Comments from Dr. Erik Garshick in letter dated August 11, 1997 to Genevieve Shiroma

Comment 1: Previous communications that I have had with OEHHA have stressed the uncertainties of the shape of the exposure-response relationship in the retrospective cohort data, particularly in the setting of under ascertainment of death between 1977-1980. A letter that I wrote in 1994 is quoted in the ARB/OEHHA responses to public comment, but the letter is not quoted in its entirety. In the second paragraph I noted that the shape of the exposure-response relationship was not as positive as originally reported, with a reference to my 1991 letter to the EPA. I also stressed the need to obtain additional information regarding the mortality experience of this cohort. It was not my intention for my concluding paragraph of that letter to be interpreted as implicit approval for this version or any past version of the OEHHA risk assessment document. Furthermore, in May 1995 I submitted my comments to the U.S.EPA regarding the use of retrospective cohort study for risk assessment. The comments made in that letter mirror my previous and current comments to OEHHA. A copy of this letter was submitted previously to you at the public hearing on July 1. Although any governmental agency must be satisfied with assumptions made regarding assessing risk, there also should be general approval of the scientific community. I do not believe that your current document fully expresses the uncertainty of the estimates of risk that you have presented, nor does the current retrospective cohort data allow the calculation of unit risk with confidence.

Response: *We appreciate your comments and suggestions made to us in the past. It was not our intent to suggest that you were endorsing or had approved our risk assessment. We simply were indicating that in response to suggestions made by you in the past, that a range of risk was developed because it was thought to be more appropriate than a single point estimate. The range of risk can better incorporate the weight of evidence for diesel exhaust, while still indicating the uncertainty in identifying a single estimate.*

OEHHA appreciates the concerns raised in the comment regarding the need to truncate the study in 1976. Clearly, the need to truncate the analysis at 1976 does potentially affect their power to distinguish any trend in response with exposure. First, it reduces the numerical power of the calculations. Secondly, it reduces the duration of the follow-up period for a disease with a long latency. These two limitations increase the likelihood of a false negative result, rather than a false positive result. However, whatever trends were observed through 1976, they do represent the period covered. OEHHA believes the information to be useful for this purpose. The analyses conducted by OEHHA, as well as those conducted by Dr. Crump (1991) do not suggest significant differences when the last four years are included or excluded in the analysis.

OEHHA has also addressed the comment's concern regarding the assignment of past exposures. Qualitatively, we have described the uncertainties as to both the levels of exposure and which workers were actually exposed to diesel exhaust before 1959. Quantitatively, OEHHA provided a large range of risk estimates to characterize uncertainty. We think it is most probable that this range encompasses the uncertainty introduced by the limited exposure information available.

Chapter 1: General Comments

Comment 2: Conclusions Regarding Causal Inference. I agree with the conclusions of the documents written by the World Health Organization, IARC, and the Health Effects Institute regarding the evidence for the carcinogenicity of diesel exhaust in humans. Based on the same data, you go beyond these conclusions in the current draft document The weight of the evidence does suggest that whole diesel exhaust is a human lung carcinogen, however the human studies have limitations. These limitations are mainly due to lack of exposure histories, and a short duration of follow-up (just over 20 years) of exposed workers in the best studies. However, based on the strong likelihood that diesel exhaust may cause lung cancer in humans, and that more additional definitive studies are expensive and time consuming, it still is important to regulate human exposure.

***Response**: Comment noted. We acknowledge your comment that "The weight of the evidence does suggest that whole diesel exhaust is a human lung carcinogen, however the human studies have limitations." OEHHA also maintains that the weight-of-evidence suggests that diesel exhaust is a human lung carcinogen. We also acknowledge that there are limitations to human studies. Our intent is to clearly identify the limitations, while indicating the weight of evidence. The limitations are underscored by the presentation of a broad range of risks..*

Comment 3: Use of Animal Data For Quantitative Risk Assessment. Since the most likely mechanism of lung cancer in rats exposed to diesel exhaust is attributable to particle overload, my opinion is that it is not possible to use the animal data to determine the human risk of lung cancer.

***Response**: With respect to the commenter's and others' concerns regarding the use of the animal data, OEHHA shared them with the Scientific Review Panel on October 16, 1997. There was a thorough discussion of the issue of using the rat data for quantitative human risk assessment at the meeting. The sense of the panel was that the rat data and calculations provide useful information and should be left in the document. However, since human epidemiologic evidence was available on which to base the human risk estimate, the human data should be used to form the range of risks. Therefore, OEHHA now bases the range of unit risk estimates only on the epidemiological information.*

Comment 4: Differing Analyses Of the Railroad Worker Retrospective Cohort. Study. In previous communications to the Office of Environmental Health Hazard Assessment, I have pointed out that there is considerable uncertainty in the slope in the relationship between cumulative exposure to diesel exhaust and lung cancer. Although the younger workers in the retrospective cohort study had the greatest risk of dying of lung cancer, based on our reanalysis of these data, it is not possible to use a positive slope to definitely describe the relationship between cumulative exposure and lung cancer mortality. I believe that the use of a slope as derived in the OEHHA assessment has not been justified. Using years of exposure (months of exposure unweighted for estimated exposure level) starting in 1959, the slope is not positive, and appears flat or negative. The lack of a positive slope between cumulative exposure does not imply the study is negative, but is due 'to weaknesses in exposure assignment, changing exposures over time, and the lack of exposure data pre-1959. In addition, contributing to the uncertainty of the

slope are unrecognized deaths in the years 1977-1980 since relatively few "cells" contribute to the effects of 10-14 years and 15-17 years of exposure as originally presented. When the study is truncated in 1976, important person-years of follow-up are excluded and it is even more difficult to determine the true slope.

***Response**: We agree with the general conclusion regarding the Garshick et al. (1988) cohort study that "The lack of a positive slope between cumulative exposure does not imply the study is negative, but is due to weaknesses in exposure assignment, changing exposures over time, and the lack of exposure data pre-1959." Furthermore, it appears to us that these weaknesses would likely move the resulting slope toward non-significance.*

The comment indicates that the use of a slope, based on cumulative exposure and lung cancer, as derived in the OEHHA risk assessment, has not been justified. In the health document analysis we discussed the many analyses and reanalyses of the cohort data. In calculating the potential human risk we reviewed published calculations as well as deriving our own. To provide a complete perspective, we felt it important to calculate risk from the original published study as well. As indicated in the Garshick (1991) letter, reanalyses (while not published) suggests a less positive slope, therefore a lower risk. Furthermore, in our proposed reconstructions, we assume a greater exposure (to simulate changes in the industry) which also results in a lower risk calculation. Consequently, the risk calculation based on the published cohort data appears to represent a "ceiling" of risk. That is, based on the 1991 reanalyses and our reanalyses, we would expect the actual risk to be below the one directly calculated from the published study. However, to address the issue raised in the comment, the risk estimate derived directly from the published results, without reanalysis, will not be included in the revised range of risk.

Comment 5: Two major differences in the development of the various analyses using the retrospective cohort data to examine lung cancer mortality has been the modeling of age (as noted in Appendix F), and the inclusion of a "background" level of particulate among the unexposed workers. However, in an examination of the analyses presented, it seems that both Dr. Stanley Dawson, the principal author of the California risk assessment, and Dr. Kenny Crump, a principal critic, have used similar methods in adjusting for age despite the arguments offered in Appendix F that different methods are used. The remaining difference therefore is the method used to account for background exposure. The difference between these analyses should be more clearly examined since different assumptions are made by using "exposure" weights for workers without actual diesel exposure.

***Response**: We have spent a considerable effort evaluating the impact of the method for modeling for age. At the time of release of our previous draft report, it appeared that the method of modeling for age did significantly affect the results. The information provided in the comments submitted by Dr. Garshick, in the comments provided by Dr. Crump, and in further internal analyses has allowed us to better understand the influence of this issue on the results reported. First, the influence of method of control for age in these analyses is dependent on the exposure pattern under consideration and the method used to account for background exposure.*

For this reason we provided further discussion of this issue in the revised document. We will summarize these issues briefly here.

When considering the exposure paradigm described in the original study (Garshick et al. 1988), the published results exhibited a clear upward trend. In the Crump et al. (1991) reanalysis of this paradigm, controlling for age by "attained age", the risk did not increase monotonically with 'elapsed time.' The Garshick reanalysis (1991), adding a third age variable, led to lower point estimates of the effects of attributable to diesel exhaust with much less of a suggestion of an exposure-response relationship. While the various methods to control for age may result in different visual trends, it is important to acknowledge the large error bars around each point. These results led us to consider further reanalysis of the data using modified exposure paradigms.

In their report, Crump et al. (1991) also used a ramp exposure pattern. They reported that the dose response slope was negative, but discovery of a programming error in their use of a computer program (Dawson, 1995) resulted in reanalysis of the data. Crump (1995, 1996, 1997) reported the reanalyzed slopes were not statistically significant; however, the analyses did not consider the background exposure of the clerks/signalmen to be zero. When we set the background exposure level to zero using the ramp exposure paradigm, this resulted in statistically significant positive slopes. Thus, this addresses the second point of the comment regarding the importance in the method used to account for background exposure.

Finally, we also used a roof exposure pattern to consider the dieselization and engine improvements over time. We presented results using a variety of controls for age and consistently found the positive slopes were statistically significant. In each case exposures for clerks and signalmen was considered to be zero.

