July 7, 2008

Dr. Ruth M. Lunn
NEIHS
Research Triangle Park, NC

Delivered electronically: lunn@niehs.nih.gov

Re: Comments on the RoC draft background document on styrene

Dear Dr. Lunn,

In response to a May 20, 2008 Federal Register notice (73 FR 29139), the American Composites Manufacturers Association is pleased to provide the attached comments on the draft background document for the National Toxicology Program Reports on Carcinogen expert panel on styrene, scheduled to meet July 21-22, 2008.

NTP's draft background document for styrene reviews the styrene epidemiology literature in Chapter 3. NTP's document describes several key studies, primarily of 12 cohorts, and their strengths and weaknesses. Some mention is made of null or negative findings, but NTP's document tends to focus on the positive findings, both statistically significant and non-significant, which results in an emphasis on these findings over findings of no effect. In addition, NTP's document presents the data in such a way that it is difficult to compare risk estimates across exposure categories and studies.

Although NTP's document is a background document and is not by itself intended to provide interpretations or conclusions, in order to support a sound process of interpretation the available studies should be laid out objectively, with the appropriate information and analyses. In ACMA's comments, we discuss the NTP document's description and analysis of individual styrene occupational epidemiology studies and cohorts, as well as the strengths and limitations of each and their bearing on a weight-of-evidence analysis of the question as to whether styrene should be considered carcinogenic. Finally, ACMA's comments discuss the styrene epidemiology literature in the context of the Bradford Hill Criteria.

Sincerely,

John Schweitzer
Senior Director of Government Affairs
1010 N. Glebe Road, Suite 450
Arlington VA 22201
(734) 622-0162
jschweitzer@acmanet.org

Comments on the Epidemiology Data Reviewed in the May 22, 2008 NTP Report on Carcinogens Draft Background Document for Styrene

Prepared for
John Schweitzer
Senior Director of Government Affairs
American Composites Manufacturers Association
1010 N. Glebe Road, Suite 450
Arlington, VA 22201

Prepared by
Julie E. Goodman, PhD, DABT
Gradient Corporation
20 University Road
Cambridge, MA 02138

July 3, 2008

Table of Contents

Page

1 Introduction 1

2 Review of Individual Epidemiology Studies 2
- 2.1 Included and Excluded Studies 2
- 2.2 Presentation of Cohorts 4
- 2.3 Presentation of Results 5
- 2.4 Study Issues 7
  - 2.4.1 Healthy Worker Effect 7
  - 2.4.2 Exposure Metrics 8
  - 2.4.3 Exposure Misclassification 10
  - 2.4.4 Confounders 12
  - 2.4.5 Statistical Analyses 12
  - 2.4.6 Consistency of Results 14

3 Bradford Hill Criteria 16
- 3.1 Strength of Association 16
- 3.2 Consistency 16
- 3.3 Specificity 17
- 3.4 Temporality 17
- 3.5 Exposure-Response 17
- 3.6 Plausibility 18
- 3.7 Coherence 19

4 Conclusion 20

References 21

List of Tables

Table 1 Cohorts Included in Quantitative Analyses in the NTP Report on Carcinogens Draft Background Document for Styrene

List of Figures

Figure 1 The Reinforced Plastics and Composites Industry. Adapted from Cohen *et al.* (2002)
Figure 2 Major Epidemiologic Studies of the SBR Industry. Adapted from Cohen *et al.* (2002)
Figure 3 Major Epidemiologic Studies of the Styrene Monomer and Polymer Industry
Figure 4 Major Epidemiologic Studies of Styrene: Other
Figure 5 An Example of Multiple Comparisons in a Styrene Study

1 Introduction

The 2008 National Toxicology Program (NTP) Report on Carcinogens: Draft Background Document for Styrene (Draft Document) reviews the styrene epidemiology literature in Chapter 3. NTP describes several key studies, primarily of 12 cohorts, and their strengths and weaknesses. Some mention is made of null or negative findings, but NTP tends to focus on the positive findings (risk estimates > 1), both statistically significant and non-significant, which results in an emphasis on these findings over findings of no effect. In addition, NTP presents the data in such a way that it is difficult to compare risk estimates across exposure categories and studies.

Although the NTP report is a background document and is not by itself intended to provide interpretations or conclusions, in order to support a sound process of interpretation, the available studies should be laid out objectively, with the appropriate information and analyses. Below we discuss NTP's description and analysis of individual styrene occupational epidemiology studies and cohorts, as well as the strengths and limitations of each and their bearing on a weight-of-evidence analysis of the question as to whether styrene should be considered carcinogenic. Finally, we discuss the styrene epidemiology literature in the context of the Bradford Hill Criteria.

2 Review of Individual Epidemiology Studies

NTP aims at providing a comprehensive review of the epidemiology literature on the carcinogenicity of styrene, discussing the strengths and limitations of individual studies and occupational cohorts. Below we discuss the studies considered by NTP and NTP's presentation of cohorts and study results, examining whether they are laid out objectively with the appropriate information and analyses presented (with selected examples shown for illustration). We then describe additional facets of individual studies and cohorts that should be considered in a weight-of-evidence analysis that addresses the carcinogenicity of styrene in humans.

2.1 Included and Excluded Studies

NTP reviews the styrene epidemiology studies that were previously reviewed by IARC (1994; 2002) and Cohen *et al.* (2002), as well as several epidemiology studies published since 2002 (although they did not explicitly describe their literature search strategy). The majority of these studies are of 12 cohorts and all of the studies are discussed in some level of detail in their Tables 3-1 to 3-8 and/or in the text. The studies of reinforced plastics and composites (RPC), styrene-butadiene latex rubber (SBR) and styrene/polystyrene (PS) industries are shown here in Figures 1 to 3, which graphically trace the history of the examination of particular cohorts in studies over time. Figure 4 describes two studies of other cohorts: an occupational cohort in Finland reporting urinary concentrations of a styrene metabolite (Anttila *et al.*, 1998) and a cohort of students who attended high school adjacent to facilities that produced synthetic styrene-butadiene (Loughlin *et al.*, 1999).

All of these studies are described in a qualitative assessment of the data, but the majority is excluded in quantitative analyses described in Tables 3-6, 3-7, and 3-8 of the Draft Document. Our Figures 1 to 4 show the studies included in the quantitative analyses in bold-bordered boxes. It is evident from these figures that most of those excluded are earlier studies of a cohort later analyzed in a subsequent included study. In some cases, included studies are a follow-up of the same population in an earlier excluded study. For example, the Danish cohort studied by Kolstad *et al.* (1993) was also analyzed in the Kolstad *et al.* (1994; 1995) studies. In other cases, a cohort is combined with other cohorts for a study with larger power – the Coggon *et al.* (1987) study of a United Kingdom population is excluded because that cohort is included in the Kogevinas *et al.* (1993; 1994) studies. While it is appropriate to exclude these studies in quantitative analyses to ensure cohorts aren't given more weight because they are represented in more than one study, it is still critical that one qualitatively analyze *all*

studies of a cohort and determine whether the results are consistent. If they are not, reasons why should be explored (as discussed in Section 2.3).

NTP also excludes studies that "did not tabulate results for the major cancer sites but focused on exposure response for leukemia and other lymphohaematopoietic cancers (Delzell *et al.*, 1996; 2001; Macaluso *et al.*, 1996; Graff *et al.*, 2005)." Some of these studies, however, have additional information which is not included in other studies of the same cohort and which should have been included in the analyses (Alder *et al.*, 2006). For example, the studies by Sathiakumar *et al.* (2005) and Graff *et al.* (2005) review the same rubber industry workers, but Sathiakumar *et al.* (2005) measured exposure by period of follow-up, employment duration, job classification, and time since first hire, while Graff *et al.* (2005) conducted analyses using cumulative exposure and annual number of exposure peaks > 50 ppm as exposure metrics. Graff *et al.* (2005) also adjusted certain analyses for exposure to 1,3-butadiene and dimethyldithiocarbamate (DMDTC), while Sathiakumar *et al.* (2005) did not. NTP discusses both studies qualitatively, but excludes the Graff *et al.* (2005) study from the quantitative analysis. As discussed in more detail in Section 2.3.3, an evaluation of a cohort should consider together the consistency among each analysis of an endpoint. In this case, while Sathiakumar *et al.* (2005) reported statistically significant associations in some analyses, Graff *et al.* (2005) found no consistent exposure-response trend with all leukemia, chronic myelogenous leukemia, or chronic lymphocytic leukemia, after adjusting for 1,3-butadiene. This inconsistency should have been explored, particularly because Graff *et al.* (2005) arguably describe exposure more completely. Further, other studies of the same cohort that were also excluded estimated exposure (Meinhardt *et al.*, 1982; Matanoski *et al.*, 1997; Macaluso *et al.*, 1996) and consideration of these could have provided valuable information.

One cohort that is not excluded in Table 3-6 of the Draft Document was that comprising over 50,000 Danish men studied by Kolstad *et al.* (1994; 1995). Subjects considered to be "highly exposed" (15,863 individuals) were also included in the Kogevinas *et al.* (1993; 1994) studies.[1] In Tables 3-7 and 3-8, NTP reports SMRs from the Kogevinas data set calculated with country-specific data in the IARC (1994) report, excluding the Danish cohort. Although this prevented individuals from being double-counted in the pooled estimates, it is not accounted for in the text or other tables, in which risk estimates are based on all subjects in the Kogevinas *et al.* (1993; 1994) cohorts. As stated by IARC (1994), in describing the Kogevinas study, "The increase in mortality from neoplasms of the lymphatic and

[1] As discussed below, although Kolstad *et al*. (1994, 1995) describe subjects as having "high" and "low" exposures, they are actually referring to subgroups with high and low *probabilities* of exposure. Exposures of laminators in both groups were equal, but the probability of being a laminator was higher in the "high" group.

hematopoietic tissues observed in the international cohort partly reflects the findings in the Danish cohort." Thus, risk estimates wholly or partially based on over 15,000 individuals, who comprise a large percentage of the total number of people studied in the entire Draft Document and about one-third of the subjects in the Kogevinas (1993; 1994) cohort, are essentially given twice as much weight in NTP's qualitative analyses.

