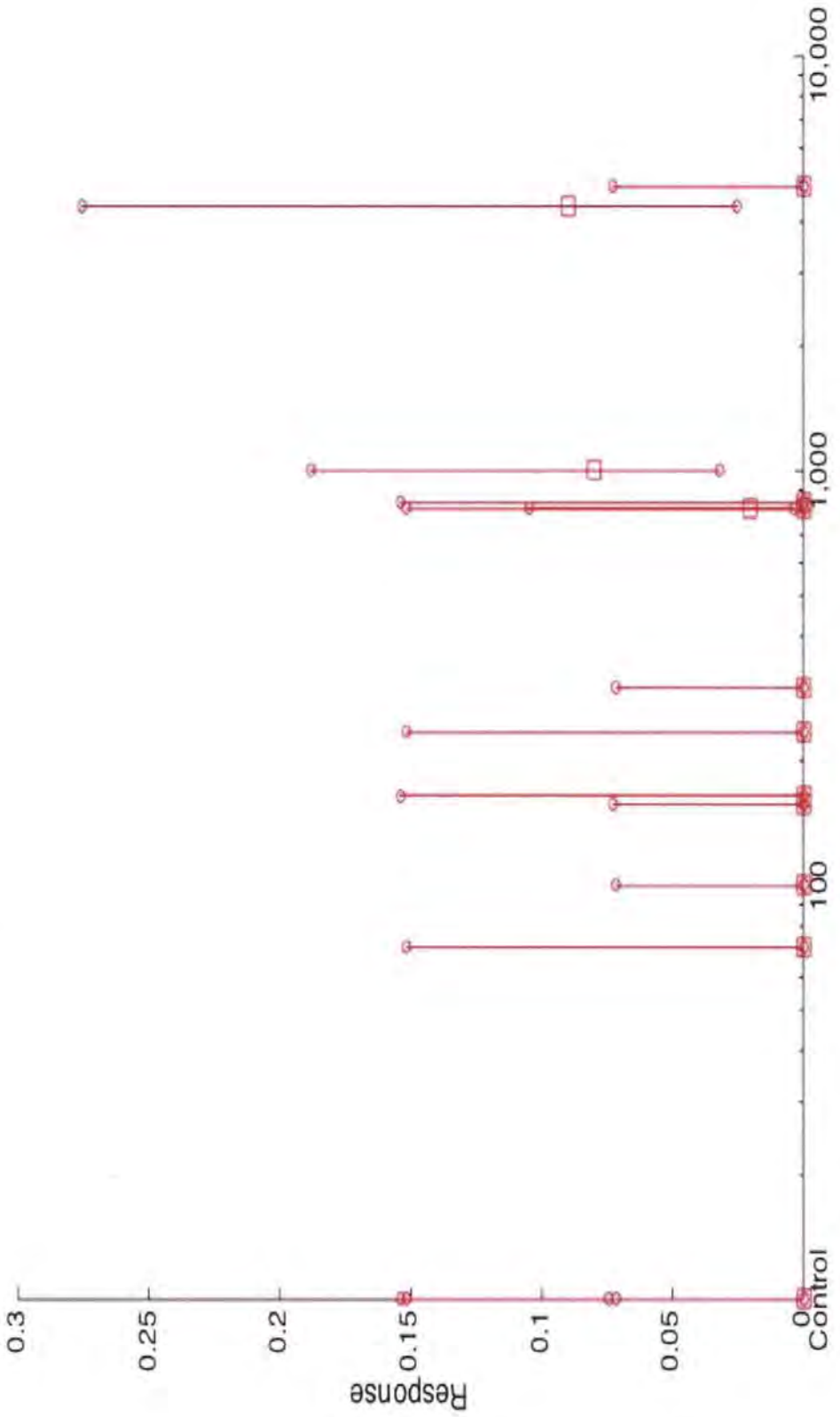
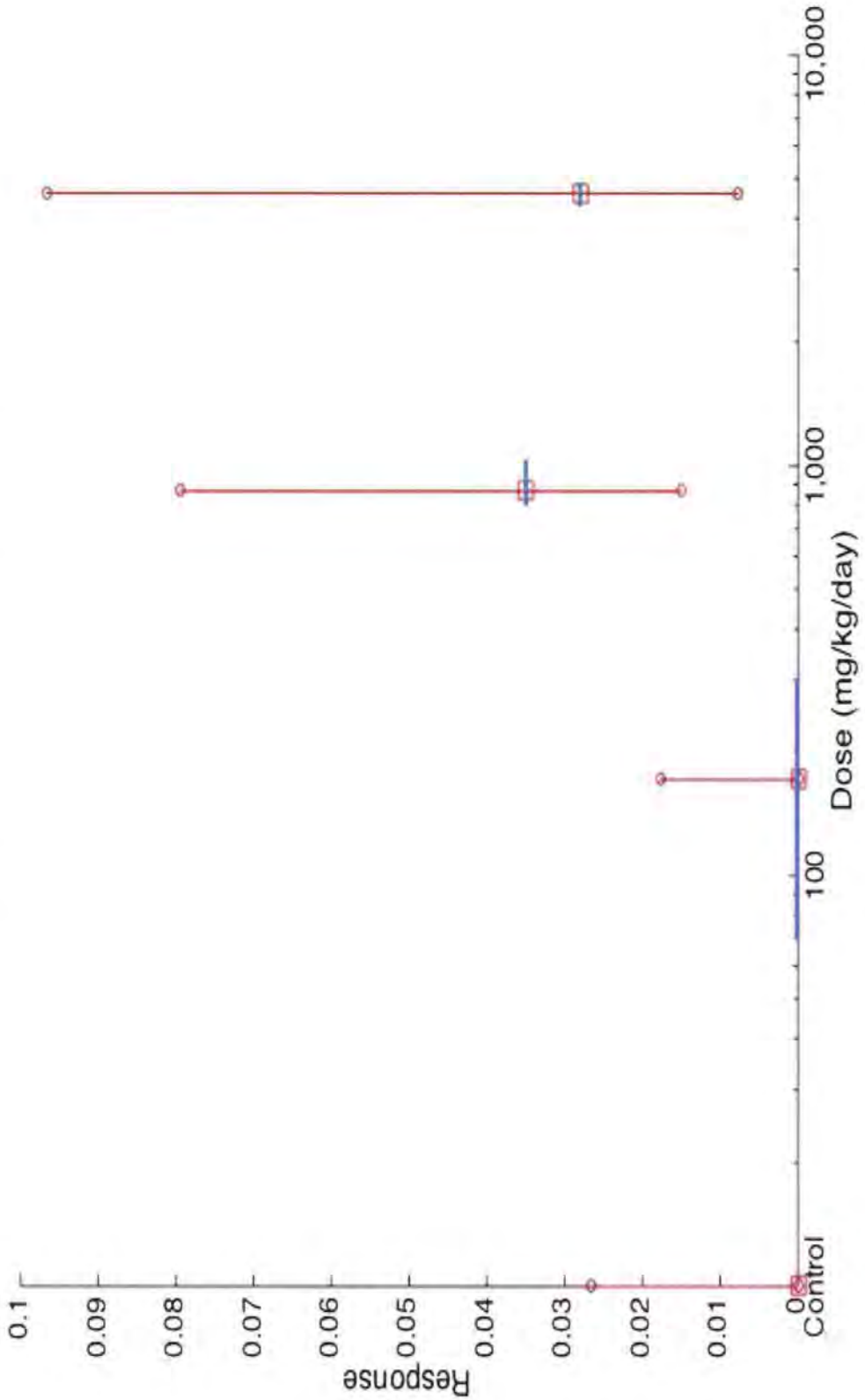


Figure 8: Hemangiosarcomas in male CD-1 mice poly-3 adjusted and clustered by similar doses



Document Six

Response to comments prepared by Christopher Portier (dated October 4, 2016) for the glyphosate EPA SAP meeting

Robert E. Tarone

Dr. Portier claims to show highly significant dose-response relationships between glyphosate exposure and renal tubule tumors, lymphomas, and hemangiosarcomas among male mice in analyses summarizing five mouse carcinogenicity studies. His conclusion is flawed, because of the use of inappropriate statistical methods and the omission of relevant tumor data.