While the slope reported for the various analyses and reanalyses vary somewhat, the predicted risk values remain fairly stable within our range of uncertainty. That is, the results of the cohort study are generally consistent with the results of the case-control study, and with the increased risk reported overall as part of the weight of the evidence regarding diesel exhaust exposure and human lung carcinogenicity.

In response to the comment regarding the need to more clearly examine the use "background" level of particulate among the unexposed workers, we have conducted additional analyses as mentioned above. In the original analyses (Garshick et al. 1988) lung cancer rates of unexposed railroad workers were compared to the rates of exposed workers categorized by the duration of exposure. The unexposed workers (clerks/signalmen) were assumed to have zero exposure to diesel exhaust for the whole study period. Thus, the background exposure is not an influencing factor in the original analyses since the unexposed workers have zero exposure. This is consistent with the work of Woskie et al. (1988a) who reported that the clerks/signalmen exposure was in the background range of the national average and the work of Hammond et al. (1988) who stated that the exposures of the clerks "…almost certainly do not represent diesel exhaust from locomotives…" We also note that the Garshick reanalysis (1991) pointed out that

unless the clerks/signalmen group concentration is considered to be zero, a cumulative exposure for the unexposed group will occur.

However, as noted above, Crump et al. (1991) and Crump (1995, 1996, 1997) performed analyses for the ramp exposure pattern without adjusting the measurements of the unexposed workers to zero. Using this approach, Crump (1996) reported non-significant results for slope estimates. When we performed analyses for the ramp exposure pattern with unexposed workers at zero exposure, we found significantly positive slope estimates. As a specific example, for internal controls on attained age and calendar year, the p-value fell from the marginally significant value of 0.037 to the significant value of 0.006 when the background was set at zero. Consequently, for dose-response slope results reported using the ramp exposure pattern, it is important to consider how the background exposure was incorporated. We also performed analyses with a the roof exposure pattern. In all of the reported roof pattern analyses, background was zero. In conclusion, we agree with the comment regarding the importance of considering the background concentration when conducting these analyses.

Comment 6: Chapter 6: Carcinogenic Effects. Page 6-49, 2nd paragraph: I agree that most human carcinogens have a latency of at least 10 years. However, it would be more complete to state that the latency for most human carcinogens is generally in the range of 20 years or more. A limitation of the epidemiological studies in humans is the lack of studies with many workers with long term exposure that is well characterized (more than 25 to 30 years).

Response: *The statement regarding latency has been modified to address this issue.*

Comment 7: Chapter 7, Section 7.3, Human Risk Estimates From Epidemiological Studies. Page 7-15, 4th paragraph: The coefficient for the risk of lung cancer attributable to work in a diesel exhaust job in the case-control study published by our group was for exposure that was assumed to start in 1959. It was not known which workers had exposure pre-1959 (up to an additional 10 to 15 years) since the railroad industry converted to diesel power after World War II, or the intensity of the exposure relative to exposure assessed in the early 1980's. A risk assessment done only using exposure post 1959 would assign an artificially high risk to each year of exposure and inflate the risk. However, the estimates of diesel exhaust exposure chosen (125 $\mu g/m^3$ and 500 $\mu g/M^3$) are high based on the measurements made in railroad workers, and also include background non-diesel particles. The use of these estimates of exposure would tend to lead to a lower risk per μg of presumed diesel exhaust exposure.

Response*: The comment suggests that there are two factors that may modify the exposure estimates used for the case-control study. One factor is the length of exposure, calculated from 1959. Inclusion of exposures prior to 1959 would likely result in a lower risk estimate. The other factor was the high estimates of diesel exhaust exposure. Lower concentrations would likely result in a higher risk estimate. Thus, the two factors raised would tend toward off-setting one another, although one factor may predominate. In any case, we have reevaluated the exposure estimates for the case-control study, and have recalculated the risk estimates accordingly.*

Comment 8: Page 7-18, and page 7-17: The relative risks obtained for the cohort excluding shopworkers are used to develop risk estimates by obtaining the slope of the relative risk plotted versus cumulative exposure. Although the major findings of our study have been replicated when attained age is used in the analysis and when exposure is based on job title held in 1959, we have shown based on our 1991 letter to EPA that the analysis using cumulative exposure based only on age in 1959 does not adequately control for attained age. The slope of the relative risk obtained based on years of exposure (with a 5 year lag) is not positive. Truncation of the study in 1976 due to under ascertainment of death between 1977-1980 even with adequate control of attained age leads to considerable uncertainty in the effects of 10-14 years of exposure, and eliminates the category of 15-17 years of exposure from analysis. Thus the use of these data as presented in Table 7.8 for risk assessment is not justified.

Response: *We have reviewed the Garshick (1991) letter and are unable to identify where it is shown that "the analysis using cumulative exposure based only on age in 1959 does not adequately control for attained age." We identified the information in the letter regarding the use of other attained age models and we did not see a discussion indicating that use of cumulative exposure based only on age in 1959 does not adequately control for attained age. This issue of truncation is an interesting one. In any case, we are no longer using the risk estimate based on the published results as part of the range of risk: the analysis of Table 7.8 of the 1997 draft TSD is not used in the new version of the TSD.*

***Comment 9*: Appendix D: Meta-Analysis.** I would not use the relative risk of 1.82 for the results of retrospective cohort study, but would use a relative risk of 1.45 for the workers age 40-44 in 1959.

Response: *Dr. Garshick is referring to the cohort study of railroad workers for which he is the lead author. The estimate of 1.82 (95% C.I. = 1.30 - 2.55) represents the relative hazard of dying of lung cancer for workers with ≥15 yr cumulative exposure, excluding shop workers and hostlers, the groups most likely to have had exposure to asbestos. The RR estimate for workers with ≥15 yr cumulative exposure, including the shop workers and hostlers (who constituted about 23% of the entire cohort), was 1.72 (95% C.I. = 1.27 - 2.33). In contrast, the RR estimate recommended by the commenter, is 1.45 (95% C.I. = 1.11 - 1.89). However, as noted in Table 3 of the report, when this latter group is stratified by years of putative diesel exposure, 30.8% had ≥ 20 yr, 31.6% had 10-19 yr, 13.6% had 1-9 yr, and 23.9% had 0 years of exposure. Were we to follow the commentator's recommendation, this would be inconsistent with the data extraction procedures used throughout the rest of the meta-analysis, which involved using the estimates derived from the group with the longest or most intense exposure to diesel exhaust. In this study, the authors stratified estimates by duration of exposure, not just by age in 1959. The estimate that we used was based on this latter analysis, in which we extracted the RR estimate for those who had worked for 15+ years.*

Comment 10: Appendix E. Page E-3: It is not clear that attained age categories in 10 year age intervals adequately controls for attained age although this stated. The results with 5 year age intervals should be presented. It is reasonable to exclude the last 4 years of follow-up, and exclude the shopworkers for the reasons stated in the text.

Response: *We conducted some analyses using both 5 and 10 year intervals and found little difference in the results. To simplify calculations in some cases we conducted calculations only using the 10-year age intervals.*

Comment 11: Appendix E. Page E-4: The table on this page lists the models considered. Model 3 includes terms for attained age and calendar year, whereas model 4 includes terms for age cohort and calendar year. These 2 models seem to be the main models of interest, since there is controversy on how to adjust for age in the regression models. The exposure term is expressed as a continuous variable, but it is stated that exposure was also expressed in several categorical variables in the same models to assess the fit of the slope.

Response: *The results for models 3 and 4 were not substantially different. This indicates that control for age was not an important factor for the ramp or roof analyses. Regarding the exposure term, calculations for all the models were done separately for each of the two ways of expressing exposure, either as a continuous or a categorical variable.*

Comment 12: Appendix E. The results, presented in Tables E-2 and E-3 show similar values for slope for most of the models tested, regardless of difference in fit. Figure E-2 shows a categorical analysis, demonstrating relative risk that initially rises, then decreases for the last points. The model fitting cumulative exposure as a continuous variable is "anchored" at zero, and assigned a relative risk of 1.0. Therefore, relative to this point, these other groups "drive," the slope of the line to be positive. In Figure E-3, 3 categorical data points are presented. Again, the slope of the line for cumulative exposure as a categorical variable is positive, largely because the origin is "anchored" at zero exposure, and the subsequent values are positive. The graphs presented do not prove that the models using a single slope adequately describes the relationship between cumulative exposure and lung cancer.

Response: *The comment is incorrect in stating that the model fitting cumulative exposure as a continuous variable model is anchored at zero with an assigned risk of 1.0. Instead, we used the unexposed group as one of the points in the analysis. Only in the categorical analysis does the unexposed group have a relative risk fixed at 1. In the continuous analysis, the regression line is free and can be seen not to go through the point of zero excess relative risk and zero exposure in any of the figures, E-2 through E-6. The procedure to calculate cancer risk from epidemiological studies has been used in a number of our previous analyses such as arsenic, nickel, cadmium and hexavalent chromium.*

Comment 13: Appendix E. Page E-7: The Armitage-Doll model is used as an additional modeling tool. A ten year lag is assumed implying that exposure in the 10 years before death doesn't contribute to cancer. This excludes over half the years of documented exposure in a study that is only 18 years in duration, 1959-1976. Exposure pre-1959 is assigned based on the percentage of diesel engines in service, but since 10 years of actual exposure is discounted, assigned exposure is used substantially to predict lung cancer risk. Given the relatively short duration of known exposure and subsequent follow-up in this cohort, it is hard to justify the use of a 10 year lag to model exposure.