2.2 Presentation of Cohorts

NTP reviews each cohort in the RPC, SBR, and PS industries separately, as shown here in Figures 1 to 3 (other cohorts shown in Figure 4), because significant differences exist in exposure among industries, with RPC clearly having the highest (Cohen *et al.*, 2002). NTP appears to include most, if not all, studies of each cohort, and describes how each cohort was followed, as well as each study's strengths and limitations.

Tables 3-1, 3-2, 3-3, 3-4, and 3-5 in the Draft Document provide a brief description of the study design, subjects, methods, exposure assessment, and results for the studies reviewed by NTP by industry (Table 3-4 includes other cohort studies that do not fit into any of the industry categories, and Table 3-5 includes case-control and ecological studies). Generally, several studies of each cohort are included, and in most cases, the results of the most recent study are provided in the tables. For example, in Table 3-1, Okun *et al.* (1985) and Ruder *et al.* (2004) are listed in the same row, but the subsequent information provided in that row is taken from only Ruder *et al.* (2004) because it is the most recent follow-up. Likewise, in Table 3-3, Ott *et al.* (1980) and Bond *et al.* (1992) are listed in the same row, but the information comes from the most recent study (Bond *et al.*, 1992). In contrast, both the Wong (1990) and the follow-up Wong *et al.* (1994) studies appear separately in Table 3-1. According to the report, separate entries are made for related studies if there were major differences between publications, such as differences in the study design or population composition. In a footnote to Table 3-1, NTP stated that both Wong studies were included "since the excess of respiratory cancer [in the earlier study] was the basis for the nested case-control study [in the earlier study]." NTP is not entirely clear on what constitutes a "major difference." For example, they do not state by how much study populations must vary to be considered major. This would be helpful for the reader, particularly for understanding the consistency, or lack thereof, of risk estimates in a cohort.

It is difficult to get a full picture of the cohorts from Tables 3-1 to 3-5 in the Draft Document. Because of the large number of cohorts and studies of each cohort, it would be helpful for NTP to include

figures (such as our Figures 1 to 4, which were adapted from Cohen *et al.* [2002]) to show how each cohort was followed over time. In addition, a table, such as our Table 1, would show how large each study was, in terms of the number of study subjects, follow-up time, and the period of follow-up. Table 1 also shows how exposures were measured and which confounders and co-exposures were discussed in each study. Although most of this information is in the text and in Tables 3-1 to 3-5 of the Draft Document, it is much easier to compare across studies in Table 1 here. One need only look across a row to determine the information provided in a study, and down a column to determine which studies examined exposure in a certain way. In a similar vein, Table 3-6 of the Draft Document can be used to show which endpoints (*i.e.*, cancer types) were examined in each study. By scattering this information throughout the text, and in some cases incompletely reporting it, the Draft Document hampers the process of weighing individual studies and comparing outcomes across studies.

Our Table 1 currently includes only those studies reviewed in NTP's Table 3-6, which are representative of the 12 major styrene cohorts. This could be expanded with more rows, one for each study, or each row could be expanded to include more information about each cohort (and this information could be provided in accompanying text, if appropriate). In any event, the Draft Document would benefit from a clearer description of the included cohorts.

2.3 Presentation of Results

The presentation of study results in the Draft Document includes valuable information, but it is often difficult to ascertain the overall results for an endpoint in a cohort or an industry. In both the text and Tables 3-1 to 3-5, NTP tends inappropriately to focus on positive outcomes, both statistically significant and non-significant, and sometimes uses terminology that suggest that non-significant positive risk estimates are indicative of an association. There do not appear to be any criteria for when they show non-significant positive effects, and, although NTP sometimes reports negative findings, they do not do so consistently throughout the tables and text. NTP should have a balanced description of positive (> 1) and null and negative (≤ 1) risk estimates for each endpoint, so it is clear to the reader when the positive outcomes, particularly the non-significant ones, are likely representative of causal associations between styrene and cancer risk.

Tables 3-1 to 3-5 present the study design, subjects, methods, exposure assessment, and results for the studies reviewed by NTP (by industry in Tables 3-1 to 3-3, in other cohorts in Table 3-4, and in case-control and ecological studies in Table 3-5). The risk estimates presented in Tables 3-1 to 3-5 are

for the most part limited to significant and non-significant *excess* cancers (morbidity or mortality). For example, in Table 3-1, NTP lists cancers with non-significant excess mortality in the high exposure workers in the Ruder *et al.* (2004) study, but does not list the cancers with non-significant lower mortality. In seeing all the results together, the reader might come to different conclusions than by seeing only the non-significant positive findings.

Risks for each cancer are generally calculated several ways in each study (*e.g.*, by job category, average exposure, cumulative exposure, years of employment). It is not made clear to the reader whether risk estimates that were not discussed in the Draft Document were never calculated or were calculated but not positive and/or statistically significant. (A table such as our Table 1 here could help establish this.) Although in certain cases it may be appropriate to focus on certain study results, they should still be put in the context of other metrics. For example, Kogevinas *et al.* (1994) found lymphohematopoietic cancers increased with time since first exposure ($p_{trend} = 0.012$) and by average levels of exposure ($p_{trend} = 0.019$), but not with cumulative exposure ($p_{trend} = 0.65$). The authors noted, "Workers who had been exposed for less than two years tended to have slightly higher mortality rates for neoplasms of the lymphatic and hematopoietic tissues than longer term workers." Although this is described in the text, it is not described in Table 3-1, nor is it made clear that this analysis was even conducted. In addition, results should be put in the context of those estimated in other studies including the same individuals. Although Kogevinas *et al.* (1994) found this association with lymphohematopoietic cancers, Coggon *et al.* (1987), whose study population made up part of the cohort in the Kogevinas *et al.* (1994) study, found a deficit of deaths from this cause (6 observed, 14.9 expected, statistical significance not discussed). This inconsistency is not discussed in the Draft Document.

The decisions to describe or not to describe specific results should be consistently applied and should follow a stated criterion. Also, results should be described in a clear and consistent manner. If non-significant positive results are shown, than so too should non-significant negative results. If all risk estimates are not shown, then those that are shown should be put in the context of those not described in detail, and also those in other studies of the same cohort. Most of the relevant information from each cohort is described somewhere in the text or the tables of the Draft Document, but the report would benefit from a more consistent reporting of study results.

2.4 Study Issues

NTP discusses several issues with each study, including those addressed by IARC (1994; 2002) and Cohen *et al.* (2002). There is no discussion, however, of the bearing of these issues on the interpretation of study results. These issues include the healthy worker effect, exposure metrics and misclassification, confounders, statistical analyses, and the consistency of results within individual studies. These issues arise in several instances throughout the Draft Document, but we are not going to discuss each time they occur. Rather, we will discuss the overarching issues with a few examples of each, and why it is important to bear these in mind when interpreting each study.

2.4.1 Healthy Worker Effect

NTP notes that a high proportion of all employed styrene workers are excluded from epidemiology analyses because of short durations of employment. NTP suggests this could lead to what is known as the healthy worker effect (HWE), or the selection of occupational cohorts with advantageous health statuses, biasing results towards the null (*i.e.*, weakening any associations between styrene exposure and disease status). According to Li and Sung (1999):

> Many investigators have argued that the HWE is of little or no consequence in interpreting data on cancer mortality. The reason for this is that it is unlikely that factors predicting eventual cancer deaths would be presented at 20 years of age, when many people become employed, which may not be true for factors that predict other causes of death. In other words, most cancers are not associated with a prolonged period of ill-health that would affect employability for a long time before death occurred….
>
> The decline of the HWE with time since first employment may be because the effect of selective exclusion from entry into work only operates during the period when an illness impairs employability. For example, a man who dies from obstructive lung disease may have been too ill to obtain a job for 10 years before his death, but it is less likely to have restricted him from employment 40 years before his death.

Given the long follow-up time for many of the styrene cohorts and that the outcome of interest is cancer, it is unlikely that the HWE will have much bearing on results. Also, most of the styrene studies included short-term workers – the minimum employment duration required for inclusion ranged from one day (Ruder *et al.*, 2004) to five years (Nicholson *et al.*, 1978), mostly between one month and a year.

It is also possible that the inclusion of short-term workers in these studies lead to the opposite of the HWE, *i.e.*, a perceived increase in cancer risk. This is because people who engage in short-term

employment may have lifestyle factors – which will stay with them throughout life – that affect cancer risk. That is, short-term employees, of which there are many in the styrene industries, could have higher cancer risks than long-term workers or the general population. For example, short-term workers tend to smoke more and have lower socioeconomic status than long-term workers (see Wong *et al.*, 1994). Thus, if effects are noted in short term workers, they should be interpreted with that possibility in mind.

2.4.2 Exposure Metrics

Styrene exposures vary greatly within and among industries and over time. The highest exposures occur in the RPC industry, followed by the SBR industry, and then the PS industry (NTP, 2008). In addition, levels of exposure have been decreasing in all three industries over time. For example, Kolstad *et al.* (1994) found that in Denmark, mean styrene levels in the RPC industry were 180 ppm between 1964 and 1970, 88 ppm between 1971 and 1975, and 43 ppm between 1976 and 1988. In Norway, the median of the long-term styrene measurements in the RPC industry from 1992 to 1996 was 7.1 ppm (Lenvik *et al.*, 1999). Current average worker exposure for the SBR and PS industries is estimated to be 5 ppm or less (Miller *et al.*, 1994; Matanoski *et al.*, 1993). Macaluso *et al.* (1996) calculated that time-weighted average (TWA) exposures have declined from 1.8 ppm in the 1940s to 0.1 ppm today in the SBR industry. Exposures within each industry can also vary by job category and employment duration.