Dr. Portier's analyses are based on the Cochran-Armitage trend test statistic and the poly-3 trend test statistic. The p-values reported for both test statistics in Portier's comments are based on their asymptotic standard normal distributions (i.e., they provide approximate tests). This can be OK (i.e., the approximate p-values may be somewhat close to exact p-values) when there are sufficient numbers of animals with tumors, or when the dose levels are approximately evenly spaced. Neither is the case with the glyphosate mouse studies. The tumor rates are very low and the doses are nowhere near equally spaced. Because glyphosate was given at such high exposure levels, particularly in the highest dose groups, the test statistics are extremely skewed – i.e., they come nowhere near being distributed as standard normal variables.

I can give an example based on the Cochran-Armitage test statistic, for which both the exact p-value and its standard normal approximation can be computed (all p-values presented below were calculated using StatXact®, version 9). Consider the p-value of 0.008 for the mouse renal tumors in the 1997 CD-1 mouse study reported in Table 2 of Portier's comments. The corresponding tumor rates were 0/50, 0/50, 0/50 and 2/50. Calculating the Cochran-Armitage test statistic using dose levels 0, 165, 838, 4348 gives an exact one-sided p-value of 0.062 and an approximate one-sided p-value of 0.0078.

The small approximate p-values in Portier's tables frequently reflect the extreme skewness of the test statistics because of the large highest dose level. The use of approximate tests results in summary p-values for renal tumors and lymphomas in male mice that are too small because of the failure of the asymptotic normal distribution to approximate the exact distribution of the test statistics.

Based on the data in Table 6 of Portier's comments there might be reason for concern about a possible association between glyphosate exposure and hemangiosarcomas in male mice. The table shows that very few hemangiosarcomas were observed, but that all but one were observed in the highest exposure groups. The table, however, omits all hemangiosarcomas reported in the 2001 Swiss mouse study and the 2009 CD-1 mouse study. Hemangiosarcomas are reported by organ or system in the summary tables for these 2 studies, and it will be assumed that no animal has more than one hemangiosarcoma in the calculation of tumor rates. For the 2001 Swiss mouse study (study 13 in the 2015 Critical Reviews in Toxicology review paper by Greim, et al.) the hemangiosarcoma rates for males are 0/50, 0/50, 2/50, and 0/50. For completeness, the hemangiosarcoma rates for females in this Swiss mouse study are 1/50, 1/50, 0/50, and 0/50. For the 2009 CD-1 mouse study (study 14 in the Greim et al. review) the hemangiosarcoma rates for males are 2/51, 1/51, 2/51, and 1/51. Again for completeness, the hemangiosarcoma rates for females in this CD-1 mouse study are 1/51, 1/51, 1/51, and 1/51. Thus, the hemangiosarcoma data omitted from Table 6 provide no support for an increase in hemangiosarcomas at highest exposure levels in male mice, and provide no support at all for an increase in hemangiosarcoma rates in glyphosate-exposed females (consistent with the other 3 mouse studies).

In his comments, Portier expresses the opinion that two-sided tests are not appropriate in evaluating the rodent glyphosate studies. There is no statistical justification for this view. Perhaps in situations in which the type of tumor being evaluated is vanishingly rare, such a claim could be supported. The role of the nominal significance level is to protect against false positive findings in both directions (i.e., direct association of tumor rates with dose level and inverse association of tumor rates with dose level). Several of the trend test statistics in the glyphosate rodent studies are in the direction of an inverse association between exposure level and tumor rate, and, in fact, the one-sided exact p-value of the Cochran-Armitage trend test for renal tumors in the 1993 CD-1 mouse study (Table 2) is 0.042 in the direction of an inverse association. For hemangiosarcomas in male mice, the exact one-side p-values for a positive trend for the five studies in Table 6 are 0.503, 0.004, 0.062, 0.50, and 0.65, with the approximate test statistic in the direction of an inverse association between glyphosate level and hemangiosarcoma rate for three of the five studies. A number of inverse associations are observed in the individual glyphosate rodent

studies, and accordingly, two-sided p-values should be used in summarizing the results of these studies.

An appropriate statistical analysis based on exact p-values because of the small numbers of animals with tumors and the skewed distribution of the test statistics would not provide convincing evidence that glyphosate induces renal tumors, lymphomas, or hemangiosarcomas in male mice.

Document Seven

Comments of Christopher J. Portier, PhD.
USEPA (EPA-HQ-OPP-2016-0385-0094)
Glyphosate Issue Paper: Evaluation of Carcinogenic Potential
Response to comments prepared by Robert E. Tarone (dated October 27, 2016)
November 16, 2016

Disclaimer: This work was done with my own resources and on my own time. I have received no reimbursement for any of these comments and no other party has contributed to the drafting of these comments. These comments are solely my opinion and my responsibility.

Dr. Tarone has 3 main points to make in his comments regarding my evaluation of the mouse carcinogenicity data on glyphosate. I will take these in reverse order.

1. Dr. Tarone contends that the use of one-sided p-values is not justified. The purpose of this hazard evaluation is to determine if exposure to glyphosate is a carcinogenic hazard; it is not to determine if glyphosate can either reduce tumor rates or increase tumor rates. Thus, the justification for the use of a one-sided test is based upon the alternative of interest.
2. Dr. Tarone contends I have not used the data on hemangiosarcomas from the studies by Kumar (2001) and Wood *et al.* (2009). In fact, I did include the numbers by Wood *et al.* (2009) based on the numbers provided in Table 5.3-1 in the EFSA Renewal Assessment Report, Addendum I (8/31/2015) which records zero tumors in all groups exposed to glyphosate. The discrepancy between the numbers of EFSA and those of Dr. Tarone have to do with the inclusion of hemangiosarcomas in liver and kidney which do not appear to be included in the pathology designated as "hemangiosarcoma of multiple organs". This also appears to be the case in the historical control data set referenced in the EFSA Table 5.3.1 and cited by the EPA (Giknis and Clifford, 2005). I suggest EPA clarify this issue for the SAP. I did not use the data from Kumar (2001) because I did not have it at the time I first did this analysis; that is corrected below.
3. Dr. Tarone contends that, by not using the exact p-values, I am overstating the significance of the observed trends. He then goes on to cite some examples of where this changes the results and finally concludes that "*An appropriate statistical analysis based on exact p-values because of the small numbers of animals with tumors and the skewed distribution of the test statistics would not provide convincing evidence that glyphosate induces renal tumors, lymphomas, or hemangiosarcomas in male mice.*" While I agree with Dr. Tarone that, in cases where tumors are rare, the approximate p-value can overstate the significance of the findings, I strongly disagree with his final conclusion. Dr. Tarone has failed to address all of the analyses considered in my comments, and in doing so, has mislead the reader to believe all significance disappears. This is not the case as I now demonstrate.

   The spreadsheet attached ("AllTestValues.xlsx") to this set of comments re-evaluates all of the analyses in my previous comments using the chi-squared

test with the one-sided approximate p-value), the exact test[1], and an evaluation against the use of a historical control frequency of tumors[2] using the numbers presented in the EFSA Table 5.3-1 and also cited by the EPA (Giknis and Clifford, 2005). Note that there are problems with the use of these historical control values in the analyses as noted in my previous comments. However, they are sufficient for the analysis being provided here and demonstrate why a careful evaluation against historical controls is necessary.