Response: *The comment is correct that the Armitage-Doll model is used as an additional modeling tool. It was suggested at the 1996 Scientific Workshop that use of a multistage model would help address the issue regarding control for age. Consequently, OEHHA conducted additional analyses of the cohort data using a multistage model. In use of multistage models the "lag" refers to the time from the development of the first cancer cell until the time of observation. The Garshick et al. (1988) cohort study bases lung cancer rates upon death certificates reporting lung cancer as the cause of death. Thus, the time of observation in the cohort study is the time of death. The lag refers to time from carcinogenesis until time of death from disease. This lag period is important in cohort studies since the endpoint used in the calculations is death from the disease and not clinical detection of disease. For both the ramp and the roof patterns of exposure, the best fit of the data to the Armitage-Doll model was determined and found to be a model which used 7 sequential transitions necessary to produce the first cancer cell and then a 10-year lag until death.*

For this multistage model, the lag period depended on the specific analysis. The seven-stage model analysis with the next-to-the-last stage sensitive to diesel exhaust used a five year lag. The five-year lag value is reported by the model as the best fit lag value. In contrast, the seven-stage model analysis with the last stage sensitive to diesel exhaust used 10 years for the detection lag. This provided a better fit to the data than a 5-year lag. These models produced highly significant slopes for both the ramp and roof patterns.

In analyses recently conducted by Crump (1997) he investigated the exposure-response relationship for the next-to-the-last-stage affected by diesel exhaust exposure using a lag period of 5 or 10 years. The results did not indicate a progressive increase in lung cancer relative risk with increasing exposure. Despite these trends, the models generally obtained a statistically significant slope. The lag period chosen, 5 or 10 years, did not appear to affect the results.

Comment 14: Appendix F. The discussion listed here recounts historical events, particularly relating to the use of age in various regression models. However, Dr. Dawson does use attained age in some of the models he presents in Appendix E. This should also be noted here.

Response*: That information has been noted in the document as suggested.*

Comments of the Health Effects Institute, letter dated August 18, 1997 to Genevieve Shiroma from Daniel Greenbaum

Comment 1: Health Effects: Animal data. The principle data from studies of effects in animals is data showing that rats, when exposed to high levels of diesel exhaust, develop lung tumors. While this is a significant and clear showing, a number of other studies have suggested that this response is specific to rats exposed at these high levels, and is likely related to an overload of the rats lung clearance mechanism. There is evidence that other species do not develop tumors, even at these high levels; that this response in rats is not specific to diesel exhaust but has been seen in response to a number of other particles; and that since the lung overload is unlikely to occur in humans exposed at much lower ambient levels, it is difficult to extrapolate the effects seen in rats to humans at ambient levels.

Specifically, HEI has published the results of scientific studies in this area over the past two years which are not cited in this document, and which add important information which should be considered:

> *HEI Research Report Number 68* contains an in-depth report of the results of a study HEI funded to compare the carcinogenicity of diesel exhaust and carbon black in rat lungs. Some of the key results have been reported in Nikula *et al.* , (Fundam. Appl.-Toxicol. 25:80-94, 1995) and are cited in the California EPA Health Risk Assessment. However, the complete findings, together with a summary of HEI's peer-review process can be found in the more comprehensive HEI Research Report.
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> *Pulmonary Toxicity of Inhaled Diesel Exhaust and Carbon Black in Chronically Exposed Rats. Part I.- Neoplastic and Nonneoplastic Lung Lesions.* JL Mauderly *et al.* , 1994.

In addition, the results of two companion studies would add important information to Section 5.4 of the California Health Risk Assessment - "Tests Assessing Primary DNA Damage. These companion studies used tissue samples from the animals exposed to diesel exhaust or carbon black in Dr. Mauderly's study and form Parts II and III of Research Report Number 68.

> *Part II.- DNA Damage* K Randerath *et al.* , 1995
>
> *Part III.- Examination of possible Target Genes* SA Belinsky *et al.* , 1995

I am pleased to enclose copies of these reports. In sum, they suggest that the particles, and not the organic chemicals adsorbed to diesel particles, are the likely cause of lung tumors seen in the rats, likely due to a mechanism related to particle overload. This finding does not preclude the possibility that a different mechanism, based on effects of the organic chemicals, could be operating at lower levels in humans, but suggests that that is not the mechanism in rats.

***Response**: Section 5.4 (Tests Assessing Primary DNA Damage) of the document has been changed to include the reference by Randerath et al. (1995). Sections 5 and 6 have also been changed to reflect the association between the references by Nikula et al. (1995) and Mauderly*

et al. (1995), and Swafford et al. (1995) and Belinsky et al. (1995). We appreciate the information submitted, have reviewed the information, and have cited the reports in the health effects document.

OEHHA has decided, following peer review including suggestions made by the Scientific Review Panel, to provide calculations of human cancer risk using rat lung tumor data on an informational basis, but use only the human cancer risk estimations based on human data in the final range of unit risks. We agree that the effects of particle overload impacts on the validity of extrapolation of the rat lung tumor data to lower level exposure in humans. However, we do not view the argument that rat lung tumor data are irrelevant to humans due to particle overload in rats in quite the same way as the commentator.

Sections 6 and 7 of this document state that the mechanism of action by which diesel exhaust induces lung tumors in rats is not established. One proposed mechanism for diesel exhaust-induced rat lung tumors is that exposure to diesel exhaust particulate matter at high concentrations exceeds pulmonary clearance capabilities and causes chronic inflammation. This inflammation leads to macrophage and/or neutrophil-induced oxidative DNA damage resulting in mutations which are instrumental in the induction of lung tumors, and also to cell proliferation which may be mechanistically important to the promotion of the rat lung tumors. This mechanism has also been invoked for carcinogenicity caused by other insoluble particles (e.g. carbon black, titanium dioxide). Rat lung tumor induction due to high dose (2.2 mg/m^3 or higher) exposure to diesel exhaust may share some commonality of mechanism with other carcinogenic insoluble particles; this possibility is discussed in the document. Several authors (e.g. Driscoll, 1996; Nikula et al. , 1997) have hypothesized that this mechanism may have an exposure threshold of action, and tumor induction due to this mechanism would also have a threshold. Gaylor and Zhang (1996) have suggested using the Moolgavkar-Venzon-Knudson clonal expansion carcinogenicity model that small increases in non-necrotic cell proliferation rates which may be undetectable may result in significant increases in tumorigenicity. They also state that 1) a nongenotoxic carcinogen that increases the cell proliferation rate via the cell division rate is not likely to have a threshold dose; 2) dose response curves for cell proliferation and tumor incidence do not necessarily mimic each other. These increases in cell proliferation may be effected either by a stimulated increase in cell division or by an inhibition of apoptosis (programmed cell death).

The rat diesel exhaust carcinogenicity studies included in this document that have evaluated diesel exhaust-induced lung cell proliferation (Heinrich et al. , 1995; Nikula et al. , 1995; 1997) used an insensitive measure of cell proliferation (histopathological comparison to controls). More appropriate measures for making quantitative comparisons of cell proliferation (e.g. labeling index determinations using bromodeoxyuridine (BrdU) DNA labeling) have not been employed, making it premature to state that a true threshold of diesel exhaust-induced lung cell proliferation has been determined. Also, lung cell necrosis has not been noted in any of the rat diesel exhaust carcinogenicity studies. The studies by Driscoll et al. (1996, 1997) did not study diesel exhaust but rather utilized other insoluble particles (a quartz, carbon black) which, unlike diesel exhaust, have no directly genotoxic component which would have implications for low-dose response and therefore limits their applicability to explaining mechanisms of diesel

exhaust-induced rat carcinogenicity. The work of Gaylor and Zheng (1996) is therefore useful in illustrating that cell proliferation, which is one of a number of potential components of the mechanism of rat lung tumor induction by diesel exhaust, may not exert a threshold effect on carcinogenicity. This information does not prove that diesel exhaust-induced carcinogenicity exhibits low dose linearity. However, the clonal expansion carcinogenicity modeling data and the diesel exhaust-induced genotoxicity (including oxidative DNA damage) and Ah (dioxin) receptor binding data indicate that diesel exhaust-induced carcinogenicity may exhibit low dose linearity without the existence of a threshold. A recent report by Borm et al. (1997) indicates that incubating rat lung epithelial-derived cells with human polymorphonuclear lymphocytes (PMN) (either unactivated or activated by preexposure to phorbol myristate acetate) increases DNA adduct formation caused by exposure to benzo[a]pyrene; addition of more activated PMN in relation to the number of lung cells further increased adduct formation in a dose-dependent manner. The authors suggest that "an inflammatory response in the lung may increase the biologically effective dose of polycyclic aromatic hydrocarbons (PAHs), and may be relevant to data interpretation and risk assessment of PAH-containing particulates.". These data raise the possibility that low dose diesel exhaust exposure may result in levels of neutrophil influx which would not necessarily be detectable via histopathological examination as acute inflammation but which might be effective at amplifying any potential diesel exhaust genotoxic effect. WHO (1996) has noted that modeling of human cancer risk from rat lung tumor data should take into account the effects of both particles (carbon core) and extractable organic matter (PAHs, nitro PAHs).