Exposure measures for the studies in Table 3-6 of the Draft Document are shown here in Table 1. Most studies did not have individual exposure measurements and used several types of exposure metrics to estimate cancer risks. These metrics included: job category, employment or exposure duration, average (ppm) or cumulative (ppm-years) exposure, employment start date, time since first hire or exposure, and hourly employee (yes/no). As stated by IARC (1994): "The use of alternative exposure models in the analysis provided a means for examining the dependency of results on the various assumptions made when estimating past exposures." All of these exposure estimations, however, are based on many assumptions, some of which may have lead to misclassification (Delzell *et al.*, 2001). According to NTP:

> Classification of workers by individual job titles (McMichael *et al.* 1976a, Ruder *et al.* 2004) or job-exposure matrices (Bond *et al.* 1992, Delzell *et al.* 2001, Kogevinas *et al.* 1994a, Matanoski *et al.* 1997, Santos-Burgoa *et al.* 1992, Seidler *et al.* 2007, Wong *et al.* 1994) may, at least partly, have reduced misclassification of exposure. However, in a validation test within the styrene-butadiene rubber industry, styrene exposure ranks

> correlated poorly with styrene measurements (Matanoski *et al.* 1993), clearly illustrating that it may be difficult to obtain valid exposure estimates in this industry. Macaluso *et al.* (2004) generally found estimates of styrene exposure in the styrene-butadiene rubber industry to be lower than industrial hygiene measurements but did not conduct a thorough validation of their exposure estimates. In the Danish studies of the reinforced plastics industry, duration of employment was abstracted from national pension fund records. Based on a small validation study, the estimates of duration of employment from the national pension fund records did not correlate well with information obtained from a questionnaire from a sub-sample of 671 employees from 8 companies. It was determined that up to 40% of the workers classified as short-term workers by the national pension fund were classified as long-term workers by the questionnaire, while the opposite misclassification occurred among 13% of the workers classified as long-term by the national pension fund (Kolstad *et al.* 1994)...
>
> The study by (Anttila *et al.* 1998) was the only one that relied on individual measurements of exposure; exposure status thus was well documented for these subjects. On the other hand, other studies have shown considerable intra-individual (Symanski *et al.* 2001) and intra-company (Kolstad *et al.* 2005) variability in styrene exposure in the reinforced plastics industry; group-level exposure assessment (Delzell *et al.* 2001, Kogevinas *et al.* 1994a, Macaluso *et al.* 1996, Matanoski *et al.* 1997) may therefore be preferable (Armstrong 1998).

Other limitations not addressed by NTP include a discussion of what the exposure categories mean. For example, Kolstad *et al.* (1994; 1995) defined their low and high exposure groups by the percentage of employees in a company involved in some aspect of reinforced plastic manufacture based on the recollection of two suppliers (who agreed with employers on the classification of 281 of 309 companies, although no analyses were conducted to determine the effect of the disagreement on results). Kolstad *et al.* (1994; 1995) made no attempt to determine any particular individual's exposure, so a person in either exposure group could have had high, low, or no exposure at all. In fact, Kolstad *et al.* (1994) estimated that only ~43% of all employees of companies producing reinforced plastics had any exposure overall (based on the assumption that 25% of employees in the "low exposure" category and 75% of employees in the "high exposure" category were exposed). This could have had a large impact on results in these studies as well as those by Kogevinas *et al.* (1993; 1994), in which the Danish "high exposure" group comprises about one-third of their study population.

In addition, each type of exposure metric differs in its implications. Using a job category as an exposure metric may be informative regarding average exposures, but is not informative regarding cumulative exposures. Within a job category, individual exposures could greatly vary, both in terms of the average exposure and exposure duration. Also, either previous to or subsequent to working at a specific job, some workers could have worked somewhere (either at the same plant or elsewhere) with other exposures.

Cumulative exposure is the same for one person exposed for 1 year to 10 ppm styrene and another person exposed for 10 years to 1 ppm, but the latter person's average exposure is one-tenth of that of the first person (1 *vs*. 10 ppm). An earlier employment start date likely correlates with higher exposures than a later date, but says nothing of exposure duration. Likewise, exposure duration has no correlation with average exposure, as exposure durations of the same length are correlated with much higher exposures in the earlier part of the 20th century verses later. Time since first hire addresses the issue of latency, but also has no bearing on average or cumulative exposure. Ever being an hourly employee was considered because it has been suggested that hourly employees have higher exposures than salaried employees, but this has not been verified (Sathiakumar *et al.*, 2005). If associations are noted with one type of exposure measure and not another, this should be made explicit, and reasons why should be considered in a weight-of-evidence analysis.

When individual exposure measures are not available, the chances of misclassification are high. Although all of the studies reviewed by NTP likely took measures to prevent misclassification of exposure, it could not always be entirely avoided. The effects of exposure misclassification on risk measures are discussed below

2.4.3 Exposure Misclassification

As is generally the case in occupational studies, some exposed subjects were erroneously classified as non-exposed, and some non-exposed subjects were classified as exposed, in the styrene studies. NTP and others have suggested that the exposure misclassification was likely to be non-differential, and therefore would have biased findings towards the null (*i.e.*, if an association existed, it would have appeared to be weaker, or closer to the null value). Non-differential misclassification occurs when, regardless of disease, each exposed and non-exposed subject had the same probability of being misclassified. Some individuals have mistakenly interpreted non-differential misclassification to mean that an equal fraction of subjects are misclassified in the diseased and non-diseased groups. If this indeed were to occur, then risk measures would be biased towards the null. This is because if some percent of the "exposed" study group was actually unexposed and vice versa, then the exposure levels in the two groups would overlap, and actual differences between exposed and unexposed individuals, if they existed,

would appear smaller (Wacholder *et al*., 1995).[2] In fact, in some cases of low sensitivity and specificity, bias beyond the null can occur (Wacholder *et al*., 1995).

By definition, "bias refers to a systematic tendency and not to a particular result" (Wacholder *et al*., 1995). Non-differential misclassification actually means that every subject, regardless of disease status, has an equal chance of being misclassified, but because which subjects are misclassified is a matter of chance, the actual fraction of subjects in a particular study misclassified in the diseased and non-diseased groups is likely to be different. Even if misclassification is non-differential *on average*, due to random variation, misclassification rates in a single study will most likely be differential (Jurek *et al.*, 2005; 2008), and may bias results in any direction. This was demonstrated in a study by Sorahan and Gilthorpe (1994), who presented relative risks from simulated cohort studies with various degrees of non-differential misclassification. This analysis showed that a considerable percentage of studies with non-differential misclassification present produced risk estimates that were larger than those from data sets classified correctly.

According to Wacholder *et al*. (1995), "Several papers published since 1990 have shown that there are special circumstances where there is a bias towards exaggeration of effects. Dosemeci *et al.* identified a scenario where non-differential misclassification of exposure more often than not leads to an overestimate of the odds ratio in an *intermediate* exposure category when there are more than two exposure levels. Other papers that have appeared since the textbooks cited by Sorahan and Gilthorpe' were published during the 1980s, have identified circumstances where an overestimate is more likely than an underestimate. These include particular forms of non-differential misclassification when an exposure is not binary, when grouping has occurred, or when the errors in a continuous exposure are correlated with their true value."

The styrene studies reviewed by NTP each have two or more exposure groups. NTP and others (*e.g.*, Cohen *et al.*, 2002) have suggested that misclassification in these studies was likely to be non-differential, leading to risk estimates that were biased towards the null. In fact, the process of non-differential misclassification could have led to biases away from the null in any of these studies, thus exaggerating risks, if they do exist. In addition, there are other factors that could have affected the risk estimates, including confounders, discussed below.

[2] This is based on the assumption that diseases are properly classified. As discussed by NTP, all but three cohort studies were based on mortality data, which may provide less reliable information about diagnosis and may exclude cases alive at the end of follow-up or dead from another cause.

2.4.4 Confounders

With respect to the styrene epidemiology literature, the confounders most often discussed are 1,3-butadiene and DMDTC in the SBR industry. Several studies acknowledge them but are unable to adjust for them in statistical analyses owing to the way in which exposure is defined (*e.g.*, Sathiakumar *et al.*, 2005). Other studies, however, have adjusted for these factors (*e.g.*, Graff *et al.*, 2005). For example, Graff *et al.* (2005) found that after controlling for 1,3-butadiene, styrene did not have a consistent exposure-response trend with all leukemia, chronic myelogenous leukemia, or chronic lymphocytic leukemia.

It is generally accepted that people working in the RPC industry have the highest exposures to styrene and, as stated by NTP, "unlike the two other industries, is characterized by exposure to few other suspected carcinogens (Jensen *et al.* 1990)." There are other factors, however, that could confound the association between styrene exposure and cancer risk. For example, life-style factors have been suggested to a play role in the differences between risk factors in short- and long-term workers (Kolstad and Olsen, 1999). Few studies have information on smoking, body mass index, or alcohol consumption, all of which are risk factors for cancer (NCI, 1996). Employment duration varied considerably among studies, and it is likely that some short-term workers also worked in other jobs with other chemical exposures.[3] Kolstad *et al.* (1995) suggested confounding by social class can also be an issue, "as a high proportion of the study population were unskilled workers known to experience an excess mortality." Wong *et al.* (1994) concurred, stating, "The most likely explanations for these increases [in mortality] were low socioeconomic class, smoking, and lifestyle factors characteristic of short term workers." These investigators also cited tobacco, alcohol, and diet as the most important risk factors for esophageal cancer. Although NTP acknowledges these confounders, they do not discuss their bearing on the weight-of-evidence analysis.

2.4.5 Statistical Analyses

Most studies used standard statistics to calculate risk estimates, and the limitations of these analyses have been noted elsewhere. Because of their impact on the weight-of-evidence analysis below, a

[3] The impacts of the confounders associated with short-term exposure could be quite large. For example, in the European cohort, the proportion of short-term workers varied from as low as 9% in Finland to as high as 81% in Denmark (Kogevinas *et al.*, 1994).

few limitations bear mentioning here. These include issues with small numbers of observed and expected cases, multiple comparisons, and the analysis of multiple SIRs and SMRs.