For renal tumors, all of the individual studies for which the p-value was less than 0.05 for the approximate test have p-values greater than 0.05 and less than 0.065 for the exact test. Thus, we go from 3 significant studies to 3 marginal studies. However, there are a few important issues to consider on these numbers. The study by Sugimoto (1997) is the most extreme outcome possible and it is not possible with only 2 tumors to get a p-value smaller than the 0.059 value with the exact test. Similar statements hold true for the 1983 study and the 2001 study. The point is that for rare tumors, the exact test has a limited ability to identify a positive finding even though it uses the exact p-value. Thus, doing a direct evaluation against the historical controls is warranted. The historical control test shows statistical significance identical for all of the tests to those in my previous comments. Especially clear is the findings from analyzing all of the data simultaneously.

For Malignant Lymphomas, all analyses are marginal for the 1993 study and significant for the 1997 and 2009 studies. The results for the 1983 study do not change and the results for the 2001 study is mixed. In general, the findings for all of the analyses presented in my previous comments are less significant, but still support an overall conclusion of a positive response for malignant lymphomas, especially where all of the CD-1 mice are combined for analysis.

Finally, for Hemangiosarcomas, after including the findings from the Kumar, 2001 study, there are very minor differences in the overall significance of the findings regardless of the method used to compute the p-values. For the

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[1] The exact test p-value was calculated using a permutation approach where 20,000 random data sets with the same fixed number of total tumors as the original data set were analyzed and compared to the original one-sided Z-statistic for the trend test. The p-value for this test is based upon the fraction of the 20,000 experiments resulting in a Z-statistic greater than or equal to that observed in the original data set.

[2] The p-value for the historical control test was calculated by randomly generating 10,000 datasets under the null hypothesis of no difference in tumor response across all groups with a true p-value equal to the historical control values. The p-value for this test is based upon the fraction of the 10,000 experiments resulting in a Z-statistic greater than or equal to that observed in the original data set.

1997 study, we again have a case where the smallest p-value possible from the exact test is the one given. The findings for the combined analyses of all of the studies do not change and remain clearly significant.

In summary, even with the use of the p-values from the exact test, the only conclusion that can be drawn from the mouse carcinogenicity data is that there is "very strong evidence in mice for renal tumors, hemangiosarcomas and malignant lymphomas. EPA's exclusion of doses above 1000 mg/kg/day is unscientific and their argument of a lack of significance above this dose is unsupported."

Document Eight

Comments on the laboratory animal carcinogenicity studies evaluated in EPA's Glyphosate Issue Paper: Evaluation of Carcinogenic Potential, and an assessment of the comments by Dr. Christopher Portier on these studies

by Joseph K. Haseman
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I. Introduction

Because of my many years of experience working with the National Toxicology Program (NTP) in the design, analysis and interpretation of their rodent cancer bioassays, I was asked to examine a document entitled "Glyphosate Issue Paper: Evaluation of Carcinogenic Potential", prepared by the EPA, whose purpose is to assess the carcinogenic potential of glyphosate for human health. The document deals with epidemiology, laboratory animal cancer, and genetic toxicity studies, but the rodent cancer data will be the focus of my comments. I will also comment on Dr. Christopher Portier's evaluation and statistical analysis of the glyphosate laboratory animal cancer data and his critique of the EPA report.

II. EPA's evaluation of the glyphosate rodent cancer data

The EPA evaluated six mouse and nine rat glyphosate carcinogenicity studies. No significant increasing trends in tumor incidence were seen in three of the mouse studies, while three studies showed isolated statistically significant ($p<0.05$) trends - hemangiomas in one study, hemangiosarcomas in in a second study, and malignant lymphoma in a third study. This third study also showed a statistically significant trend for lung adenocarcinoma, but this trend was not statistically significant when all lung tumors were considered (i.e., lung adenoma or adenocarcinoma combined).

In five of the nine rat studies, no significant carcinogenic effects were found. The other four studies showed isolated statistically significant trends, but in different tumor types that were also different from the tumor types that produced significant trends in mice – one study showed a significant trend for testis tumors, a second for thyroid c-cell tumors, a third for liver tumors, and a fourth for mammary gland tumors. The second study cited above also showed a statistically significant trend for liver adenoma, but no significant trend for all liver tumors (adenoma or carcinoma combined).

Based on these results, the EPA concluded that "Based on the weight-of-evidence, the agency has determined that any tumor findings observed in the rat and mouse carcinogenicity studies for glyphosate are not considered treatment-related. Tumor findings observed at the highest doses tested were also not reproduced in studies in the same animal strain at similar or higher doses. Furthermore, even if the high-dose tumors were considered treatment-related, these findings are not considered relevant for human health risk assessment based on the use pattern and potential exposures for glyphosate."

III. Dr. Portier's evaluation of the glyphosate rodent cancer data

The EPA's conclusion given above disagrees with the International Agency for Research on Cancer (IARC), who in March, 2015 concluded that glyphosate was a probable human carcinogen (Group 2A), based in part on "sufficient" evidence of carcinogenicity in laboratory animals.

Dr. Christopher Portier played a key role in IARC's classification of glyphosate as a Group 2A probable human carcinogen, so it is not surprising that his report (dated 10-4-16) disagrees with the EPA's assessment of glyphosate, and he defends IARC's classification of glyphosate as a probable human carcinogen. My comments will focus on those portions of Dr. Portier's report that deal with the rodent cancer studies.

I have many major concerns with Dr. Portier's report. For example,

(i) his statistical analyses of tumor trends is an approximate rather than an exact trend test, and this approximate test can exaggerate the statistical significance of trends (especially for rare tumors) by as much as 10 fold;

(ii) Dr. Portier misuses poly-3 tumor rates in several ways. First of all, his "poly-3 tumor rates" for the 24 month studies are nothing more than the overall observed tumor rates – there is no survival adjustment at all.
For the 18 month studies, Dr. Portier's "poly-3 tumor rates" are nothing more than the observed overall tumor rates extrapolated to 24 months by a poly-3 adjustment that erroneously assumes that all tumor-free animals in the 18 month study survived until the end of the study;

(iii) his attempts to pool tumor data across unrelated studies are fatally flawed and greatly exaggerate the significance of the pooled data;

(iv) he attributes various actions and statistical evaluations of the data to the EPA that were in not in fact carried out;

(v) his efforts to incorporate historical control data into the overall evaluation are flawed;

(vi) he underestimates the importance of pre-neoplastic lesions in the development of cancer; and

(vii) he apparently fails to appreciate that the frequency of significant increasing trends in tumor incidence across the 15 glyphosate studies is totally consistent with chance expectation. Moreover, the significant trends that were observed showed no consistency regarding the target site or type of "carcinogenic effect".

Dr. Portier presents 23 specific points of emphasis concerning the EPA's animal carcinogenicity data findings. The most important of these are

Point 4 [echoed in Point 10]: "EPA consistently dismisses significant findings in rat [and mouse] studies because of a lack of a preneoplastic finding."

Response – Pre-neoplastic lesions are important for certain tumor types, and should be considered in an overall weight of evidence evaluation of the data, especially for marginal increases in tumor incidence. For example, when interpreting (and dismissing the biological significance of) the marginal increase in kidney tumors seen in male mice in the 1983 Monsanto (Knezevich and Hogan) glyphosate study, Dr. Robert Squire, a world-renowned expert in rodent pathology, stated "I know of no instance where a renal carcinogen was given at a dose sufficient to induce tumors without also inducing tubular toxicity and hyperplasia".

Point 5 [echoed in Point 11 for mice]: "EPA consistently dismisses significant findings in rat studies because of a lack of a significant pairwise comparison even though there is a significant trend."