Additionally, some parameters of the "particle overload" hypothesis are incompletely characterized. Alveolar type II cell epithelial hyperplasia has been noted after diesel exhaust exposure, but the measures of cell proliferation used were relatively crude and unsuitable for use in a quantitative estimate of cell proliferation as would be required for biologically-based modeling. It should also be noted that uncertainties exist regarding the magnitude and biological importance of particle overload for diesel exhaust-induced rat lung carcinogenicity. Mauderly et al. (1994) included data from a rat bioassay on the number of neutrophils/mL present in bronchoalveolar lavage fluid from the exposed and control animals (males and females combined). Active oxygen species generated by activated neutrophils are one component of the inflammatory response to diesel exhaust exposure that might be mechanistically important to the induction of tumorigenesis. The number of neutrophils was increased approximately 50-75% for the high carbon black group compared to the low carbon black group; the increase for the high diesel exhaust group was 20-40% compared to the low diesel exhaust group. However, the tumor incidence (males and females combined) for the high carbon black and diesel exhaust groups were approximately 3-fold greater than that for the low carbon black and diesel exhaust groups, respectively. Similarly, the differences in the severity scores for alveolar macrophage hyperplasia and alveolar epithelial hyperplasia in rats that died or were killed after 18 months of exposure between the low and high diesel exhaust groups (approximately 25 and 20%, respectively) do not correlate well with tumor incidence. It would be expected that a better correlation between tumor incidence and indices of inflammation and cell proliferation would exist if diesel exhaust-induced rat lung tumors were solely due to particle overload.

Hattis and Silver (1994) examined lung burden data from diesel exhaust rat carcinogenicity studies and came to the conclusion that "there is continuing accumulation of diesel-derived dust in the lungs of rats throughout life, even at low doses". They also found that this was not predicted by models developed to represent diesel exhaust particulate matter accumulation under "overload" versus nonoverload conditions. Finally, they have found that at high diesel exhaust exposure levels, the increase in the ratio of internal diesel exhaust particulate matter burden to external exposure is not very large, being slightly larger than a factor of 2 at most, and state that "Although dust overloading is a real phenomenon, it is not a very large effect and thus would not be expected to give rise to dramatically lowered estimates of risk at low exposure levels.". It is therefore premature to conclude that the carcinogenic response in rats to diesel exhaust is completely nonspecific.

Furthermore, the rat diesel exhaust lung tumor data discussed in Section 6 of this document is insufficient for the purposes of determining if a threshold for diesel exhaust-induced carcinogenicity exists. As an example, in the study by Mauderly et al. (1987), rats exposed to 350 µg/m^3 diesel exhaust demonstrated a non-statistically significant increase in lung tumor incidence (1.3% compared to 0.8% for controls; relative risk of 1.4). To determine if a difference in lung tumor incidence of that magnitude is significant at a 95% confidence level would require approximately 4000 (Dr. Mauderly estimated 15,000) animals/group. Thus, while the available data do not allow a conclusion that diesel exhaust induces increases in lung tumors at concentrations of less than 2.5 mg/m^3, the data are insufficient for determining that there is a threshold for diesel exhaust-induced rat lung tumors.

Finally, Section 6.1.1.1 of this document lists two studies (Pepelko and Peirano, 1983; Heinrich et al. , 1986a) in which the authors describe statistically significant lung tumor induction in mice in response to diesel exhaust exposure. Takemoto et al. (1986) reported increased tumor incidences in mice which were not statistically significant, but which IARC (1990) determined to be statistically significant after the data were reanalyzed. Section 6.1.3 has been changed to include a study by Ichinose et al. (1997) which describes lung tumor induction in mice after intratracheal exposure to diesel exhaust particulate matter. Of the six mouse diesel exhaust inhalation or intratracheal instillation carcinogenicity studies described in this document, three definitely contain positive results, one probably contains positive results, and two are negative. This indicates that the description of the mouse carcinogenicity results as "mixed" is justified, and it cannot be accurately stated that the only rodent species susceptible to diesel exhaust-induced lung tumors is the rat.

Comment 2: Health Effects: Human Studies. There have been a number of occupational studies of the health effects of long-term exposure to diesel exhaust. Overall, they have found a consistent, small (RR 1.2 - 1.5) association between exposure and increased levels of lung cancer. That association is consistent, and based on HEI's analysis, appears to persist after

controlling for smoking, the single largest possible other explainer of effects, but the association is weakened by an inability to control for other confounders, the absence of exposure measurements for the workers taken at the time of their exposure, and questions about one's ability to estimate a dose-response relationship from these studies.

Specifically, the quantitative risk estimation contained in the *Health Risk Assessment* faces two challenges: (1) the absence of concurrent measurements of exposure during the studies, and the uncertainty about how past exposure relate to the amount and character of today's diesel exposure, is the major limitation to using these data for quantitative risk assessment, and (2) a variety of factors missing data, possible misclassifications of workers as exposed or unexposed, and highly correlated measures of exposure and age - make it difficult to estimate a single, reliable dose-response curve from these data.

- *Integrating Conclusions* - There is some biological rationale for extrapolating the rat tumor data to workers exposed to high (greater than 1,000, $\mu g/m^3$) concentrations of diesel particulate matter, and perhaps to those exposed to 100 - 1,000, $\mu g/m^3$. The rat data do not support the assumption that exposure to diesel exhaust particulate at the levels found in most ambient settings (1 to 10 $\mu g/m^3$) would be sufficiently high to overwhelm lung clearance and induce lung tumors.

  Given the consistent, small associations seen in occupational studies between diesel exhaust exposure and lung cancer, the possibility of a different biological mechanism for lung cancer, based on the mutagenic potential of the organic compounds adsorbed to the diesel particles cannot be excluded. However, uncertainties about the ability to develop a consistent dose-response from these studies, and especially the absence of concurrent exposure measurements, limit one's ability to produce a quantitative estimate of risk from these studies.

***Response**: The comment's summary of effects found in occupational studies is in close agreement with OEHHA's conclusions. The meta-analysis in Appendix D of the 1997 draft TSD, Appendix C of the current draft supports the association summarized in the comment. Lack of concurrent measurements of past exposure does limit the quantitative use of studies, as mentioned in point (1) in the comment. In the case of the Garshick et al. (1987a, 1988) studies, a range of assumptions about the historic exposure gives a likely envelope around the actual pattern of exposure concentrations.*

The factors mentioned in point (2) of the comment do make it difficult to estimate a single, reliable dose-response curve from these data; so the draft TSD constructs a considerable range of likely relationships.

In regard to the lack of support for extrapolation of the rat data to effects on humans at the levels found in most ambient settings (1 to 10 mg/m^3), the current TSD no longer includes predictions of human risk from rat data in the range of human risks. We acknowledge the comment's point that such extrapolation may be appropriate for occupational settings.

In regard to the last paragraph of the comment, the draft TSD does detail uncertainties of the quantitative risk assessment and acknowledges the limitations by developing a range of risks.

Comments on the Executive Summary. In several key respects, this document does not accurately reflect HEI's conclusions, or the broader scientific knowledge about diesel health effects, and the uncertainty-inherent in that knowledge. Specifically:

Comment 3: On the top of Page 13, the Summary cites HEI's works as finding that "the epidemiological data are consistent in showing associations between diesel exhaust and lung cancer," and that the "carcinogenicity of diesel exhaust had been convincingly demonstrated in rats." While these statements are accurate, in as far as they go, they do not reflect the full opinion of HEI that I have presented here today. Specifically, the HEI Diesel Working Group found that the absence of reliable exposure data in the epidemiology studies limits one's ability to use them for quantitative risk assessment, and that the rat data, while convincing for that species, is not likely to be relevant for humans exposed at much lower, ambient levels of diesel exhaust.

On Page 14, the estimates of numbers of cases of cancer caused are presented as a range. However, considering the high level of uncertainty around these numbers, and the tendency of the media and others to leap to the highest number and publish it as the likely risk, it is incumbent upon the agencies to add specific conditional language after discussing any range to caution the media and public against drawing quick conclusions about the "right" number, particularly since the agencies themselves have said there is no "best estimate."

***Response**: With respect to the commentator's concerns regarding any selective citation of the HEI findings, the summary Section 1.5 of Part B addresses sources of uncertainty in the dose response assessment. This section states that "The HEI (1994) cited the general lack of exposure information as limiting the ability to perform a reliable quantitative risk assessment using any of the human data." This statement has been added to the Executive Summary.*

Comment 4: Finally, the statement in the final paragraph on Page 14, that "at recent and current ambient concentrations, diesel exhaust *produces* a significant increase in the likelihood of cancer (italics added)", is not supported by the data, is not consistent with the body of the *Health Risk Assessment* (see, for example, the last paragraph of the summary of the Assessment (p. 1-10)), and seemingly is not necessary to meet the statutory test noted in the same paragraph. This type of statement, if not carefully crafted, can undermine efforts to develop and communicate to the broader public an objective, thoughtful view of what the science is and is not telling us about the health effects of diesel exhaust.

***Response**: Wording will be clarified in revised executive summary.*

Joint Comments of the following organizations submitted August 22, 1997:

Aggregate Producers Association of Northern California, Agricultural Council of California, Almond Haulers and Processors Association, American Automobile Manufacturers Association, American Road and Transportation Builders Association, American Trucking Associations, Associated California Loggers, Association of Energy Services Companies, Association of General Contractors of California, Association of American Railroads, California Cattlemen's Association, California Chamber of Commerce, California Cotton Ginners Association, California Grain and Feed Association, California Independent Oil Marketers Association, California Independent Petroleum Association, California League of Food Processors, California Manufacturers Association, California Mining Association, California Refuse Removal Council of Southern California, California Rental Association, California Trucking Association, Coalition of Petroleum Services, Construction Industry Manufacturers Association, Engine Manufacturers Association, Equipment Manufacturers Institute, Forest Resources Council, National Mining Association, NISEI Farmers League, Raisin Bargaining Association, Western Independent Refiners Association, Western States Petroleum Association.