Although most of the styrene epidemiology studies follow from hundreds to tens of thousand individuals, the observed and expected numbers of cancer incidence and mortality are generally quite small in each study. As a result, the risk estimates are often not stable (*e.g.*, see Bond *et al.*, 1992). For example, NTP stated, "The biomonitored workers (Anttila *et al.* 1998) showed a 3-fold increased risk of pancreatic cancer (SIR = 3.64, 95% CI = 0.75 to 10.6)." While it is true that the risk estimate is over 3, it is non-significant and highly unstable, as evidenced by the wide confidence interval. It is based on 3 observed and 0.8 expected cases.[4] This SIR is simply too unstable to support or refute an association. To suggest a "3-fold increased risk" is not an appropriate representation of the data. The number of subjects and the stability of each risk estimate always should be considered when interpreting results.

In almost every study considered by NTP in the Draft Document, dozens to hundreds of statistical analyses are conducted. As most studies consider a p-value of 0.05 to be the cutoff for statistical significance, even if exposure had no effect on outcome, 5 out of 100 analyses would be expected to result in statistically significant findings due to chance alone. In addition, if a dataset is broken down into many analyses of subsets of categories, even when certain tests aren't independent of one another, there is the possibility that some of these comparisons, many of which are susceptible to fluctuation owing to low sample size, are likely to result in statistically significant findings simply by chance. For example, Table 3 in the Kogevinas *et al.* (1994) study (Figure 5 here) lists 74 SMRs and their 95% CIs, each representing an analysis of a particular subset or categorization of the subjects in a study. Of these, six are statistically significant:

- all neoplasms (SMR = 91, 95% CI = 83-99)
- all neoplasms, < 10 years since first exposure (SMR = 84, 95% CI = 72-97)
- lymphatic and hematopoietic cancers, < 2 years exposure, 10-19 years since first exposure (SMR = 183, 95% CI = 112-283)
- lymphatic and hematopoietic cancers, < 2 years exposure, < 10 years since first exposure (SMR = 43, 95% CI = 16-93)
- non-Hodgkin's lymphoma (SMR = 0, 95% CI = 0-99); and

[4] It should also be noted that there were 0 observed and 1.0 expected case among individuals whose measurement was taken < 10 years before (SIR = 0.00, 95% CI = 0.00-3.76) and the overall SIR was 1.66 (95 % CI = 0.34-4.85). Neither of these is discussed by NTP, but both should have been considered in a discussion of the findings in the 10+ group.

- leukemia, < 2 years exposure, 10-19 years since first exposure (SMR = 215, 95% CI = 103-395).

There is no exposure-response relationship for either time since first exposure (< 10, 10-19, ≥ 20 years) or exposure duration (< 2, ≥ 2 years) for any cancer type, and there are no associations with other lymphohematopoietic cancers in this table. Also, four of six of these SMRs are < 100, and one would not conclude that this implies styrene was protective of cancer under these conditions. Given these facts and the null results for the 68 other risk estimates, these six significant findings are most likely due to chance. To determine if a statistically significant finding is representative of a true effect, one must determine whether the particular effect has consistency and interpretability within the study and is consistent with findings across other studies, as well. If only a few of many risk estimates are statistically significant and there is no consistency among them, the possibility that the results are due to chance needs to be considered in a weight-of-evidence analysis.

To determine whether an exposure-response relationship exists, one must compare risks across exposure groups. This is most appropriate when using risk estimates based on internal comparisons (*e.g.*, relative risks or odds ratios) or risk estimates based on populations with similar age distributions. The age distributions used in calculations of SMRs and SIRs, which are the risk estimates calculated in the majority of styrene studies, are always based on the population (or sub-population) under study (McElvenny *et al.,* 2004). Thus, it is not entirely appropriate to compare SMRs and SIRs within and across studies, unless one assumes that the age distribution among the populations and sub-populations under study are similar. Nonetheless, if SMRs or SIRs are the only information available, assessing them in exposure-response relationships can be a critical analysis for determining whether a causal association exists. One must just be aware of these caveats and interpret the results of these types of analyses with care.[5]

2.4.6 Consistency of Results

In most styrene epidemiology studies, risk estimates for the same cancer endpoint were calculated a number of ways. They were calculated using different types of exposure metrics, and within each metric, at several exposure levels (Table 1). To properly interpret the results of a study, one must determine whether the risk estimates from *all* analyses are consistent, and, if not, determine what factors were likely to contribute to the inconsistency. In the Draft Document, single results are often reported

[5] This line of reasoning also applies for comparing SMRs and SIRs under other circumstances, such as across studies.

without putting them in the context of other risk estimates for that endpoint. Including the positive findings, while excluding the null and negative findings, from the tables and/or the discussion might make an association appear that doesn't in fact exist.

If an exposure measure is divided into ordinal categories and an association is observed at a middle category but not a higher one, this is not consistent with causation. If this is the case, it should be discussed. For example, Wong *et al.* (1994) found statistically significant increases in cancers of the bronchus, trachea, and lung in individuals exposed for < 1 year ($p < 0.01$) and between 2 and 4.9 years ($p < 0.01$), but not in subjects exposed from 1-1.9 years ($p > 0.05$), 5-9.9 years ($p > 0.05$), or ≥ 10 years ($p > 0.05$). Thus, these results are not indicative of an association between duration of styrene exposure and cancer of the bronchus, trachea, and lung.

It should be noted that if an association is noted with one type of exposure measure, but not another, it is not necessarily inconsistent *per se*. For example, if an association is seen with average exposure but not cumulative exposure, it might mean that a threshold must be reached at some point in time, and the amount of time spent at an exposure level is not as important. Kogevinas *et al.* (1994) found that lymphohematopoietic cancers were increased by time since first exposure, but not by cumulative exposure or duration of exposure. This could be interpreted as meaning a threshold must be reached and a period of time must elapse for styrene to exert its carcinogenic effects, or, on the other hand, it is possible that the statistically significant results are due to chance. When this occurs, it is crucial to determine explanations why to understand the association between styrene and cancer risk.

Overall, if statistically significant effects are seen with exposure to styrene, one must look for consistency of such effects within a study. If a biologically plausible explanation is suggested for an effect that was not expected, one must recognize the *ad hoc* nature of such an explanation (*i.e.*, that it was invoked to account for an inconsistency after it was found). These data can provide a basis to hypothesize about the kind of exposure necessary for an effect to occur, but the hypothesis of such an effect must be tested in other studies to confirm or refute it.

3 Bradford Hill Criteria

The postulates or criteria proposed by Sir Austin Bradford Hill in 1965 are often considered when one is trying to determine whether a chemical exposure is associated with a particular disease in humans (Hill, 1965). These postulates include strength of association, consistency, specificity, temporality, exposure-response, plausibility, and coherence. NTP suggests that "the most consistent findings were for increases in lymphohematopoietic malignancies, and pancreatic cancer." Thus, we address the question as to whether styrene exposure can lead to an increased risk of these malignancies based on the Bradford Hill Criteria. It should be noted that in order to assess these criteria, we have critically reviewed all of the data, particularly the individual risk estimates in the most recent studies of each cohort. These data are described in detail in Dr. Lorenz Rhomberg's comments, but are not shown here.

3.1 Strength of Association

There is no instance of a "strong" effect of styrene in any study based on any styrene exposure metric. The overwhelming majority of risk estimates for styrene and pancreatic or any lymphohematopoietic cancer are not statistically significant, and those few that are significant are not markedly large (*i.e.*, most are below 2 or 3). In addition, most analyses were based on a small number of observed cases, which resulted in unstable estimates, vis-à-vis wide confidence intervals that almost always included 1. Those confidence intervals that did not include 1 were generally close to 1.

3.2 Consistency

Unlike many compounds for which there are sparse human data, there are several cohorts for which styrene risk estimates have been calculated based on several exposure metrics. This allows for the determination of whether noted effects are consistent within and among cohorts and studies. As discussed here in Sections 2.1, 2.2, and 2.3, the way in which data are presented in the Draft Document make it difficult to assess consistency within and across studies. This is primarily because the report stresses significant and non-significant positive effects over null and negative effects, leaving the reader with the impression of a high degree of consistency. There are no consistent effects within or among studies and cohorts for any cancer type, including pancreatic and lymphohematopoietic cancers. In addition, significant and non-significant negative associations reported for certain cancer types were often as strong as positive associations reported for others. Just as it is unlikely that these negative associations are

reflective of a protective mechanism for styrene, the few positive associations are unlikely to reflect a causal association.

3.3 Specificity

There are two ways in which one can examine specificity: one can determine whether (1) one disease is specific to an agent or (2) whether one agent is specific to a disease. NTP indicates that styrene is associated with pancreatic and lymphohematopoietic cancers, diseases with very different modes of action, thus violating (1). The second criterion is often difficult to meet because many diseases or symptoms have multiple causes (some which may be unknown). With few exceptions (*e.g.*, asbestos and mesothelioma, vinyl chloride and angiosarcoma), cancer is not associated with one specific factor. Thus, this criterion is not met for styrene.

3.4 Temporality

Temporality is an obvious requirement because a cause must always precede its effect. This is also the least definitive of Hill's criteria because many events occur prior to the appearance of the disease that may have nothing to do with its causation. Although styrene exposure clearly occurred before cancer incidence or mortality in these studies, so too could have exposures to other factors.

3.5 Exposure-Response

If styrene were associated with cancer, then one would expect an exposure-response relationship within studies and among industries. Because workers in the RPC industry have higher exposures than do those in the SBR and PS industries, one would expect stronger associations among RPC workers. This is not in the case for pancreatic or lymphohematopoietic cancers. In addition, within studies, there were very few instances of an increase in cancer risk with an increase in exposure.

For pancreatic cancer, Kogevinas *et al.* (1994) found a near-significant trend ($p = 0.068$) with cumulative exposure in the European cohort, but there was no trend based on cumulative exposure indicated by Wong *et al.* (1994) in a US cohort. There were also no trends of increased pancreatic cancer risks with job/exposure category, employment duration, exposure duration, or time since first hire in the US cohort.