Response - I agree with Dr. Portier that a trend test is more sensitive than pairwise comparisons for detecting carcinogenic effects, and the focus of my comments will be on the trend tests. One of my most important findings

(discussed later in this report) is that the frequency of statistically significant ($p<0.05$) trends across the 15 glyphosate studies is actually lower than chance expectation. Moreover, there is no consistency to the significant trends that are observed. Thus, the "significant trends" are most likely due to chance, not to glyphosate.

Point 8: "The strong positive response seen for thyroid c cell carcinomas in female rats in one of these studies [Stout and Reucker] should be considered positive".

Response - This "strong positive response" noted by Dr. Portier is a single carcinoma in the mid dose group! I suspect that Dr. Portier intended to refer to thyroid c cell adenoma, but this pattern of response (2/57 - 2/60 - 6/59 - 6/55) is hardly a "strong positive response". Even when the carcinoma in the mid dose group is included, the overall incidence of thyroid c cell tumors (2/57 -2/60 - 7/59 - 6/55) in female rats is only barely significant by a trend test ($p=0.042$; see Table 4.7 of the EPA report) and this non-monotonic pattern of tumor response does not support Dr. Portier's conclusion on page 11 that "the dose response is clear."

This is exactly the sort of marginal tumor trend that is likely due to chance because of the large number of tumor types/sites evaluated in nine rat studies and also because the concurrent control tumor rate appears to be abnormally low. The control rate of thyroid c cell tumors in female rats in the Lankas study (the only other rat study for which I have complete tumor incidence data) is 6/47, which is actually greater than the thyroid c-cell tumor rate seen in the high dose group in the Stout and Ruecker study.

Dr. Portier concludes (page 11) that this single marginal trend is the only definitive carcinogenic effect seen in the nine rat studies of glyphosate. In my view, the overall scientific weight of evidence is overwhelming that this marginal trend is due to chance and not to glyphosate.

Points 13 and 15: The EPA misuses historical control data.

Response - Ironically, it is Dr. Portier who misuses historical control data, at least as it relates to renal tumors. The historical control database that he uses for the Monsanto (1983) male mouse glyphosate study is inappropriate because the kidneys from the historical control database were not as carefully scrutinized as the kidneys from the Monsanto study, and thus the

historical data will almost certainly under-report the tumor rate that would be found with a more rigorous histopathology evaluation.

That is, the original kidney slides from the Monsanto study received an additional level of review (revealing a tumor in the control group that had been missed from the initial routine examination), and additional step sections of the kidney were also taken and examined for possible tumors. This additional rigor was not present in the historical control data, so using this database as a reference point provides an "apples vs. oranges" comparison, unless one re-evaluates the historical control data with the same rigor used in the Monsanto glyphosate study, including additional kidney step sections to look for additional tumors.

Point 14: "EPA relies on two-sided p-values for trend tests when one-sided p-values would be more appropriate for identifying adverse effects" (a point made multiple times in his report).

Response - Dr. Portier's assertion that the EPA is using two-sided trend tests for the statistical analysis of tumor trends is incorrect. The EPA report clearly states: "In the current evaluation, the Cochran-Armitage Test for Trend (Snedecor and Cochran, 1967; one-sided) was used." I could find no mention in the EPA report of any use of two-sided tests.

I suspect that Dr. Portier is trying to understand why his trend test p values are so much more significant than the EPA's when they are applying the same trend test to the same tumor data. However, the answer is not one-sided vs. two -sided tests; rather it is approximate vs. exact tests.

Dr. Portier (and IARC) used an approximate one-sided trend test, which can greatly exaggerate the statistical significance of the trend, especially for rare tumors, whereas I have confirmed (at least for the rare tumors) that the trend test p values reported by the EPA are one-sided p values from an exact trend test. There is no need to carry out flawed approximate tests that exaggerate statistical significance, when exact tests are available.

The exaggerated statistical significance associated with the use of approximate trend tests can be extreme. For example, in the 1983 Monsanto male mouse study, even a single tumor in the high dose group would have been sufficient for the approximate Cochran-Armitage trend test to indicate

a statistically significant (p=0.045) trend (see Appendix A), whereas the exact trend test p value is p = 50/198 or p = 0.25. That is a huge difference!

Some might argue that no one would ever use an approximate trend test when there were only 1 or 2 total tumors across all the dosed and control groups (and the exact trend test is so easy to calculate), and even if they did, they would certainly recognize this error and discount the exaggerated statistical significance associated with the approximate trend test.

They would be wrong.

Twice Dr. Portier applied the approximate trend test to a "two-tumor trend" (i.e., 0/50 - 0/50 - 0/50 - 2/50) and then regarded the exaggerated p value (p=0.008) to be both a biological and statistically significant response (renal tumors in his Table 2 and hemangiosarcoma in his Table 6). However, the exact trend test p value in both cases is

$$p = (50/200) \times (49/199) \text{ or } p = 0.062,$$

which is not even significant at the $p<0.05$ level.

The exaggerated statistical significance associated with Dr. Portier's approximate trend test is exacerbated by the unequal spacing of the doses. This is illustrated below by a comparison of exact and approximate trend tests applied to the two-tumor trend of 0/50 - 0/50 -0/50 - 2/50 for three different dose metrics.

	Trend test p values	
Doses	Exact	Approximate
0, 8, 9, 10	p=0.062	p=0.122
0, 1, 2, 3	p=0.062	p=0.029
0, 1, 2, 100	p=0.062	p=0.007

This comparison illustrates another serious flaw associated with the approximate trend test used by Dr. Portier and IARC when applied to rare tumors. Under the null hypothesis of no treatment effect, the probabilities associated with the possible distributions of tumors across groups should not depend upon the doses used (which are assumed under the null hypothesis to have no effect on tumor incidence). The actual doses are relevant to the ranking of the possible distributions of tumors regarding a possible treatment

effect, but the doses should have no influence on the actual probabilities of the various tumor distributions under the null hypothesis. This explains why the exact p values are the same in the example above, regardless of dose. Note, however, that the approximate trend test p values in this example depend upon the dose metric and vary by a factor of nearly 20.

Another example: the EPA Cochran-Armitage trend test for the Monsanto kidney tumor data (1/49, 0/49, 1/50, 3/50) indicates a non-significant trend (p=0.065; see EPA's Table 4.12), whereas Dr. Portier's (and IARC's) trend test analysis of the same data indicates a significant trend (p=0.03; see his Table 2). Dr. Portier incorrectly asserts that this is because the EPA is using a two-sided test. However, the real reason is that the EPA is using an exact trend test, while Dr. Portier and IARC are using an approximate test.

Incidentally, if a dose metric of equally spaced doses had been used (which would be reasonable, since the doses are approximately equally spaced on a log scale), then the trend in kidney tumors noted above would not be significant (p=0.082) even by Dr. Portier's flawed approximate trend test.

Finally, the Pathology Working Group (PWG), who examined carefully the kidney tumors in the 1983 Monsanto male mouse study "firmly" and "unanimously", agreed that "the incidences of renal tubular-cell neoplasms in this study are not compound related."

The trend test p values for the kidney tumor rates in the other studies reported by Dr. Portier are also exaggerated, as is demonstrated below.

Year of study	Kidney tumor rates	Cochran-Armitage trend test p values	
		Approximate[1]	Exact
1983	1/49 - 0/49 - 1/50 - 3/50	0.03	0.065
1997	0/50 - 0/50 - 0/50 - 2/50	0.008	0.062
2001	0/49 - 0/49 - 1/50 - 2/50	0.04	0.063

[1] as reported by Dr. Portier in his Table 2

Note that none of these trends are statistically significant at the $p<0.05$ level when an exact trend test is used. In fact, it could be argued that the most significant trend for the kidney tumor data is the decreasing trend in kidney tumor incidence seen in the 1993 Atkinson study: 2/50 - 2/50 - 0/50 - 0/50. By a one-sided exact trend test that evaluates deceases in tumor incidence,

this decrease is significant: p=0.042. By a one-sided exact trend test that evaluates increases in tumor incidence, the p value is 0.98.