Comment 1: OEHHA omits important qualifications to HEI's 1995 conclusion that "the carcinogenicity of diesel exhaust has been convincingly demonstrated in rats". In fact, HEI also found that diesel exhaust does not produce lung tumors in hamsters and that results in mice were equivocal. OEHHA should reference HEI's statement that the data "suggests a species-specific carcinogenic mechanism operates in the rat, and that caution is needed in extrapolating the rat data to humans."

***Response**: The animal studies are discussed in Chapter 5, starting on page 6-1. Studies in mice, rats, and hamsters are discussed. OEHHA does not agree that the data in animals to date indicates a species-specific mechanism in rats. Hamsters appear to be resistant to lung tumorigenesis in general. Primates have been inadequately tested. While the mouse studies have mixed results, inhalation exposure to diesel exhaust in some studies and intratracheal instillation of diesel particles in mice produced statistically significant increased incidences of lung tumors relative to controls. Skin painting with diesel extracts produces skin tumors in a mouse model.*

After public and peer review including suggestions made by the Scientific Review Panel, OEHHA has decided to focus on the epidemiological data to estimate human risks. We agree that caution is warranted in extrapolating the rat lung tumor data to humans. The document still describes estimates of unit risk from animal data for informational purposes.

Comment 2: OEHHA omitted the subpopulation of railroad workers in the Garshick cohort with the assumed greatest exposure (shop workers).

***Response**: As indicated by Dr. Garshick (1991), the shop workers who worked in the diesel repair shops shared job codes with workers in non-diesel shops where there was no exposure to diesel exhaust. In addition, the scientific presentations and discussion at the January 1996 meeting affirmed that shop workers were heterogeneously exposed. Some shops entailed diesel*

exhaust exposures in their operations, others did not. In the original Garshick et al. (1988) cohort study, the investigators reported results including and excluding the job classifications of shopworkers and hostlers. They reported that with both shopworkers and hostlers excluded from the analysis, the effect of diesel exhaust exposure remained significant and of comparable magnitude to the whole cohort. Similarly, in the Crump et al. (1991) analysis the presence of an effect did not seem to be affected much by the inclusion or exclusion of shopworkers.

With respect to its exclusion of the shop workers, OEHHA has provided its rationale in Section 7.3.3. paragraph 5. There was substantial heterogeneity of exposures in the broad classification of shop worker. Many of the shop workers were in shops near the engine sheds which had the very high exposures when engines were running without modern ventilation systems. Many other shop workers were in facilities that did not involve diesel exposures. For the exposed shops, measurements by Woskie et al. (1988b) were about twice those of other train workers. There is no useful information on the proportion of workers in the unexposed or lesser exposed shops. Where OEHHA included shop workers in our analyses (1994 OEHHA draft), we assumed that one-half were unexposed. This value was considered an unbiased estimate in the absence of information as to the actual proportion. However, given the greater uncertainty in the shop workers' exposures, and the information that came to our attention during the 1996 Scientific Workshop, OEHHA thought it prudent to simply exclude these workers from the analyses.

Comment 3: Changing simple assumptions about worker age and length of employment eliminates the positive relationship between diesel exhaust exposure and lung cancer in the Garshick data reported by OEHHA. In fact, these changes result in a model prediction which tracks much more closely with the actual lung cancer incidence data from the Garshick cohort.

***Response**: The point that the comment is making is not clear. While age and employment are "simply" assumptions, they are also critical ones. Appendices E and F address the impact of varying assumptions on the association between diesel exhaust exposure and lung cancer. OEHHA has found that the association holds for a variety of reasonable assumptions and approaches.*

Comment 4: Certainly, the weight of scientific opinion suggests that the two studies most central to the Cal-EPA proposal, the rat study authored by Dr. Joe Mauderly and the railroad worker cohort study authored by Dr. Eric Garshick, are not appropriate for use in quantitative human health risk assessment. The latter research draws conclusions in the absence of actual exposure data.

***Response**: With respect to the use of the Mauderly et al. rat data, we shared the comments of Dr. Mauderly with the Scientific Review Panel on October 16, 1997. There was a thorough discussion of the issue of using the rat data for quantitative human risk assessment at the meeting. The sense of the panel was that rat data and calculations provide useful information and should be left in the document; however, because human epidemiologic evidence was available on which to base the human risk estimate, the human data should be used to form the range of risks. Therefore, OEHHA now bases the range of unit risk estimates only on the epidemiological information.*

With respect to the Garshick et al. (1988) cohort study, OEHHA continues to use the data from this study as one basis for estimating unit risk factors for humans. While this study did not have actual exposure measurements for the period 1945-1980, the industrial hygiene studies of Woskie et al. (1988b) provide relevant diesel exhaust occupational exposure information for the different occupations of interest. The Woskie exposure information used locomotives that were typical of the industry during the 1960's and early 1970's and the studies were conducted during the period 1982-1983, a time just after the Garshick cohort study's follow-up period. Therefore, OEHHA, and others who have used the Garshick data, have had to reconstruct the historical exposures using the Woskie (1988b) data and working backwards in time. As discussed further in Part B Section 7.3.2.2.2, Reconstruction of the Time Course of Concentration, OEHHA combined both Bureau of Labor Statistics annual data on the extent of dieselization in the railroad industry and primarily anecdotal information bearing on the greater smokiness of engines of the earlier era in estimating the past exposures of the study population. OEHHA conducted analyses using both the "ramp" pattern (developed by Dr. Crump) which takes into account the extent of dieselization in the industry and its own "roof" pattern (which incorporated information indicating that early diesel engines were much smokier). Therefore, the OEHHA "roof" pattern has more area under the exposure-time curve than the "ramp" pattern. As a result, the reconstruction developed by OEHHA provides higher estimates of past exposure and lower estimates of carcinogenic potency than the "ramp" exposure pattern.

OEHHA believes that these approaches help bracket the exposure uncertainty in the risk assessment. For instance, the "ramp" and "peak" reconstructions estimates of potency differ only about 3 to 10 fold. Furthermore, the range of extrapolation is not large. The state-wide average concentration of diesel exhaust particulate is 2.2 mg/m^3. This value is only one-thirtieth of the occupational exposures measured by Woskie et al. (1988b). The uncertainty involved in the extrapolation from the occupational exposure levels to the ambient levels of concern is itself unusually small.

OEHHA furthermore has determined that it will present a range of risks rather than a single unit value. This range encompasses the values which resulted from analyses of the Garshick et al. (1988) cohort study. It also includes values derived from the pooled relative risk estimates of the meta-analysis. The resulting range of human risk values captures much of the uncertainties stemming from the choice of study and exposure scenarios.

Comments of Dr. Joe Mauderly, Lovelace Research Institute, letter dated July 7, 1997 to Genevieve Shiroma

I find the health assessment section to be improved in several respects from the last review draft, and that many of the issues identified in my previous comments have been successfully resolved. On the other hand, I find the present draft deficient in its use of rat lung tumor data to generate quantitative estimates of human lung cancer risk from environmental exposures to diesel exhaust. I have been closely involved in the use of animal studies to understand health risks from inhaled diesel exhaust, and in continuing efforts to understand the appropriate interpretation of the rat lung tumor results. It is in my opinion, and one that is broadly held, that current scientific evidence strongly suggests that lung tumor results from rats exposed chronically to high concentrations of diesel soot should not be used to estimate human lung cancer risk from environmental exposures. Several types of information supporting this view are reviewed in my attached comments. The fact that the rat-based estimates of risk are lower than those OEHHA derived from human epidemiological data, and thus do not drive the risk assessment, does not make their 'inclusion appropriate, nor does the desire to portray a range of estimates.

General Comments

These comments only pertain to Sections 1, 6, and 7. I did not review other sections in detail.

Comment 1: Based on current scientific understanding, it is not appropriate to use existing lung tumor data from rats to generate quantitative estimates of unit human lung cancer risks from environmental exposures to diesel soot. This is true regardless of whether or not the estimates based on rat data drive the risk assessment. Recent credible scientific reviews, such as those of the Health Effects Institute and the Clean Air Scientific Advisory Committee of the U.S. Environmental Protection Agency have come to this conclusion. Additional information since those reviews have lent further support to the above view, and none has lent support to the contrary. The general basis for this position is stated below and supporting evidence is described in more detail in the comments that follow.

Meaningful increases in lung tumors in diesel soot-exposed rats only occur at exposure rates overwhelming particle clearance defenses and inducing a strong prolonged and progressive inflammatory, and cell proliferative response. Although soot particles accumulate in small amounts at lower exposure levels, there appears to be a threshold exposure rate for triggering progressive lung disease in rats. The apparent threshold for induction of this progressive rat response is at least two orders of magnitude above the rates of environmental exposures to diesel soot. This threshold behavior is not characteristic of current models of chemical carcinogenesis; moreover, it has been shown that soot-associated organic mutagens are not important in the rat lung tumor response to diesel soot. Syrian hamsters and mice do not develop sustained cell proliferation or lung tumors at soot exposure rates carcinogenic in rats; thus it is well-proven that cancer risk estimates for these other rodent species cannot be derived from the rat data. Chronic exposure of nonhuman primates to diesel soot does not induce the cell proliferative response associated with development of lung tumors in rats. Although lifetime cancer studies of diesel exhaust have not been conducted in nonrodent species, there is no scientific basis for assuming

that lung tumors would be induced through the same mechanisms producing tumors in rats, and there is a growing body of evidence that they would not.