For all lymphohematopoietic cancers combined, in the European cohort, Kogevinas *et al.* (1994) reported a significant trend with time since first exposure (p_{trend} = 0.012) and average exposure (p_{trend} = 0.019), but not with cumulative exposure (p_{trend} = 0.65). In Europeans whose time since first exposure was < 10 years, however, the risk, although non-significant, was < 1, (SMR = 0.6, 95% CI = 0.32-1.03), and the observed trend may be more a product of unusually low risks in the lowest latency group (time since first exposure was < 10 years) rather than elevated risks in the higher latency groups (10-19 and ≥ 20 years since the first exposure). There were also no trends of increased lymphohematopoietic cancer risk with job/exposure category, employment duration, exposure duration, cumulative exposure, or time since first hire in the US cohort (Wong *et al*., 1994).

No consistent trends of increased risk were noted for individual lymphohematopoietic cancers (*e.g.*, non-Hodgkin's lymphoma, Hodgkin's lymphoma, multiple myeloma, and leukemia) either in the Europoean and US RPC cohorts or the US SBR cohort (Kolstad *et al*., 1994, 1995; Kogevinas *et al*., 1993, 1994; Sathiakumar *et al*., 2005; Graff *et al*., 2005). Associations were examined by job/exposure category, employment duration, exposure duration, cumulative exposure, employment start date, time since first hire, time since first exposure, and average exposure.

3.6 Plausibility

For virtually all known carcinogens, a particular organ or set of organs is known to be the primary site of tumor formation (Cole *et al.*, 2003). For example, benzene is known to affect the blood cell forming system, causing acute myeloid leukemia (Snyder and Andrews, 1996). There may be one target site or multiple target sites, and sites may vary depending on the species of animal observed. Within a species, however, for any given carcinogen, the target sites are consistent. Most national and international guidelines for the classification of chemical agents as carcinogens emphasize the importance of site-specific effects (Huff and Haseman, 1991; IARC, 1986).

Huff *et al.* (1991) and Gold and Zeiger (1997) have compiled data from NTP cancer bioassays and from the general scientific literature for 394 and 1298 different chemicals, respectively, and categorized the chemicals by tumor site. These compilations clearly show that each chemical produces tumors at a limited number of sites within each species or strain tested. Using a random subset of 81 NTP studies, Huff and Haseman (1991) found that analysis of site-specificity was a more accurate predictor of carcinogenicity than analysis of the total number of tumors at all sites combined. Of 45 chemicals shown to be carcinogenic by site-specific analyses, less than half (22) showed a significant increase in overall

tumor incidence. These studies provide confirmation of the historical understanding that a carcinogen "elicits cancers located at sites definitely related to the particular carcinogen" (Hueper and Conway, 1964). Because pancreatic and lymphohematopoietic cancers vary to such a great degree, there is little plausibility that styrene is a causal factor for these different cancer types.

3.7 Coherence

To address coherence, one must determine whether all of the known facts related to the case fit together in a consistent manner. Pancreatic and lymphohematopoietic cancers do not have similar biological modes of action, nor is there anything in mechanistic studies of styrene to suggest the pancreas or the lymphohematopoietic system will be targets of this chemical. There is also no concordance in the way of epidemiology evidence to suggest styrene causes cancer. Taken together, the coherence criterion is not met to support a role for styrene in carcinogenesis in humans.

4 Conclusion

In the Draft Document, NTP describes several key epidemiology studies of the potential carcinogenicity of styrene, aiming to provide a comprehensive review of the literature, discussing the strengths and limitations of individual studies and occupational cohorts. NTP presents the data in such a way that it is difficult to compare risk estimates across exposure categories and studies, however, and tends to emphasize both significant and non-significant positive findings over negative findings. Although this is a background document and is not by itself intended to provide interpretations or conclusions, in order to support a sound process of interpretation, the available studies should be laid out objectively, with the appropriate information and analyses.

Many styrene cohorts were analyzed in one or more studies, and although NTP generally references each study, they are not consistent in how they present information and analyses among these studies. In particular, NTP often emphasizes results from an earlier study of a cohort, when results from later studies are available. NTP also describes several issues within individual studies, but does not discuss their bearing on results. These include the healthy worker effect, which NTP says likely biases results toward the null, but this is generally not the case for cancer. Also, most studies did not have individual exposure measurements and used several types of exposure metrics to estimate cancer risks. All of these exposure estimations are based on many assumptions, some of which may have lead to misclassification, which could have biased results towards or away from the null. Confounders, including 1,3-butadiene in the SBR industry or lifestyle factors in all industries, could have also affected risk estimates. Small numbers of observed cases and multiple comparisons within studies may have lead to spurious significant results. Finally, risk estimates that were not consistent within a study, both within an ordinal exposure measure (*e.g.*, an association is observed at a middle category but not a higher one), or among categories (*e.g.*, an association with cumulative, but not average, exposure) do not support a causal association.

NTP suggests that "the most consistent findings were for increases in lymphohematopoietic malignancies, and pancreatic cancer." We addressed the question as to whether styrene exposure can lead to an increased risk of these malignancies based on the Bradford Hill Criteria, taking all of the above factors into consideration. We found that, based on these criteria, the epidemiology data does not support an association between styrene exposure and pancreatic or lymphohematopoietic cancer risk.

References

Alder, N; Fenty, J; Warren, F; Sutton, AJ; Rushton, L; Jones, DR; Abrams, KR. 2006. "Meta-analysis of mortality and cancer incidence among workers in the synthetic rubber-producing industry." *Am. J. Epidemiol.* 164(5):405-420.

Anttila, A; Pukkala, E; Riala, R; Sallmén, M; Hemminki, K. 1998. "Cancer incidence among Finnish workers exposed to aromatic hydrocarbons." *Int. Arch. Occup. Environ. Health* 71:187-193.

Bond, GG; Bodner, KM; Olsen, GW; Cook, RR. 1992. "Mortality among workers engaged in the development or manufacture of styrene-based products - An update." *Scand. J. Work. Environ. Health* 18(3):145-154.

Coggon, D; Osmond, C; Pannett, B; Simmonds, S; Winter, PD; Acheson, ED. 1987. "Mortality of workers exposed to styrene in the manufacture of glass-reinforced plastics." *Scand. J. Work. Environ. Health* 13(2):94-99.

Cohen, JT; Carlson, G; Charnley, G; Coggon, D; Delzell, E; Graham, JD; Greim, H; Krewski, D; Medinsky, M; Monson, R; Paustenbach, D; Petersen, B; Rappaport, S; Rhomberg, L; Ryan, PB; Thompson, K. 2002. "A comprehensive evaluation of the potential health risks associated with occupational and environmental exposure to styrene." *J. Toxicol. Environ. Health B* 5(1-2):1-263.

Cole, P; Trichopoulos, D; Pastides, H; Starr, T; Mandel, JS. 2003. "Dioxin and cancer: A critical review." *Regul. Toxicol. Pharmacol.* 38:378-388.

Delzell, E; Macaluso, M; Sathiakumar, N; Matthews, R. 2001. "Leukemia and exposure to 1,3-butadiene, styrene and dimethyldithiocarbamate among workers in the synthetic rubber industry." *Chem. Biol. Interact.* 135-136:515-534.

Delzell, E; Sathiakumar, N; Hovinga, M; Macaluso, M; Julian, J; Larson, R; Cole, P; Muir, DCF. 1996. "A follow-up study of synthetic rubber workers." *Toxicology* 113:182-189.

Frentzel-Beyme, R; Thiess, AM; Wieland, R. 1978. "Survey of mortality among employees engaged in the manufacture of styrene and polystyrene at the BASF Ludwigshafen works." *Scand. J. Work Environ. Health* 4(Suppl. 2):231-239.

Gold, LS; Zeiger, E (Eds.). 1997. *Handbook of Carcinogenic Potency and Genotoxicity Databases.* CRC Press, Boca Raton, FL, 754p.

Graff, JJ; Sathiakumar, N; Macaluso, M; Maldonado, G; Matthews, R; Delzell, E. 2005. "Chemical exposures in the synthetic rubber industry and lymphohematopoietic cancer mortality." *J. Occup. Environ. Med.* 47(9):916-932.

Heuper, WC; Conway, WD. 1964. "Creosote." Charles C. Thomas (Springfield, IL) In Hueper, WC, and Conway, WD. 1964. "Chemical Carcinogenesis and Cancers." Charles C. Thomas, Springfield, Illinois. p19-39, 606-626.

Hill, AB. 1965. "The environment and disease: Association or causation?" *Proc. R. Soc. Med.* 58:295-300.

Hodgson, JT; Jones, RD. 1985. "Mortality of styrene production, polymerization and processing workers at a site in northwest England." *Scand. J. Work Environ. Health* 11(5):347-352.

Huff, J; Cirvello, J; Haseman, J; Bucher, J. 1991. "Chemical associated with site-specific neoplasia in 1394 long-term carcinogenesis experiments in laboratory rodents." *Environ. Health Perspect.* 93:247-270.

Huff, J; Haseman, J. 1991. "Long-term chemical carcinogenesis experiments for identifying potential human cancer hazards: Collective database of the National Cancer Institute and National Toxicology Program (1976-1991)." *Environ. Health Perspect.* 96:23-31.

International Agency for Research on Cancer (IARC). 1986. "Long-Term and Short-Term Assays for Carcinogens: A Critical Appraisal." IARC Scientific Publication No. 83. 564p.

International Agency for Research on Cancer (IARC). 1994. "IARC Historical Multicentric Cohort Study of Workers Exposed to Styrene. Report of the Epidemiological Study and the Industrial Hygiene Investigation." IARC Internal Report 94/002. 328p.

International Agency for Research on Cancer (IARC). 2002. "IARC Monographs on the Evaluation of Carcinogenic Risks to Humans. Volume 82: Some Traditional Herbal Medicines, Some Mycotoxins, Naphthalene and Styrene: Section on Styrene." World Health Organization (WHO). p437-550.

Jurek, AM; Greenland, S; Maldonado, G. 2008. "How far from non-differential does exposure or disease misclassification have to be to bias measures of association away from the null?" *Int. J. Epidemiol.* 37(2):382-385.

Jurek, AM; Greenland, S; Maldonado, G; Church, TR. 2005. "Proper interpretation of non-differential misclassification effects: Expectations vs observations." *Int. J. Epidemiol.* 34(3):680-687.