One of the most significant trends seen in all of the glyphosate studies is the increasing trend in hemangiosarcomas in male mice in the 1993 Atkinson study. Dr. Portier and the EPA disagree regarding the denominators for the tumor rates (compare EPA's Table 4.14 with Dr. Portier's Table 6,) but to illustrate the exaggerated significance of Dr. Porter's p value, I will assume that his tumor rates are correct: 0/50 - 0/50 - 0/50 - 4/50. It is very easy to calculate the exact trend test p value for this distribution of tumors. It is

$$P = (50 \times 49 \times 48 \times 47)/(200 \times 199 \times 198 \times 197) \text{ or } p=0.004$$

In his Table 6, Dr. Portier reports the p value for this trend to be p=0.0004, which exaggerates by a factor of 10 the exact p value for this trend!

Dr. Portier's statistical analysis also misuses the (survival-adjusted) poly-3 test, a test which he himself developed. For example, in the last column of his Tables 2, 4, and 6, he reports the results of two trend tests, one a Cochran-Armitage trend test based on the observed tumor rates, and the other supposedly a poly-3 trend test based on the poly-3 tumor rates.

However, this is very misleading. Application of the poly-3 test requires individual animal survival and tumor data. Since Dr. Portier almost certainly did not have this information, he could not carry out a meaningful poly-3 test. Instead, his "poly-3 test" for the 24 month mouse studies is apparently nothing more than a repeat of the Cochran-Armitage trend test applied to the observed tumor rates. This is confirmed by the fact that the p values for the unadjusted and poly-3 adjusted tumor trends are identical for all the 24 month studies reported in Dr. Portier's Tables 2, 4, and 6. This is also confirmed by the fact that the graphs of his "poly-3 rates" from the 24 month studies (see http://eomsociety.org/images/PDF/PortierOLII.pdf) are clearly the observed tumor rates. Thus, for the 24 month studies his "poly-3 tests" make no adjustment for survival at all.

For the 18 month studies, Dr. Portier's "poly-3 tumor rates" are nothing more than the observed overall tumor rates extrapolated to 24 months by a poly-3 adjustment that erroneously assumes that all tumor-free animals in the 18 month study survived until the end of the study. Thus, this "Portier extrapolation tumor rate" has little scientific relevance, since many animals

died before 18 months, and their survival times are not taken into account, as they must be in the calculation of an appropriate poly-3 tumor rate. This matter is discussed more fully in Appendix B.

Points 16-18: Dr. Portier opines (Point 18) that some of the 15 studies considered by the EPA should be excluded, and that the EPA analysis should have included some positive results that he provided to them.

Response: I leave that judgment to others. My primary analysis will be based on the15 studies considered relevant by the EPA, and the significant tumor trends that they identify and report. However, in response to Dr. Portier's Points 19-21 below, I will use only the studies that he deems adequate when evaluating the pooling of results across studies to illustrate how his pooling (even when limited only to the studies that he deems to be relevant) greatly exaggerates the statistical significance of the data.

Points 19-21: "EPA did not analyze the consistency across mouse studies on the findings relating to renal tumors..., malignant lymphomas,...and heamngiosarcomas [sic]."

Response - In my opinion, Dr. Portier's statistical analysis, which pools observed and extrapolated tumor rates from five 18 and 24 month studies, involving different strains of mice with different tumor background rates, pathology protocols and dosage regimens, spanning a period of nearly 30 years, and incorrectly assuming that in the 18 month studies all tumor-free animals survived until the end of the study, is fatally flawed.

Dr. Portier carries out two trend test analyses for the pooled tumor data, and the results are reported in the final column of Tables 3, 5, and 7. The first apparently uses observed ("original") tumor rates from both the 18 and 24 month studies together in a single trend test, which is inappropriate and is the same "serious error" that he attributes to the EPA's use of historical control data. That is, one cannot sensibly use observed tumor rates from both 18 and 24 month studies together in a single huge trend test analysis.

The second trend test analysis claims to use a poly-3 adjustment. However, as noted above, Dr. Portier's graphing of these "poly-3 tumor rates" (see http://eomsociety.org/images/PDF/PortierOLII.pdf) makes it clear that this trend test uses observed tumor rates from the 24 month studies and the

fatally flawed "Portier extrapolation tumor rates" from the 18 month studies. This too is inappropriate.

Another problem is Dr. Portier's combining of dosed groups in order to have a manageable number of dosed groups for a trend test. He condenses the 15 dosed groups to three: 15-310, 310-1500, and >1500 mg/kg/day (see http://eomsociety.org/images/PDF/PortierOLII.pdf). This pooling is inappropriate, since it (for example) regards a dose of 15 mg/kg/day in an 18 month study Swiss Webster mouse study and a dose of 300 mg/kg/day in 24 month CD-1 mouse study as equivalent doses with regard to producing a tumor effect. The pooling of dosed groups is also subjective, so a critic could argue that the specific choice of how the dose groups were pooled was done after the studies were over so as to maximize the significance of the trend.

In summary, Dr. Portier pooled tumor rates from five disparate studies of different study lengths, with different strains of mice that used different pathology protocols and different doses (inappropriately combining dosed groups), using an approximate trend test that exaggerates the statistical significance of the trend.

Dr. Portier carried out two trend tests, one based on the observed tumor rates in the 18 and 24 month studies (which is inappropriate) and the other based on observed tumor rates in 24 month studies and the fatally flawed Portier extrapolation tumor rates in 18 month studies, which is also inappropriate. Proper poly-3 adjusted tumor rates were not used in either analysis. I strongly recommend discounting all the pooled results Dr. Portier presents in Tables 3, 5, and 7.

My recommendations for evaluating the glyphosate rodent cancer data is given in the following section.

IV. My own evaluation of the glyphosate rodent cancer data

Any global assessment of the carcinogenic potential of glyphosate in the rodent cancer bioassays must recognize the fact that when multiple rodent carcinogenicity studies are carried out on a specific test agent, it is inevitable that some statistically significant ($p<0.05$) increasing trends in tumor incidence will be found. Some could be due to chance; others could be reflecting real carcinogenic effects. When making an overall weight of

evidence decision on this matter, the following three questions should be asked (and answered):

(1) Does the overall frequency of these significant trends exceed what would be expected by chance alone?

(2) Regardless, is there a consistency of target sites across the studies that show significant trends?

(3) Regardless, are there one or more tumor increases that are so strong statistically (e.g., $p<0.0001$) that it would be extremely unlikely that such responses could be a chance occurrence?

The EPA (2016) report gives 11 statistically significant ($p<0.05$) tumor trends for the glyphosate mouse and rat studies, with the single strongest trend being $p=0.002$ (see Tables 4.1, 4.4, 4.7 (2), 4.9, 4.10 (2), 4.14, 4.15, 4.16, and 4.17). How likely would it be to find this many significant increasing trends in tumor incidence occurring by chance? Answering this question requires knowledge of (1) how many studies were evaluated by the EPA; and (2) how many trend tests were made within each study.

The EPA evaluated the results from 9 rat studies and 6 mouse studies (males and females evaluated separately). Using tumor incidence data provided to me from four representative glyphosate studies (the Monsanto and Sugimoto mouse studies; the Lankas and Stout and Reucker rat studies), I determined how many tumor types (and tumor combinations) occurred in a sufficient number of animals to permit a meaningful application of a trend test (i.e., occurred in at least three animals). These are summarized in Appendix C for mice and in Appendix D for rats. These data indicate that the numbers of meaningful trend tests are approximately 10.5 for male mice, 15 for female mice, 21.5 for male rats, and 25.5 for female rats.