The above findings do not prove that: a) there is no lung cancer risk for humans; b) that there is no lung cancer risk for humans from soot-associated organic mutagens; or c) that the risk for humans, if it exists, has a threshold. These are open issues that will have to be resolved on the basis of other information. The findings do indicate that human lung cancer risk from environmental exposures diesel exhaust, if it exists, almost certainly occurs by mechanisms different from those resulting in the rat lung tumors; thus, the rat lung tumor response is not an appropriate basis for quantitative estimates of human lung cancer risk. If this is true, and there is a growing consensus among the scientific community that it is, then it is true regardless of the numerical value, or intended use, of the estimates. It is simply inappropriate to generate human cancer risk estimates from the present rat data for any purpose.

The detailed comments that follow include several points related to the correct interpretation of the rat studies. Although detailed comments are offered for correctness, it should be understood that my summary view is that the rat studies and their lack of relevance to human risk should be discussed in summary form, and then no further manipulation of the rat data should be done for the purpose of estimating human risk.

***Response**: OEHHA has decided, following public review including suggestions made by the Scientific Review Panel, to provide calculations of human cancer risk using rat lung tumor data on an informational basis, but use only the human cancer risk estimations based on human data in the final range of unit risks. We agree that the effects of particle overload impacts on the validity of extrapolation of the rat lung tumor data to lower level exposure in humans. However, we do not view the argument for a threshold in rats in the same way as the commentator.*

Sections 6 and 7 of this document state that the mechanism of action by which diesel exhaust induces lung tumors in rats is not established. One proposed mechanism for diesel exhaust-induced rat lung tumors is that exposure to diesel exhaust particulate matter at high concentrations exceeds pulmonary clearance capabilities and causes chronic inflammation. This inflammation leads to macrophage and/or neutrophil-induced oxidative DNA damage resulting in mutations which are instrumental in the induction of lung tumors, and also to cell proliferation which may be mechanistically important to the promotion of the rat lung tumors. This mechanism has also been invoked for carcinogenicity caused by other insoluble particles (e.g. carbon black, titanium dioxide). Rat lung tumor induction due to high dose (2.2 mg/m^3 or higher) exposure to diesel exhaust may share some commonality of mechanism with other carcinogenic insoluble particles; this possibility is discussed in the document. Several authors (e.g. Driscoll, 1996; Nikula et al., 1997) have hypothesized that this mechanism may have an exposure threshold of action, and tumor induction due to this mechanism would also have a threshold. Gaylor and Zhang (1996) have suggested using the Moolgavkar-Venzon-Knudson clonal expansion carcinogenicity model that small increases in non-necrotic cell proliferation rates which may be undetectable may result in significant increases in tumorigenicity. They also state that 1) a nongenotoxic carcinogen that increases the cell proliferation rate via the cell division rate is not likely to have a threshold dose; 2) dose response curves for cell proliferation

and tumor incidence do not necessarily mimic each other. These increases in cell proliferation may be effected either by a stimulated increase in cell division or by an inhibition of apoptosis (programmed cell death).

The rat diesel exhaust carcinogenicity studies included in this document that have evaluated diesel exhaust-induced lung cell proliferation (Heinrich et al., 1995; Nikula et al., 1995; 1997) used an insensitive measure of cell proliferation (histopathological comparison to controls). More appropriate measures for making quantitative comparisons of cell proliferation (e.g. labeling index determinations using bromodeoxyuridine (BrdU) DNA labeling) have not been employed, making it premature to state that a true threshold of diesel exhaust-induced lung cell proliferation has been determined. Also, lung cell necrosis has not been noted in any of the rat diesel exhaust carcinogenicity studies. The studies by Driscoll et al. (1996, 1997) did not study diesel exhaust but rather utilized other insoluble particles (a quartz, carbon black) which, unlike diesel exhaust, have no directly genotoxic component which would have implications for low-dose response and therefore limits their applicability to explaining mechanisms of diesel exhaust-induced rat carcinogenicity. The work of Gaylor and Zheng (1996) is therefore useful in illustrating that cell proliferation, which is one of a number of potential components of the mechanism of rat lung tumor induction by diesel exhaust, may not exert a threshold effect on carcinogenicity. This information does not prove that diesel exhaust-induced carcinogenicity exhibits low dose linearity. However, the clonal expansion carcinogenicity modeling data and the diesel exhaust-induced genotoxicity (including oxidative DNA damage) and Ah (dioxin) receptor binding data indicate that diesel exhaust-induced carcinogenicity may exhibit low dose linearity without the existence of a threshold. A recent report by Borm et al. (1997) indicates that incubating rat lung epithelial-derived cells with human polymorphonuclear lymphocytes (PMN) (either unactivated or activated by preexposure to phorbol myristate acetate) increases DNA adduct formation caused by exposure to benzo[a]pyrene; addition of more activated PMN in relation to the number of lung cells further increased adduct formation in a dose-dependent manner. The authors suggest that "an inflammatory response in the lung may increase the biologically effective dose of polycyclic aromatic hydrocarbons (PAHs), and may be relevant to data interpretation and risk assessment of PAH-containing particulates." These data raise the possibility that low dose diesel exhaust exposure may result in levels of neutrophil influx which would not necessarily be detectable via histopathological examination as acute inflammation but which might be effective at amplifying any potential diesel exhaust genotoxic effect. WHO (1996) has noted that modeling of human cancer risk from rat lung tumor data should take into account the effects of both particles (carbon core) and extractable organic matter (PAHs, nitro PAHs).

Additionally, some parameters of the "particle overload" hypothesis are incompletely characterized. Alveolar type II cell epithelial hyperplasia has been noted after diesel exhaust exposure, but the measures of cell proliferation used were relatively crude and unsuitable for use in a quantitative estimate of cell proliferation as would be required for biologically-based modeling. It should also be noted that uncertainties exist regarding the magnitude and biological importance of particle overload for diesel exhaust-induced rat lung carcinogenicity. Mauderly et al. (1994) included data from a rat bioassay on the number of neutrophils/mL present in bronchoalveolar lavage fluid from the exposed and control animals (males and

females combined). Active oxygen species generated by activated neutrophils are one component of the inflammatory response to diesel exhaust exposure that might be mechanistically important to the induction of tumorigenesis. The number of neutrophils was increased approximately 50-75% for the high carbon black group compared to the low carbon black group; the increase for the high diesel exhaust group was 20-40% compared to the low diesel exhaust group. However, the tumor incidence (males and females combined) for the high carbon black and diesel exhaust groups were approximately 3-fold greater than that for the low carbon black and diesel exhaust groups, respectively. Similarly, the differences in the severity scores for alveolar macrophage hyperplasia and alveolar epithelial hyperplasia in rats that died or were killed after 18 months of exposure between the low and high diesel exhaust groups (approximately 25 and 20%, respectively) do not correlate well with tumor incidence. It would be expected that a better correlation between tumor incidence and indices of inflammation and cell proliferation would exist if diesel exhaust-induced rat lung tumors were solely due to particle overload.

Hattis and Silver (1994) examined lung burden data from diesel exhaust rat carcinogenicity studies and came to the conclusion that "there is continuing accumulation of diesel-derived dust in the lungs of rats throughout life, even at low doses". They also found that this was not predicted by models developed to represent diesel exhaust particulate matter accumulation under "overload" versus nonoverload conditions. Finally, they have found that at high diesel exhaust exposure levels, the increase in the ratio of internal diesel exhaust particulate matter burden to external exposure is not very large, being slightly larger than a factor of 2 at most, and state that "Although dust overloading is a real phenomenon, it is not a very large effect and thus would not be expected to give rise to dramatically lowered estimates of risk at low exposure levels.". It is therefore premature to conclude that the carcinogenic response in rats to diesel exhaust is completely nonspecific.

Furthermore, the rat diesel exhaust lung tumor data discussed in Section 6 of this document is insufficient for the purposes of determining if a threshold for diesel exhaust-induced carcinogenicity exists. As an example, in the study by Mauderly et al. (1987), rats exposed to 350 µg/m^3 diesel exhaust demonstrated a non-statistically significant increase in lung tumor incidence (1.3% compared to 0.8% for controls; relative risk of 1.4). Comment 9 correctly points out that the problem is a case of sample size. To determine if a difference in lung tumor incidence of that magnitude is significant at a 95% confidence level would require approximately 4000 (Dr. Mauderly estimated 15,000) animals/group. Thus, while the available data do not allow a conclusion that diesel exhaust induces increases in lung tumors at concentrations of less than 2.2 mg/m^3, the data are insufficient for determining that there is a threshold for diesel exhaust-induced rat lung tumors.

Finally, Section 6.1.1.1 of this document lists two studies (Pepelko and Peirano, 1983; Heinrich et al., 1986a) in which the authors describe statistically significant lung tumor induction in mice in response to diesel exhaust exposure. Takemoto et al. (1986) reported increased tumor incidences in mice which were not statistically significant, but which IARC (1990) determined to be statistically significant after the data were reanalyzed. Section 6.1.3 has been changed to include a study by Ichinose et al. (1997) which describes lung tumor induction in mice after

intratracheal exposure to diesel exhaust particulate matter. Of the six mouse diesel exhaust inhalation or intratracheal instillation carcinogenicity studies described in this document, three definitely contain positive results, one probably contains positive results, and two are negative. This indicates that the description of the mouse carcinogenicity results as "mixed" is justified, and it cannot be accurately stated that the only rodent species susceptible to diesel exhaust-induced lung tumors is the rat.