Kogevinas, M; Ferro, G; Andersen, A; Bellander, T; Biocca, M; Coggon, D; Gennaro, V; Hutchings, S; Kolstad, H; Lundberg, I; Lynge, E; Partanen, T; Saracci, R. 1994. "Cancer mortality in a historical cohort study of workers exposed to styrene." *Scand. J. Work Environ. Health* 20(4):251-261.

Kogevinas, M; Ferro, G; Saracci, R; Andersen, A; Biocca, M; Coggon, D; Gennaro, V; Hutchings, S; Kolstad, H; Lundberg, I; Lynge, E; Partanen, T. 1993. "Cancer mortality in an international cohort of workers exposed to styrene." *IARC Sci. Publ.* (127):289-300.

Kolstad, H; Lynge, E; Olsen, J. 1993. "Cancer incidence in the Danish reinforced plastics industry." *IARC Sci. Publ.* (127):301-308.

Kolstad, H; Olsen, J. 1999. "Why do short term workers have high mortality?" *Am J Epidemiol* 149(4):347-352.

Kolstad, HA; Juel, K; Olsen, J; Lynge, E. 1995. "Exposure to styrene and chronic health effects: Mortality and incidence of solid cancers in the Danish reinforced plastics industry." *Occup. Environ. Med.* 52(5):320-327.

Kolstad, HA; Lynge, E; Olsen, J; Breum, N. 1994. "Incidence of lymphohematopoietic malignancies among styrene-exposed workers of the reinforced plastics industry." *Scand. J. Work Environ. Health* 20(4):272-278.

Lenvik, K; Osvoll, P; Woldbaek, T. 1999. "Occupational exposure to styrene in Norway, 1972-1996." *Appl. Occup. Environ. Hyg.* 14:165-170.

Li, CY; Sung, FC. 1999. "A review of the healthy worker effect in occupational epidemiology." *Occup. Med.* 49(4):225-229.

Loughlin, JE; Rothman, KJ; Dreyer, NA. 1999. "Lymphatic and haematopoietic cancer mortality in a population attending school adjacent to styrene-butadiene facilities, 1963-1993." *J. Epidemiol. Commun. Health* 53(5):283-287.

Macaluso, M; Larson, R; Delzell, E; Sathiakumar, N; Hovinga, M; Julian, J; Muir, D; Cole, P. 1996. "Leukemia and cumulative exposure to butadiene, styrene and benzene among workers in the synthetic rubber industry." *Toxicology* 113:190-202.

Matanoski, G., Elliott, E., Tao, X., Francis, M., Correa-Villasenor, A., and Santos-Burgoa, C. 1997. "Lymphohematopoietic cancers and butadiene and styrene exposure in synthetic rubber manufacture." *Ann. New York Acad. Sci.*, 837, 157-69.

Matanoski, G; Francis, M; Correa-Villasenor, A; Elliott, E; Santos-Burgoa, C; Schwartz, L. 1993. "Cancer epidemiology among styrene-butadiene rubber workers." *Butadiene and Styrene: Assessment of the Health Hazards*. IARC Scientific Publications No. 127, M. Sorsa, K. Peltonen, H. Vainio, and K. Hemminki, eds., International Agency for Cancer Research, Lyon, France, 363-74.

Matanoski, GM; Santos-Burgoa, C; Schwartz, L. 1990. "Mortality of a cohort of workers in the styrene-butadiene polymer manufacturing industry (1943-1982)." *Environ. Health Perspect.* 86:107-117.

McElvenny, DM; Armstrong, BG; Jarup, L; Higgins, JP. 2004. "Meta-analysis in occupational epidemiology: A review of practice." *Occup. Med.* 54(5):336-344.

McMichael, AJ; Spirtas, R; Gamble, JF; Tousey, PM. 1976. "Mortality among rubber workers: Relationship to specific jobs." *J. Occup. Med.* 18(3):178-185.

Meinhardt, TJ; Lemen, RA; Crandall, MS; Young, RJ. 1982. "Environmental epidemiologic investigation of the styrene-butadiene rubber industry." *Scand. J. Work Environ. Health* 8:250-259.

Miller, RR; Newhook, R; Poole, A. 1994. "Styrene production, use, and human exposure." *Crit Rev Toxicol* 24:S1-S10.

National Cancer Institute. 1996. "Cancer Rates and Risks (4th Edition)." NIH Publication No. 96-691. 205p.

National Toxicology Program (NTP). 2008. "Final Report on Carcinogens Draft Background Document for Styrene." 436p.

Nicholson, WJ; Selikoff, IJ; Seidman, H. 1978. "Mortality experience of styrene-polystyrene polymerization workers. Initial findings." *Scand. J. Work Environ. Health* 4(Suppl. 2):247-252.

Okun, AH; Beaumont, JJ; Meinhardt, TJ; Crandall, MS. 1985. "Mortality patterns among styrene-exposed boatbuilders." *Am. J. Ind. Med.* 8(3):193-205.

Ott, MG; Kolesar, RC; Scharnweber, HC; Schneider, EJ; Venable, JR. 1980. "A mortality survey of employees engaged in the development or manufacture of styrene-based products." *J. Occup. Med.* 22(7):445-460.

Ruder, AM; Ward, EM; Dong, M; Okun, AH; Davis-King, K. 2004. "Mortality patterns among workers exposed to styrene in the reinforced plastic boatbuilding industry: An update." *Am. J. Ind. Med.* 45(2):165-176.

Santos-Burgoa, C; Matanoski, GM; Zeger, S; Schwartz, L. 1992. "Lymphohematopoietic cancer in styrene-butadiene polymerization workers." *Am. J. Epidemiol.* 136(7):843-854.

Sathiakumar, N; Delzell, E; Hovinga, M; Macaluso, M; Julian, JA; Larson, R; Cole, P; Muir, DC. 1998. "Mortality from cancer and other causes of death among synthetic rubber workers." *Occup. Environ. Med.* 55(4):230-235.

Sathiakumar, N; Graff, J; Macaluso, M; Maldonado, G; Matthews, R; Delzell, E. 2005. "An updated study of mortality among North American synthetic rubber industry workers." *Occup. Environ. Med.* 62(12):822-829.

Snyder, R; Andrews, LS. 1996. "Toxic effects of solvents and vapors." In *Casarett and Doull's Toxicology, the Basic Science of Poisons (Fifth Edition).* (Ed: Klaassen, CD), McGraw Hill, New York, p737-771.

Sorahan, T; Gilthorpe, MS. 1994. "Non-differential misclassification of exposure always leads to an underestimate of risk: An incorrect conclusion." *Occup. Environ. Med.* 51(12):839-840.

Wacholder, S; Hartge, P; Lubin, JH; Dosemeci, M. 1995. "Non-differential misclassification and bias towards the null: A clarification [Letter to the editor]." *Occup. Environ. Med.* 52(8):557.

Wong, O. 1990. "A cohort mortality study and a case-control study of workers potentially exposed to styrene in the reinforced plastics and composites industry." *Br. J. Ind. Med.* 47(11):753-762.

Wong, O; Trent, LS; Whorton, MD. 1994. "An updated cohort mortality study of workers exposed to styrene in the reinforced plastics and composites industry." *Occup. Environ. Med.* 51(6):386-396.

Tables

Table 1
Cohorts Included in Quantitative Analyses in the NTP Report on Carcinogens Draft Background Document for Styrene