Assuming that these data are representative of the other glyphosate studies (I have confirmed that these values are very similar to the number of trend tests carried out for these sex/species groups in recent NTP rodent carcinogenicity studies), then the total number of trend tests carried out for the 15 glyphosate rat and mouse studies was approximately $\{(9 \times [21.5+25.5]) + (6 \times [10.5 + 15])\}$ or 576, and the number of significant ($p<0.05$) trends expected by chance is approximately 576×0.05 or 28.8.

This expected number of significant ($p<0.05$) trends is actually much greater than the 11 significant trends given in the 2016 EPA report. Thus, even if some of the glyphosate studies are considered inadequate, as opined by Dr. Portier, and/or if additional positive trend tests are added in to what was actually observed, there is no evidence that the number of significant tumor trends found in these glyphosate studies exceeds chance expectation.

To answer the second question posed above, I focused my statistical analysis on the number of tumor sites/types in each study that could be meaningfully evaluated by a trend test, rather than on the actual number of trend tests. For example, in a given study, a specific target site (e.g., liver) could have three trend tests applied: adenoma, carcinoma, and adenoma or carcinoma combined). When this occurred, the trend test p value for the tumor combination was considered the "definitive" p value for that site, and the site was counted only once, not three times.

I found that there were seven instances in which significant ($p<0.05$) carcinogenic trends were observed for specific cancer sites/types in the fifteen glyphosate studies. Interestingly, they each occurred in a separate study, and are summarized below.

Study	Sex-species	Organ site or cancer type
Lankis	Male Rat	Testis
Wood	Female Rat	Mammary gland
Stout	Female Rat	Thyroid gland
Brammer	Male Rat	Liver
Sugimoto	Female mouse	Hemangioma
Wood	Male mouse	Malignant Lymphoma
Atkinson	Male mouse	Hemangiosarcoma

Using the data in Appendices C and D, we can count the total number of tumor sites/types that can be meaningfully evaluated for trends in the four representative glyphosate studies and use these values (which averaged 15.5 for mice and 28 for rats) to estimate the total number of tumor site/types that could be meaningfully evaluated by trend tests in all 15 glyphosate studies.

This number of tumor sites/types [namely, (15.5 x 6) + (28 x 9) or 345] is considerably less than the estimated number of actual trend tests given above (576). Even so, the number of tumor sites/types that would be expected to

show significant ($p<0.05$) trends by chance alone (0.05 x 345 or 17.25) is still far in excess of what was actually observed (7).

Note that none of the seven significant ($p<0.05$) site-specific tumor trends were replicated in other studies. In addition to this lack of consistency regarding target site, note that kidney tumors do not even appear on this list, since by an exact trend test, there were no significant ($p<0.05$) increasing trends in kidney tumor incidences in any of the studies evaluated by the EPA, although there was (arguably) a significant ($p<0.05$) decreasing trend in kidney tumor incidence in one of the studies, as noted above.

To answer the third question posed above, I note that even the most significant trend reported by the EPA ($p=0.002$) is not an unusual finding, since this is similar to chance expectation of 1.15 (576 x 0.002).

Thus, the answers to the three questions posed above are No, No, and No. This is summarized in the table below

Table 1 - Summary of the estimated number of tumor incidence trend tests carried out in the 15 glyphosate studies evaluated by the EPA and their comparison to chance expectation

	Estimated Number	Significant ($p<0.05$) trends	
		Expected	Observed
All Trend tests	576	28.8	11
Trend tests for unique tumor sites/types	345	17.25	7
Strongest trend ($p=0.002$)	1	1.15	1

Finally, we can use the data in Appendices C and D to estimate the number of specific sites/types of cancer that in theory could show possible carcinogenic effects when pooled over studies. This (conservative) estimate is 61 possible carcinogenic effects: 11 in male mice, 14 in female mice, 17 in male rats, and 19 in female rats. This is a conservative estimate, since the other 7 rat studies and 4 mouse studies not included in Appendices C and D could add additional cancer sites/types to this list.

Thus, even if the three "pooled" cancer target sites identified by Dr. Portier's fatally flawed pooled analysis summarized in his Tables 3, 5, and 7 are considered statistically significant ($p<0.05$) carcinogenic effects (and I certainly do not), this would still be consistent with chance expectation (61 x 0.05 or 3.05).

Thus, for all measures given above, the frequency of "significant trends" is consistent with or actually less than chance expectation.

V. Summary and Conclusions

Summary: The available data provide strong evidence that the frequency of significant increasing trends in tumor incidence across the various glyphosate studies is consistent with chance expectation. Moreover, the significant trends that were observed showed no consistency regarding the target site or type of "carcinogenic effect" across studies. Even the strongest tumor trend observed was consistent with chance expectation.

Furthermore, I strongly disagree with Dr. Portier's statistical analysis and conclusions for a number of reasons, the most important of which are

(1) He uses an approximate rather than an exact trend test, and his approximate trend test (especially for rare tumors) greatly exaggerates the significance of the trend, by as much as 10-fold.

(2) Dr. Portier's misuses poly-3 tumor rates. His "poly-3 tumor rates" for the 24 month studies are apparently nothing more than the overall observed tumor rates – there is no survival adjustment at all. For the 18 month studies, Dr. Portier's "poly-3 tumor rates" are nothing more than the observed overall tumor rates extrapolated to 24 months by a poly-3 adjustment that erroneously assumes that all tumor-free animals in the 18 month study survived until the end of the study. Any subsequent statistical analyses based on these flawed "poly-3 tumor rates" have little or no scientific value.

(3) For the rat data, Dr. Portier concludes that there is only one definitive carcinogenic effect: an increase in thyroid c-cell tumors in female rats, an increase that he considers a "strong, clear dose response". However, I strongly disagree with his characterization of this marginal ($p=0.04$) trend (2/57, 2/60, 7/59, 6/55), and when one considers the large number of trend

test carried out for the rat studies, and that the concurrent control tumor rate in this one study was abnormally low, the only scientifically defensive conclusion is that this marginal trend is due to chance, not to glyphosate.

(4) In mice, Dr. Portier's pooling of observed and/or Portier extrapolation tumor rates over five disparate studies, is inappropriate for a variety of reasons as discussed in this report. Moreover, even if an appropriate statistical analysis had found the three pooled tumor trends evaluated by Dr. Portier in Tables 3, 5, and 7 to be statistically significant ($p<0.05$), the frequency with which these significant pooled trends occurred would still be consistent with chance expectation.

(5) Dr. Portier's use of historical control data is flawed, and his claim that the EPA used 2-sided tests to evaluate tumor trends is false.

Conclusion: Based on the data given to me, the evidence is very strong that all of the statistically significant ($p<0.05$) increasing trends in tumor incidence observed in the various glyphosate rodent cancer studies are due to chance, not to glyphosate.

Appendix A: Application of the Cochran-Armitage trend test asymptotic approximation to hypothetical glyphosate data showing a single kidney tumor in the high dose group (i.e., 0/49 - 0/49 - 0/50 - 1/50)

Let m = total number of tumors (1)
Let N = total number of animals (198)
Let d_i denote the i^{th} dose (0, 161, 835, 4945 mg/kg/day)*
Let x_i denote the number of tumors found at the i^{th} dose (0-0-0-1)
Let n_i denote the number of animals examined at the i^{th} dose (49-49-50-50)

Then the large sample normal approximation to the Cochran Armitage trend test statistic can be written (see Corcoran, C.D. and Mehta, C.R. (2002). Exact level and power of permutation, bootstrap, and asymptotic tests of trend. Journal of Applied Statistical Methods 1: 42-51.)

Z = (T-E) / SD, where
T = sum of $d_i x_i$ = 4945
E = (m/N) x [sum of $d_i n_i$] = 1499.44
SD = square root of $\{[(m(N-m))/(N(N-1))]$ x Sum $[(d_i - (E/m))^2 n_i]\}$ or
SD = 2027.1

Z = (4945 – 1499.44) / 2027.1 = 1.70, p=0.045

*Doses reported by the EPA for this study (see Table 4.12). The doses used by Dr. Portier are slightly different: 157, 814, and 4841 mg/kg/day; see his Table 2, but yield basically the same result (Z=1.70; =0.045).