Comment 2: In developing this draft, OEHHA appears to have introduced information on carbon black to support their continued use of the rat data. Nobody questions that diesel soot contains mutagenic and carcinogenic compounds. The fact that those compounds are apparently not significant in the rat response to heavy exposures does not mean that the epidemiological data require human carcinogenicity by carbon black for validation. The fact that rats get lung tumors from heavy exposure to carbon black has no more relevance to human risk from diesel soot than the rat response to diesel soot itself. The light treatment of the carbon black epidemiological information and its predication on the rat response to carbon black detract from the document. If it is felt that the carbon black data are important to support the association between occupational exposures to diesel exhaust and lung cancer, or to support the notion that this association should be extrapolated down to environmental exposure levels, then OEHHA must do a more thorough and credible analysis of the carbon black data.

***Response**: Appendix C was not meant to be a treatise on the tumorigenicity of carbon black. It was generated in response to a comment on an earlier draft. We have removed Appendix C from the document and amended Part C by including the material as a response to comment.*

Comments on Section 1.0 Summary

Comment 3: P1-1, ¶ 1, L 1: Diesel exhaust is a triphasic mixture (gas, vapor, and particle), not a biphasic mixture.

***Response**: The term biphasic has been deleted.*

Comment 4: P1-3, ¶ 1, L 4: First, since not all diesel exhaust is particulate, it's not correct to speak of the "particulate nature of diesel exhaust". Second, the fact that the soot is the critical component of diesel exhaust for tumorigenesis in rats is clearly illustrated by the fact that filtered exhaust does not cause tumors, not by the fact that other particles do as well. The important point, not clearly conveyed by the present wording, is that the aggregate results from studies of diesel soot and the other particles indicate that the response to diesel soot is likely to be a general response of the rat lung to heavy loading with particles, rather than a response to the specific physical/chemical nature of the particles.

***Response**: Section 1.3.1 has been changed to indicate that diesel exhaust particulate matter may be the critical component of diesel exhaust for rat lung tumorigenesis at high doses. As noted in the response to Comment 1, Sections 6 and 7 of this document state that the mechanism of action by which diesel exhaust induces lung tumors in rats is not established. However, the gas and vapor phases of diesel exhaust contain a number of known animal and human carcinogens (e.g.*

acetaldehyde, formaldehyde, 1,3-butadiene, benzene, chlorinated dioxins and benzofurans), and there are data indicating that unfiltered diesel exhaust can induce lung tumors in mice (Heinrich et al., 1986). As noted in the response to Comment 1, it is therefore premature to conclude that the carcinogenic response in rats to diesel exhaust is a completely nonspecific response to diesel exhaust particulate matter.

Comment 5: P 1-5, ¶ 3, L 4: HEI is correctly quoted as finding that the carcinogenicity of diesel exhaust has been convincingly demonstrated in rats. Indeed, all authors, review panels, etc. have concluded that obvious truth. The sentence consists of a half-truth, however, because HEI (and other groups) also concluded that the carcinogenicity results from rats are unlikely to have value for predicting risk to humans exposed to low levels of exhaust.

***Response**: Our statement that the "HEI also found that the carcinogenicity of diesel exhaust had been convincingly demonstrated in rats." is found in the Executive Summary section (Section 1.3, Carcinogenicity) which addresses hazard identification. This section was not meant to reach the dose response assessment issue suggested by the comment. Rather, the section summarizes, in traditional fashion, both the animal and the human evidence as to the carcinogenicity of diesel exhaust. The HEI position regarding use of the rat findings with respect to extrapolation to "ambient exposures to diesel exhaust" relates more to dose response than hazard identification. OEHHA notes that HEI's summaries of their own epidemiology and animal health effects review chapters similarly cited the animal hazard identification findings without explicitly addressing the species mechanism/dose response issue. However, in addressing the substantive issues, OEHHA did not cite the HEI dose response position in that part of our Executive Summary dealing with dose response (Section 1.4.2). This section will be corresponding revised to address the comment.*

Comment 6: P 1-6, ¶ 7: The information in the paragraph is accurate, however, OEHHA 's portrayal of the status of the U.S. EPA's diesel risk assessment effort is another half-truth. OEHHA does not comment on the more important point - that the Clean Air Scientific Advisory Committee told EPA in its 1995 review of EPA's 1994 draft risk assessment that the present scientific evidence does not support using the rat data for estimating human risk regardless of how those risks are calculated.

***Response**: In paragraph 7, page 1-6, OEHHA describes the range of unit risks in the EPA document. We do not comment one way or the other on CASAC's review of the USEPA document. In the past we have focused our comments regarding EPA's review of a chemical based on the information provided in the EPA documents. As the EPA has not yet released a revised document, the EPA response to the CASAC recommendations is unclear. As noted earlier, OEHHA is not including the risk estimates from the animal data in the final range of risk estimates for humans.*

Comment 7: P 1-8, ¶ 8, L 1: Because OEHHA is attempting to develop risk estimates for environmental, rather than occupational, exposures, I'd argue that the overriding uncertainty regarding use of the human data is whether or not one can develop with confidence an exposure dose-response relationship from present data.

***Response**: This paragraph of the 1997 document clearly stated there is uncertainty in the model parameters including the estimate of exposure. The paragraph began "The principal uncertainties in using the human data are the representativeness of railroad workers for the general population, the choice of the analytical model, and the lack of knowledge of the exposure history of the railroad workers including possible exposure to unknown confounders."*

By analyzing the data in different ways, OEHHA presented and addressed some of the uncertainties in the dose-response results. Quantitatively, OEHHA provided a large range of risk estimates to characterize uncertainty. We feel that this range likely encompasses much of the uncertainty introduced by the limited information available.

Comments on Section 6: Carcinogenic Effects

Comment 8: P 6-24, ¶ 1, L 2: This sentence is poorly worded. Particle "accumulation" implies increased retention time, so it's redundant to speak of increased accumulation resulting in increased retention time. Our present understanding of the most correct concept is that, at some exposure rate (concentration x time factor), the rate of particle deposition exceeds the lung's clearance capacity (ability to clear mass with time), clearance is then impaired (slowed), and particles accumulate in the lung at an accelerated rate. Although this may appear as a meaningless "chicken and egg" argument, it is actually critical, because it implies that, below a critical exposure rate, clearance impairment, accumulation of substantive lung burdens of particles, and progressive lung disease wouldn't happen. Indeed, this was exactly the finding of the Mauderly *et al*. (1987) study. Since significant tumorigenesis in rats appears to have been always associated with accelerated accumulation of particles in the lung and the associated inflammatory and proliferative responses, the concept of the relationship between exposure rate and clearance is pivotal to the threshold issue.

***Response**: The sentence referred to in the comment reads as follows: "Increased particle accumulation in the rat lung results in impaired particle clearance and therefore increased retention time, leading to potentially increased mutagen exposure." Particle accumulation does not necessarily depend on increased retention time. As noted in the response to Comment 1, Hattis and Silver (1994) stated that rat bioassay data suggest that "there is continuing accumulation of diesel-derived dust in the lungs of rats throughout life, even at low doses. This is not predicted by any of the models we examined that have been developed to represent dust accumulation under "overload" versus "nonoverload" conditions.". The authors indicate that this is probably due to some proportion of the diesel exhaust particulate matter reaching the alveoli being accumulated in some very long term sequestration compartment, which is cleared either very slowly or not at all. Finally, Hattis and Silver (1994) found that at high diesel exhaust exposure levels, the increase in the ratio of internal diesel exhaust particulate matter burden to external exposure is not very large, being slightly larger than a factor of 2 at most, and stated that "Although dust overloading is a real phenomenon, it is not a very large effect and thus would not be expected to give rise to dramatically lowered estimates of risk at low exposure levels.".*

Comment 9: P 6-25, ¶ 3, L 14: The quote from Mauderly (1994) needs to be placed in context. First, there are three different Mauderly, 1994 references in the bibliography (page R-2 1), and one is listed twice (both as 1994b and 1994c). Being familiar with the references, I know that the one quoted here is the first 1994b reference ("Current Assessment -"). Second, quoting the reference without context suggests that I meant that meaningful increases in lung tumors are likely to have been missed because of the small group sizes. That's not what I meant, and it's extremely unlikely to have been true. In the Mauderly *et al*. (1987) study to which this statement referred, there was a 1.4 "relative risk' for lung tumors in rats exposed for 30 mo to 350 $\mu g/m^3$ in comparison to controls (1.3% incidence vs 0.9%). It is tempting to think that this "increase" might have meaning, because it's of the same order of magnitude of the increases suggested by some of the epidemiological studies of workers. However, the "increase" was from a control level of two adenocarcinomas in males and none in females, to one adenocarcinoma in males and two in females in the exposed group.

The group sizes were 230 controls vs 223 exposed, half male and half female. These groups were over twice as large as the typical 50/gender specified for inhalation carcinogenicity bioassays conducted by the National Toxicology Program, and the resulting "increase" would not be considered meaningful by any regulatory or research body in the world. Indeed, that was the point made in the talk from which the paper was derived. I apologize that the point wasn't made as clearly in the paper. I have calculated that one would have to expose 14,720 rats per group to test this "increase" at levels of 95% confidence and power. Although one could conduct this study, I doubt that OEHHA would care to provide the $40M required for the "bare-bones", two-group study!

My point was, and is, that it's fair to say that animal studies don't have the power to test such small differences adequately, but it's not fair to use that statement to suggest that there are meaningful expressions of carcinogenicity at low levels of diesel exhaust that we have missed because of lack of statistical power.