Reference	Job/Exposure Category	Subjects (n)	Total Follow-up (person-years)	Period of Follow-up	Minimum Employment Duration	Employment Duration (Years)	Exposure Duration (Years)	Average Exposure (ppm)	Cumulative Exposure (ppm-yr)	Employment Start Date	Time Since Hire/First Exposure (Years)	Hourly Employee	Co-Exposures/ Confounders Discussed in Study	Notes
Reinforced Plastic Industry (RP)														
Kolstad *et al.* (1994, 1995) Workers from Denmark.	Exposed: Ever worked in company producing reinforced plastic Low: 1-49% of employees in RP production High: 50-100% of employees in RP production Unexposed: Never worked in company producing reinforced plastic Exposure unknown	50,903	584,556	1970-1989	< 1 ≥ 1	< 1 ≥ 1				1964-1970 1971-1975 1976-1988 ≤ 1970 > 1970	< 10 ≥ 10			Exposure classification based on opinions of two dealers of plastic raw materials. These differed from employers' classifications. Mean styrene levels were 180 ppm (1964-1970), 88 ppm (1971-1975), and 43 ppm (1976-1988).
Kogevinas *et al.* (1993, 1994) Workers from Denmark, Finland, Italy, Norway, Sweden and the United Kingdom	Laminators Unspecified Tasks Other exposed Jobs Unexposed	40,688	539,479	1945-1991 (varies by country)			< 2 ≥ 2	< 60 60-99 100-119 120-199 > 200	< 75 75-199 200-499 ≥ 500		< 10 10-19 ≥ 20		Peroxides Styrene oxide Acetone Methylene chloride Other aromatic hydrocarbons Fibers Dust	Study examines decreasing exposure over time. Study uses part of Danish cohort described in Kolstad *et al.* (1994, 1995).
Wong *et al.* (1994). Workers in the US.	Open mould processing Mixing and closed mould processing Finishing operations Plant supports Maintenance and preparation Supervisory and professional	15,826	307,932	1948-1989	≥ 6 months	< 1 1-1.9 2-4.9 5-9.9 ≥ 10 up to 1977	< 1 1-1.9 2-4.9 5-9.9 ≥ 10 up to 1977		< 10.0 10.0-29.9 30.0-99.9 ≥ 100.0		< 10 10-19 ≥ 20			Job category analysis - all cohort members employed > 2 yr
Ruder *et al.* (2004). Workers at two boatbuilding plants in the U.S.	High exposure: Fiber glass (TWA = 42.5 ppm) or Lamination (TWA = 71.7 ppm) Low exposure: Never worked in high-exposure departments	5,204	135,588	1959-1998	> 1 d	< 1 > 1							Fiberglass Solvents Wood dust Wood finishing agents	
Styrene-Butadiene Rubber Industry (SBR)														
McMichael *et al.* (1976). Male workers at a tire plant in the US (OH).	SBR Plant 15 other work areas	6,678	--	1964-1973	> 10 yr (99%)	>2 >5							Gases or liquids that are ingredients for the particular synthetic rubber being made	
Sathiakumar *et al.* (2005). Male workers at 8 U.S. and Canadian synthetic rubber plants.	Production (polymerization, coagulation, finishing) Maintenance (shop, field) Labor (production, manintence) Laboratories Other	17,924	--	1944-1991 1992-1998 1944-1998	≥ 1yr	< 10 ≥ 10					< 20 20-29 ≥ 30	Ever Never	1,3-Butadiene DMDTC Benzene	Graff *et al.* (2005) examined cumulative exposure and freqeuency of peak exposure > 50 ppm in same cohort
Polystyrene/Styrene Production Industry (PS)														
Frentzel-Beyme *et al.* (1978). Workers at BASF Ludwigshafen, Germany.	All employees engaged in the manufacture of styrene or polystyrene	1,960	20,138	1931-1976	> 1 month									
Bond *et al.* (1992). Male workers at Dow Chemical plants in the US.	Styrene monomer and finishing Styrene-butadiene latex production Product research and development Polymerization, coloring, extrusion All styrene-based products cohort Workers unexposed to styrene Styrene/ethylbenzene only Mixed exposures to styrene, ethyl benzene, benzene, alkylbenzenes, acrylonitrile Extrusion fumes; indrenct colorants; styrene, ethylbenzene, or acrylonitrile Extrusion fumes; drenct colorants; styrene, ethylbenzene, or acrylonitrile Polymer dusts plus styrene/ethylbenzene Several other categories 1-4 ppm and ≥ 5 styrene 8-hr TWA.	2,904	89,825	1937-1986	≥ 1 yr	< 1 1-4 ≥ 5							Ethylbenzene Alkylbenzene compounds Benzene Acrylontrile Polymer dusts Styrene oligomers Mineral Oil Direct colorants Indirect colorants	
Hodgson and Jones (1985). Male workers at a plant in England.	Laboratory and manual workers (styrene production, polymerization, and processing) Manual workers with no specific occupational styrene exposure	622	8,654	1945-1978	≥ 1 yr					1945-1958 1959-1968 1969-1974			Acrylonitrile Pitch Polyvinyl chlorinde fumes Benzene Dyestuffs Antioxidants Polyolefines Ethylene Oxide	Exposure substantially < 100 ppm. Also conducted analyses stratified by age.
Nicholson *et al.* (1978). Male workers at a plant in the US (TX).	Production and polymerization Maintenance Utilities service	560	--	1960-1975	≥ 5 yr						10-19 20-29 ≥ 30		Benzene	Exposures: 5 – 20 ppm or < 1 ppm
Styrene Monitored Workers														
Anttila *et al.* (1998). Male and female workers biologically monitored by the Finnish Institute of Occupational Health.	Workers monitored by Finnish Institute of Occupational Health	2,580	34,288	1973-1983	--						0-9 ≥ 10			Time since measurement of styrene metabolite in urine.
Environmental Exposure														
Loughlin *et al.* (1999). Former students of an Eastern TX high school, located adjacent to styrene mfg facilities.	Students attending high school adjacent to SBR facility	15,403	310,254	1963/4-1992/3	≥ 3 consec. months attendance in a school year	≤ 2 ≥ 3								

Figures

Figure 1. The Reinforced Plastics and Composites Industry. Adapted from Cohen *et al.* (2002). *Studies with bold border included in NTP Draft Document Table 3-6.*

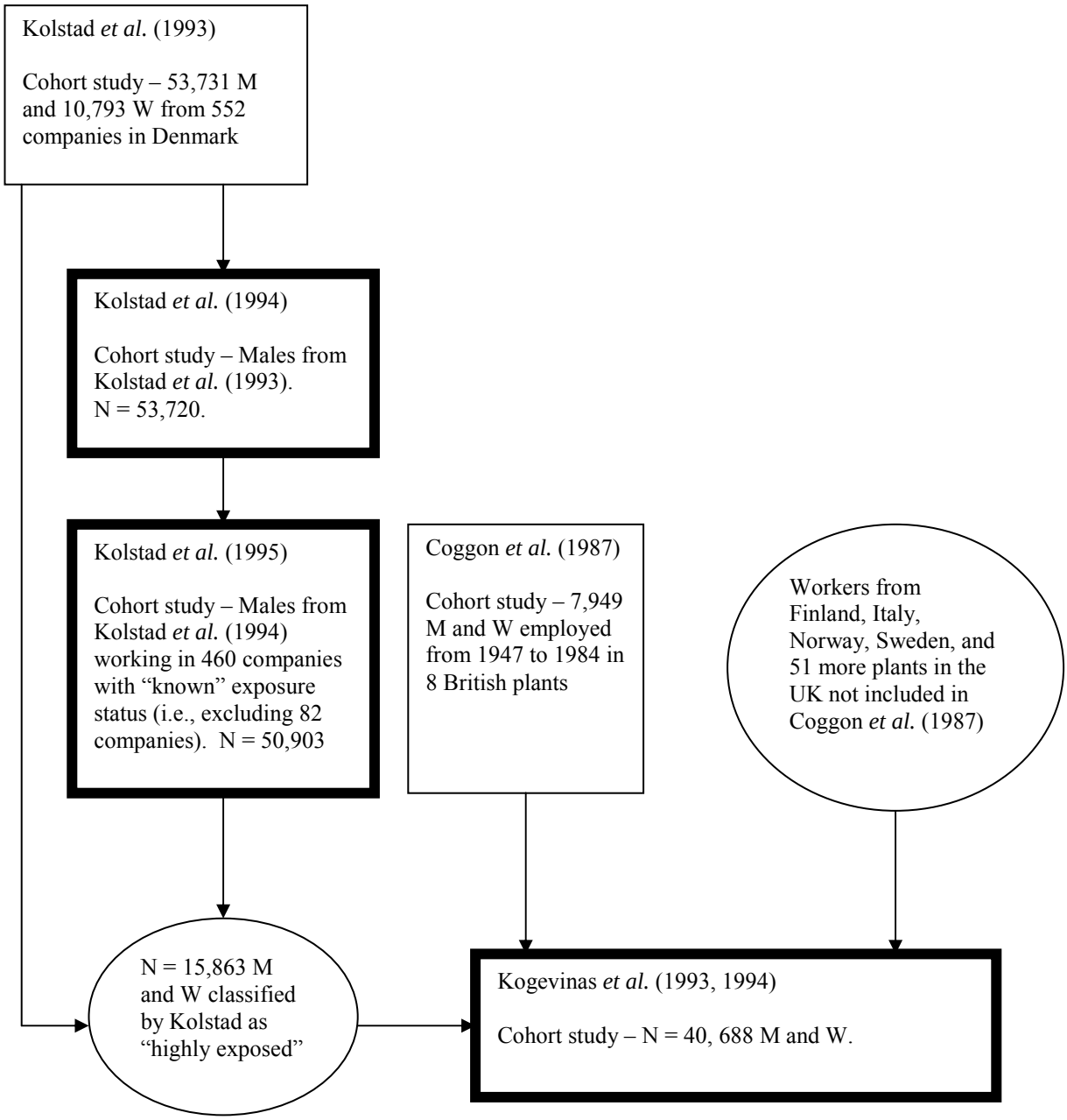

Figure 1 (Continued). The Reinforced Plastics and Composites Industry. Adapted from Cohen *et al*. (2002). *Studies with bold border included in NTP Draft Document Table 3-6.*

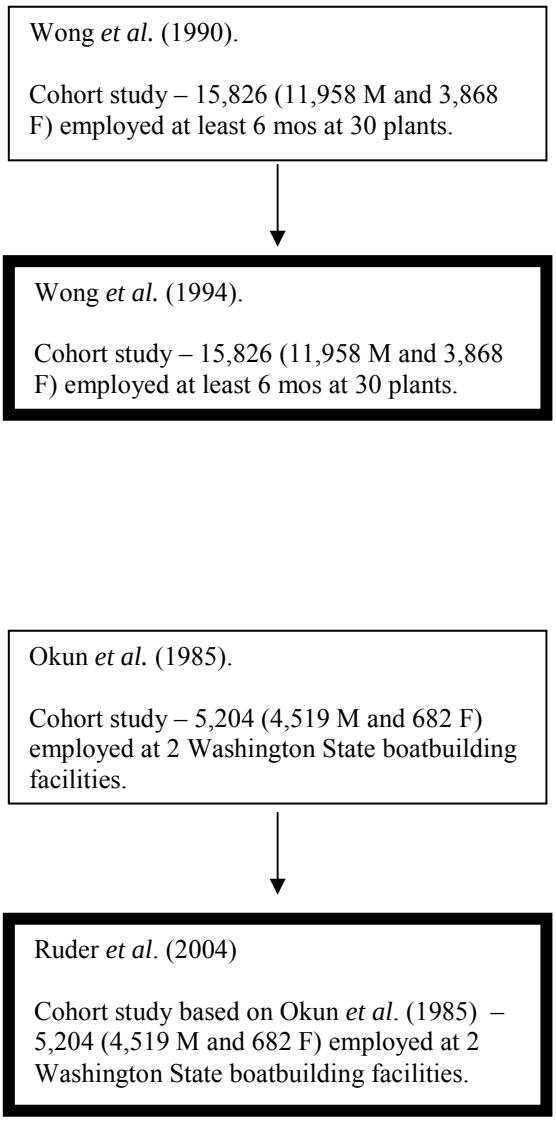

Figure 2. Major Epidemiologic Studies of the SBR Industry. Adapted from Cohen *et al.* (2002). *Studies with bold border included in NTP Draft Document Table 3-6.*