Thus, by the Cochran-Armitage large sample normal approximation, even a single tumor can be considered a "statistically significant (p<0.05) trend". The exact p value for this "trend" is p = 50/198 or p=0.25, not p =0.045.

The exaggerated statistical significance of the approximate Cochran Armitage trend test can be demonstrated in another way. Suppose that the underlying control kidney tumor rate is in the 0.3% to 1% range. Using the Monsanto doses given above, it is easy to show that for the approximate Cochran Armitage trend test carried out at the nominal p<0.05 level, the actual significance level is in the 9-11% range, depending upon the exact background tumor rate (details available upon request). This exaggerated statistical significance is unacceptable.

Appendix B: Dr. Portier's misuse of poly-3 tumor rates

The principle of the poly-3 tumor rate is that an animal dying early in the study has a lower risk of tumor development than an animal surviving until

the end of the study. The poly-3 method adjusts downward the denominator of the observed tumor rate by not giving full weight to the tumor-free animals that die early in the study.

Specifically, an animal surviving until the end of the study or an animal dying with a tumor during the study is given full weight (n=1). A tumor-free animal dying before the end of the study is given a "weight" of the proportion of the full study that it survived raised to the third power. For example, if a tumor-free animal survived half of the study, it would be "down weighted" as $n=(1/2)^3$ or n=0.125. The sum of all the n's would then be the denominator of the poly-3 adjusted tumor rate. Importantly, calculation of a poly-3 tumor rate requires knowledge of each individual animal's survival time and tumor status.

Dr. Portier's misuses poly-3 tumor rates in several ways. First of all, his "poly-3 tumor rates" for the 24 month studies are nothing more than the overall observed tumor rates – there is no survival adjustment at all!

For the 18 month studies, Dr. Portier adjusts the observed tumor rates, but does it incorrectly. For example, when attempting to extrapolate what a kidney tumor rate of 2/50 seen in the high dose group in the 1997 18 month study might have been in a 24 month study, Dr. Portier states (page 15) "in order to compare all 5 studies, we must use the Poly-3 adjustment to extrapolate the 18 month studies to estimate what we think the cancer risk would have looked like at 24 months". He further states (page 16) "In the 1997 study [high dose male mouse group], 48 animals had no tumors at 18 months; the poly-3 adjustment reduces this to 20.25 [i.e., $48\ (18/24)^3$], leading to an incidence estimate of 2/22.25=9%" .

However, appropriate poly-3 tumor rate calculations must be based on the time of death of each individual animal. Dr. Portier's calculations given above assume that all 48 tumor-free animals in the high dose male mouse group in the 1997 study survived until the 18 month terminal sacrifice, which was clearly not the case. Thus, in my view, these "Portier extrapolation tumor rates" are fatally flawed.

Appendix C: Tumors occurring with sufficient frequency for a meaningful trend test: two representative glyphosate mouse studies

	Monsanto		Sugimoto	
Site/type	M	F	M	F

Liver	3*	3*	3*	1
Lung	3*	3*	3*	3*
Kidney	1	-	-	-
Bone: Osteoma/osteosarcoma	-	-	1	-
Uterus/cervix: Leiomyoma/leiomyosarcoma	-	3*	-	3*
Uterus: Endometrial adenocarcinoma	-	1	-	-
Harderian gland: adenoma	-	1	1	1
Adrenal gland: cortical adenoma	1	-	-	-
Adrenal gland: Benign A cell tumor	-	-	-	1
Testis: interstitial cell tumor	1	-	-	-
Mammary gland: ductal adenocarcinoma	-	1	-	-
Mammary gland: adenoma/adenocarcinoma	-	-	-	2#
Skeletal muscle: Rhabdomyosarcoma	-	-	-	1
Spleen: hemangioendothelioma	-	1	-	-
All sites: Lymphosarcoma	1	1	-	-
All sites: Malignant lymphoma	-	-	1	1
All sites: Liposarcoma	1	-	-	-
All sites: Granulocytic leukemia	-	1	-	-
All sites: Hemangioma /hemangiosarcoma	-	-	1	2^
Total	11	15	10	15

*benign, malignant, and benign and malignant combined

#adenocarcinoma and adenoma or adenocarcinoma combined; too few adenomas for a meaningful trend test analysis

^hemangiomas and hemangiomas or hemangiosarcomas combined; too few hemangiosarcomas for a meaningful trend test analysis

Appendix D: Tumors occurring with sufficient frequency for a meaningful trend test: two representative glyphosate rat studies

Lankas Stout and

			Reucker	
Site/type	M	F	M	F
Pituitary gland	3*	3*	1	2#
Liver	3*	3*	3*	3*
Pancreas (islet cell)	2#	3*	2#	1
Thyroid gland: C-cell tumors	2#	3*	3*	2#
Thyroid gland: Follicular cell tumors	1	1	2#	1
Parathyroid gland: adenoma	-	-	1	1
Skin: Keratoacanthoma	-	-	1	-
Skin: Squamous cell tumors	-	-	3*	-
Subcutaneous tissue: Fibroma/fibrosarcoma	3*	-	2#	2#
Subcutaneous tissue: Fibrous histiocytoma	-	-	-	1
Adrenal gland: Pheochromocytoma	1	1	1	1
Adrenal gland: Cortical tumors	1	3*	1	3*
Brain: Glioma	1	-	-	-
Brain: Astrocytoma	-	-	1	-
Kidney: Lipoma/Liposarcoma	-	-	1	-
Thymus: Lymphoma/Lymphosarcoma	-	-	-	1
Uterus: Adenoma	-	1	-	-
Uterus: Polyp	-	1	-	1
Ovary: Granulosa cell tumor	-	1	-	1
Testis interstitial cell tumor	1	-	1	-
Mammary gland: Fibroadenoma	-	1	-	1
Mammary gland: Adenoma/adenocarcinoma	-	3*	-	3*
All sites : Lipoma	1	-	-	1
All sites: Reticulum cell sarcoma	1	1	-	-
All sites: Malignant lymphoma	-	1	-	-
Total	20	26	23	25

*benign, malignant, and benign and malignant combined

#benign and benign and malignant combined; too few malignant tumors for a meaningful trend test analysis.

Addendum

by Joseph K. Haseman
J. K. Haseman Consulting
November 23, 2016

Today I received comments from Dr. Robert Tarone (dated 10-27-16) who expressed concerns regarding Dr. Portier's statistical analysis of the glyphosate mouse data, as reported in a 10-4-16 memo. I also received a copy of Dr. Portier's response to these concerns, dated 11-16-16. I have the following response to both Drs. Portier and Tarone.

Dr. Tarone's primary criticisms echoed one of my own concerns (made independently) that Dr. Portier's use of an approximate rather than an exact trend test greatly exaggerates the significance of the trend for rare tumors. Importantly, Dr. Portier now concedes that "the approximate p-value can overstate the significance of the findings", although he attempts (unsuccessfully in my view) to minimize the impact that this has on the overall interpretation of the data.

In his latest response Dr. Portier calculates what he considers to be exact trend test p values for the three major tumors of interest in mice. His own calculations now confirm my conclusions given in the body of this report that (i) the approximate trend test can exaggerate the statistical significance of the trend by as much as 10 fold (hemangiosarcomas in the 1993 Atkinson study); and (2) none of the three renal tumor trends that Dr. Portier originally reported as significant ($p<0.05$) were actually significant at the $p<0.05$ level by an exact trend test.