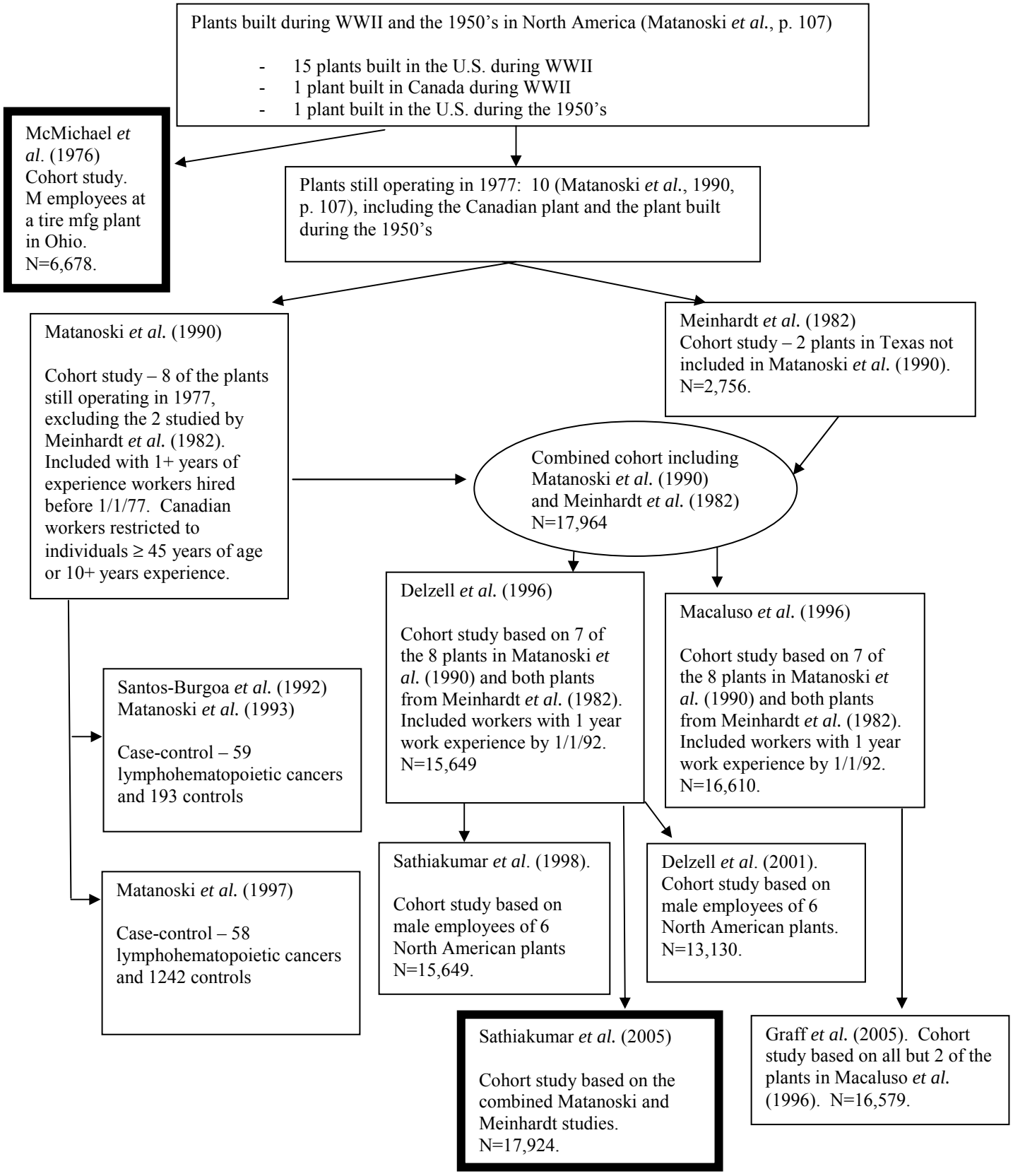

Figure 3. Major Epidemiologic Studies of the Styrene Monomer and Polymer Industry.
Studies with bold border included in NTP Draft Document Table 3-6.

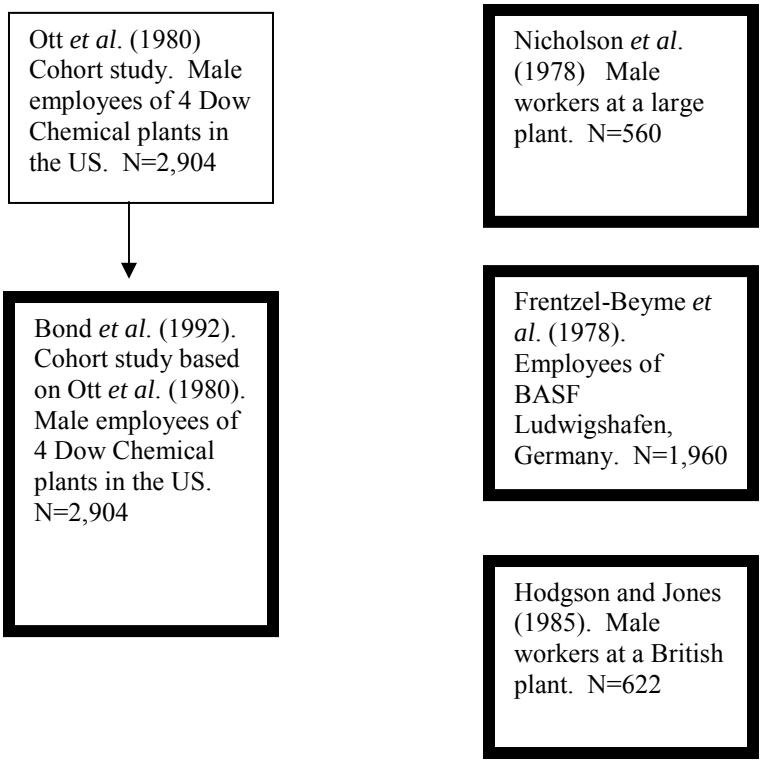

Figure 4. Major Epidemiologic Studies of Styrene: Other. ***Studies with bold border included in NTP Draft Document Table 3-6.***

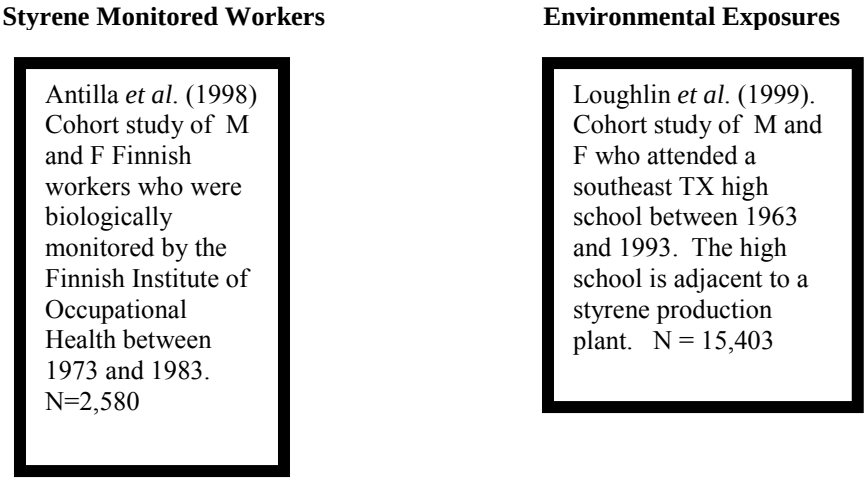

Figure 5. An Example of Multiple Comparions in a Styrene Study. ***Statistically significant findings shown in boxes.***

Table 3. Mortality from all neoplasms and neoplasms of the lymphatic and hematopoietic tissue for workers exposed to styrene by time since first exposure and duration of exposure. (SMR = standardized mortality ratio, 95% CI = 95% confidence interval)

Site*	Time since first exposure											
	<10 years			10–19 years			≥20 years			Total		
	Observed deaths (N)	SMR	95% CI	Observed deaths (N)	SMR	95% CI	Observed deaths (N)	SMR	95% CI	Observed deaths (N)	SMR	95% CI
All neoplasms												
<2 years' exposure	100	78	63–94	143	105	88–123	45	86	63–116	288	91	81–102
≥2 years' exposure	76	94	74–118	113	87	72–105	59	100	76–129	248	92	81–104
Total exposure	176	84	72–97	256	96	85–109	104	93	76–113	536	91	83–99
Lymphatic and hematopoietic (200–208)												
<2 years' exposure	6	43	16–93	20	183	112–283	3	85	18–248	29	102	68–147
≥2 years' exposure	7	92	37–190	6	61	22–132	7	173	70–357	20	93	57–143
Total exposure	13	60	32–103	26	125	82–183	10	132	64–244	49	98	72–130
Non-Hodgkin's lymphoma (200–202)												
<2 years' exposure	—	0	0–99	4	118	32–302	1	83	2–460	5	60	19–140
≥2 years' exposure	3	140	29–408	1	32	1–177	3	221	45–645	7	105	42–217
Total exposure	3	51	11–149	5	76	25–178	4	155	42–397	12	80	41–140
Hodgkin's disease (201)												
<2 years' exposure	2	82	10–295	3	280	58–819	—	0	0–1844	5	134	44–313
≥2 years' exposure	1	83	2–460	—	0	0–419	1	500	13–2786	2	87	11–314
Total exposure	3	82	17–241	3	153	32–447	1	244	6–1359	7	116	47–239
Multiple myeloma (203)												
<2 years' exposure	1	72	2–401	3	172	36–504	1	139	4–774	5	129	42–302
≥2 years' exposure	1	99	3–552	2	108	13–391	—	0	0–405	3	80	16–233
Total exposure	2	83	10–299	5	140	45–326	1	62	2–344	8	105	45–207
Leukemia (204–208)												
<2 years' exposure	3	47	10–137	10	215	103–395	1	73	2–407	14	113	62–189
≥2 years' exposure	2	62	7–222	3	75	16–220	3	194	40–566	8	91	39–179
Total exposure	5	52	17–122	13	150	80–257	4	136	37–347	22	104	65–157

* Code of the International Classification of Diseases, eighth revision, in parentheses.

Source:

Kogevinas, M; Ferro, G; Andersen, A; Bellander, T; Biocca, M; Coggon, D; Gennaro, V; Hutchings, S; Kolstad, H; Lundberg, I; Lynge, E; Partanen, T; Saracci, R. 1994. "Cancer mortality in a historical cohort study of workers exposed to styrene." Scand. J. Work Environ. Health 20(4):251-261.