This second result is especially important. The kidney tumors are a potentially important endpoint in the interpretation of the mouse study. Rather than finding three significant ($p<0.05$) kidney tumor trends (one highly significant ($p<0.01$) trend), Dr. Portier now concedes that none of these three trends were actually significant ($p<0.05$). In fact, as noted by Dr. Tarone (agreeing with my own calculations) the strongest trend for male mouse kidney tumors "is 0.042 in the direction of an inverse association." Had IARC realized all this at the time of their evaluation of glyphosate, it might well have changed their overall conclusions regarding glyphosate's carcinogenic potential.

Dr. Portier's "exact p values" are only "approximate" exact p values because he needlessly (at least for the rare tumors) used a "permutation approach where 20,000 random data sets with the same fixed number of total numbers as the original data set" to estimate the exact p value, whereas it is trivial to calculate the "exact" exact trend test p value for rare tumors.

For example, for the 2001 Kumar study, the kidney tumor incidences were 0/49 – 0/49 – 1/50 – 2/50, and the exact trend test p value is simply

p=[(50 x 49 x 48)/(198 x 197 x 196)]+[(3 x 50 x 49 x 50)/(198 x 197 x 196)]

or p=0.063. Dr. Portier's simulated "approximate" exact test p value for this tumor trend is p=0.059.

For the 1997 Sugimoto study, the kidney tumor incidence was 0/50 – 0/50 – 0/50 – 2/50, and Dr. Portier states that "it is not possible with only 2 tumors to get a p value smaller than the p=0.059 value with the exact test," even though he had previously reported this trend as being highly significant (p=0.008). However, for this pattern of tumor incidence the exact test gives a p value of p=(50 x 49) / (200 x 199) or p=0.062, not p=0.059, and Dr. Portier incorrectly reports this p value to be p=0.061 in any case.

The admission by Dr. Portier that his approximate trend test can greatly exaggerate the statistical significance of the trend is the only important "new" result in the exchange between Drs. Portier and Tarone. Obviously, I support Dr. Tarone's position in this matter.

Nothing in Dr. Portier's latest memo addresses the many other major flaws in his statistical analysis that I laid out in the body of this report, most importantly his miscalculation of meaningful poly-3 tumor rates and his fatally flawed pooled analysis over studies that inappropriately pools tumor rates and exaggerates the significance of the overall effect. In fact, Dr. Portier's latest calculations actually support my conclusion that the "significant" tumor increases observed in the glyphosate rat and mouse studies are all due to chance, not to glyphosate.

Document Nine

Additional Comments of Christopher J. Portier, PhD.
USEPA (EPA-HQ-OPP-2016-0385-0094)
Glyphosate Issue Paper: Evaluation of Carcinogenic Potential
December 12, 2016

Disclaimer: This work was done with my own resources and on my own time. I have received no reimbursement for any of these comments and no other party has contributed to the drafting of these comments. These comments are solely my opinion and my responsibility.

Dr. Joseph Haseman provided comments to the SAP critical of my analyses. I wish to respond.

Major Points:

1. Dr. Haseman criticizes my use of the approximate p-value for the trend test. In my response to Dr. Robert Tarone, I have addressed this issue in male mice by providing exact p-values (calculated using a randomization test) for all of the tumors observed as well as provided an analysis of the trends formally utilizing the historical controls cited by the EPA.

2. Dr. Haseman's analysis of p-values across the fifteen studies is flawed because (1) some of these studies are not valid tests of the compound as noted by the EFSA, EPA and others (6 valid rat studies, 5 valid mouse studies); (2) The approach does not account for rare tumors and (3) the approach does not address the issue of the same tumor site appearing in multiple studies in the male mouse. For example, using his methods for mice alone, you would (1) expect 3.8 positive findings (15.5 tumors/sites x 5 studies x 0.05) and you get 3 using the exact test and 7 using the historical control evaluation; with a significance of 0.002 or less, you would expect 0.115 positive findings and you get 1 from EPA and 1 in the historical control analysis (males only) while for a p-value below 0.02, you expect 1.15 positive findings and get 4 by the exact test and 6 by the historical control test (only males, see spreadsheet from my response to Dr. Tarone); (3) for the pooled analysis, you would expect 25*0.05=1.25 significant at p=0.05 and my analysis shows 3. Thus, if you ask the question, is glyphosate positive in mice, the answer is yes, whereas the answer in rats is probably no.

3. Dr. Haseman's dismissal of the tumor findings in mice fails to clarify the picture. For tumors with low historical control rates, the exact test has very limited power and it is where historical controls should be used to calculate the p-value for the response. For example, the p-value of 0.062 calculated by Dr. Haseman and approximated by me as 0.059 for the 2 renal tumors in the high dose of the Kumar et al. study is the smallest p-value possible under the exact test as this is the most extreme case for a marginal of two tumors (a condition of the exact test); using historical control data yields a p-value of 0.011. The historical control data set I used here was that cited by EPA. The IARC used different historical controls but for simplicity in my presentation, I used the historical controls cited by EPA. For this rare tumor, the p-value calculated by the historical controls is $p<0.01$ in 3 studies. This is even more obvious for hemangiosarcomas where the historical control base cited by the EPA shows no tumors in 26 control groups for 18 months yet the Sugimoto study saw 2 tumors in the high dose group; using combined 18 month and 24 month historical control values, I still get a p-value of 0.021. If nothing else, the SAP should request that EPA use a formal test of significance of historical controls in these evaluations.

4. The EPA draft document compares the results from the 18 month mouse studies with those of the 24 month studies and concludes they are inconsistent. The analysis presented with the poly-3 adjusted pooled data is an attempt to address this question rigorously; if the studies were indeed inconsistent, then any trend in the data should be removed by the pooled analysis. Dr. Haseman's comments fail to address these arguments by the EPA and provide no alternative method to assess the question of consistency. I believe this analysis is appropriate.

Minor Points:

1. The Poly-3 adjustments are used appropriately. EPA based their analysis on total number of animals under study and did not provide the actual survival for the animals. The analysis was simply an attempt to make the 18-month studies match the 24-month studies. There was no attempt to hide the fact that the 24-month study denominators would not change. This is a minor point since studies with and without the Poly-3 adjustment gave effectively the same results.

2. The randomization test (20,00 random samples) I used to estimate the exact p-value for the trend test is sufficient to give reasonable p-values. While it is simple to calculate p-values for responses at the extreme (e.g. only 2 tumors in the highest dose group), this is much more difficult for the pooled data and I did not have access to a program to calculate truly exact p-values. The randomization test was applied to all of my exact analyses.

3. Dr. Haseman is not actually reviewing the EPA evaluation but focuses only on my analyses. Both he and Dr. Tarone correctly point out the flaws in using the approximate trend test p-value. What he fails to see are the limitations of any of these tests for rare tumors and that using historical control data, the last 2 columns in the spreadsheet for my response to Dr. Tarone, shows these limitations. He does not address any of the additional concerns I have about missed data, inappropriate use of historical control data, inappropriate exclusion of the high-dose data, etc. in my original comments on the EPA evaluation.

4. I did not play a significant role in the IARC review; I was an invited specialist. In that capacity, I did not write any of the report or participate in the final discussion to list glyphosate as a "Probable Human Carcinogen". The positive findings for renal tubule adenomas (0/49, 0/49, 1/50, 3/50) in the Knezevich and Hogan study used by the IARC Working Group was based upon the approximate p-value and historical control data but is also significant under the exact test (EPA combines adenomas and carcinomas which resulted in 1 tumor in the control group and an exact p-value of 0.062 compared to the IARC Working Group p-value for adenomas alone of 0.02 against the concurrent control and <0.001 against the historical controls). The finding of a significant effect on hemangiosarcomas in the Atkinson et al. study is significant by the exact test ($p=0.004$) and the test against historical controls ($p<0.001$). The IARC could only evaluate 2 of the mouse studies since there was insufficient documentation available for the remaining studies.