***Response**: The sentence on page 6-25 of the 1997 document referred to in this comment read as follows: " Mauderly (1994) noted that small increases in lung tumor incidence have been observed in rats at diesel exhaust concentrations which do not induce cell proliferation or fibrosis, but group sizes have been insufficient (maximum of approximately 200) to permit the significance of the small tumor incidence increases to be evaluated with much confidence." The issue of power to detect an effect is a reasonable issue to raise if an assumption of a threshold is driving a quantitative risk assessment. The rat diesel exhaust lung tumor data discussed in Section 6 of this document is insufficient for the purposes of determining if a threshold for diesel exhaust-induced carcinogenicity exists. As an example, in the study by Mauderly et al. (1987), rats exposed to 350 $\mu g/m^3$ diesel exhaust demonstrated a non-statistically significant increase in lung tumor incidence (1.3% compared to 0.8% for controls; relative risk of 1.4). However, the animal study did not have the power to detect an effect at the exposure level of 350 mg/m^3. To determine if a difference in lung tumor incidence of that magnitude is significant at a 95% confidence level would require approximately 4000 (Dr. Mauderly estimated 15,000) animals/group. Thus, while the available data do not allow a conclusion that diesel exhaust*

induces increases in lung tumors at concentrations of less than 2.2 mg/m^3, the data are insufficient for determining that there is a threshold for diesel exhaust-induced rat lung tumors.

Comment 10: P 6-33, ¶ 3, L 4: The word should be "estimated", not "measured". It is disingenuous to suggest that the very indirect indices of exposure used in these studies were "measurements of exposure".

Response: The text does not describe "measurements of exposure", nor was this intended by OEHHA. However, the commenter's misinterpretation indicates that the wording could be improved. Therefore, we have changed the text in the interest of clarity, and have substituted "considered here as truck drivers" for "measured here as truck driving".

Comment 11: P 6-45, ¶ 3, L 5: The majority of studies may have indicated a central estimate of relative risk above 1.0, but it would be useful to indicate the portion of studies for which the lower bound of the 95% confidence interval was above 1.0.

***Response**: The commenter's suggestion has been incorporated in the revised version of the document, as follows: "While many of the studies presented multiple estimates of relative risk, 23 of the 40 studies summarized in Table 6-5 contained at least one estimate described as statistically significant or having a 95% confidence interval whose lower bound exceeded unity."*

Comments on Section 7: Quantitative Cancer Risk Assessment

Comment 12: P 7-1, ¶ 3, L 3: In the previous section, it was stated that exposures above 2.5 mg/m^3 were carcinogenic. Here, the value is 2.0 mg/m^3. Is there additional information, or is this a case of "risk creep"?

***Response**: Sections 6 and 7 have been revised to state that exposures at or above 2.2 mg/m^3 were carcinogenic.*

Comment 13: P 7-2, ¶ 5, L 2: It is not clear how the lung burdens at different exposure times were estimated. The statement suggests that the lung burden after two years of exposure was assumed to have accumulated in a linear manner. That would be an undependable assumption. The actual published data for both diesel soot and carbon black lung burdens during exposure show that the accumulation accelerates as the exposure progresses. This is particularly true for intermediate exposure levels, such as the 3.5 mg/m^3 level in the Mauderly *et al*. (1987) study, in which assuming linearity from the 24-month point results in an overestimate by a factor of two of the actual lung burden at 12 months (Wolff et a]., 1987). Assuming linear accumulations at exposure concentrations causing increases in tumors generally overestimates lung burdens during the first half of exposure.

***Response**: The section in question has been substantially reworded to make clear that in the actual model calculations lung burden was assumed constant at its average value over the course of the experiment. Linear interpolation of the data points in computing the detailed time*

course of the lung burden probably did overestimate the lung burden somewhat because of the acceleration of the time course.

Comment 14: P 7-2, ¶ 5, L 8: It is astounding that OEHHA states its view that environmental exposures would not result in human lung burdens sufficient to warrant use of a lung burden model, yet goes ahead using the rat data to derive a risk factor for humans, and even uses a lung burden model as one of the pathways for developing a risk estimate from the rat data!

***Response**: OEHHA relied more on the epidemiological data in its March 97 document than it did on the animal data. In the revised final draft, animal studies are not used to estimate unit risk factors for the range of risk. Two primary factors led to the decision to focus on the human data for quantitative risk assessment: (1) the uncertainty in extrapolating the animal data to humans because of lack of firm understanding of mechanism and (2) the availability of human data. That being said, it is still desirable to seek as much connection between animal and human results as scientifically feasible. Because of the apparently large role of particles, and therefore lung burden, in producing tumors in rats, it is reasonable to expect a role for particles, and therefore lung burden, in humans. Even with that as a working hypothesis, it is convenient to perform the analyses in humans using simple atmospheric concentration because at low exposures lung burden and atmospheric concentration have a linear relationship. The point of using lung burden in rats is to develop ways to extrapolate the animal results obtained at high exposures down to the low exposures, even if different mechanisms are involved in the two regimes. The rat calculation in the March 1997 draft is a first step in this process.*

Comment 15: P 7-3, 5, L 2: While it is appropriate to exclude the squamous cysts from the Mauderly *et al*. rat lung tumor data, it is no less appropriate for the other studies. From the descriptions in the papers and information presented at meetings, and from knowledge of the rat lung's response to particles in general, it is clear that the high-level exposure groups of all studies developed squamous cysts, just as those in the Mauderly *et al*. study. Given that it is more difficult to determine the portion of squamous cysts in the data published by some of the other laboratories, it is still unreasonable to include these lesions in the tumor counts from some studies and not others. If it is recognized as important (as I certainly agree) to exclude the cysts from rat carcinogenicity results, then it is not appropriate to list results from any study unless the cysts can be removed. As a poor alternative, if data from multiple studies are to be listed in a table, then include the cysts for the Mauderly study as well, with appropriate notations.

***Response**: OEHHA has included the Mauderly et al (1987) tumor incidence data with and without squamous cysts in the revised Chapter 7. We have clarified with footnotes in the appropriate tables which unit risk estimates result from inclusion of squamous cysts.*

It should be noted that the progression potential of squamous cysts (or benign keratinizing cystic squamous cell tumors) is controversial. They have been described by various authors as being nonneoplastic (Mauderly et al., 1994), or as having the potential to progress to malignant tumors (Kittel et al., 1993). Some recent rat diesel exhaust carcinogenicity bioassays (Heinrich et al., 1995; Dasenbrock et al., 1996) have included squamous cysts in their total tumor incidence data, indicating a lack of consensus on this issue.

Comment 16: P 7-3, para 7: Unless I have missed it, there is no attempt to validate the estimates of lung burden by comparing them to those actually measured. Although most of the earlier studies did not measure lung burdens of soot over time, data for lung burdens at 6-month intervals in the Mauderly *et al*. (1987) study were published (most thoroughly in Wolff *et al*., 1987). It would be simple to lend credibility to the present method for estimating lung burdens by comparing the result to known lung burdens. Conversely, a failure to match is a clear indication that the method for estimating lung burdens is not useful. Regardless, not including this obvious step detracts from the credibility of the estimate.

***Response**: The predictions of Yu et al. are of the same order of magnitude as the measurements in Wolff et al. These predictions are only intended to be very approximate. So the draft TSD relied on the USEPA and WHO approach without a formal independent validation.*

Comment 17: P 7-4, para 2: The significance of this observation is not obscure at all, as suggested by the text. The lung tumor response to diesel soot exposure is simply not linear; thus, higher dose groups give higher estimates than lower dose groups. Rather than missing the significance of this result, OEHHA should interpret it as one of several indications that assuming a linear relationship between exposure and carcinogenesis in the rat studies is not appropriate. This is one of several indications that estimating risks at low (environmental) exposures from carcinogenesis at high (greater than occupational) exposures is simply not appropriate, even if the rat response was relevant to potential human responses (which it undoubtedly isn't).

***Response**: The interpretation appears to require more investigation, but the comment is noted as part of the reason for diminishing the role of the animal results.*

Comment 18: P 7-4, para 7: The point about the relative lack of variability of the rat response is not clear. It is true that we imagine individual humans to have wide variabilities in their responses; however, it is not so clear that humans as a population have a much wider variability in their risk than a population of rats. Individual rats vary widely in both their cancer and non-cancer responses to diesel soot. Even in the highest-level exposure group reported (Brightwell *et al*.), less than one-half of the rats had lung tumors, although all were exposed to a uniform near lifetime exposure. The occurrence of cancer is a dichotomous variable. The rats did not all have 40% cancer - only 40% of the rats had cancer! It is not clear from the text how this variability differs from that of humans.

***Response**: An in-bred laboratory rat population is genetically homogeneous relative to the human population. In addition, in experimental toxicology studies, they drink the same water, eat the same food, breathe the same air, are the same age at start of exposure and in most studies are not pregnant. It is a very reasonable assumption that the human population has a wider interindividual variability in response than an inbred strain of rats. One of the reasons that inbred strains are used is to minimize genetic differences (Principles and Methods of Toxicology, A. Wallace Hayes, ed , 1989, p. 239). As stated in Casarett and Doull's Toxicology (fourth edition, 1991, p. 115), homozygous strains are routinely used in toxicological studies to avoid the added complications involved in the use of heterozygous animals. The field of*

pharmacogenetics exists because of the important influences of one's genetic makeup on the response to a toxicant. The human population is heterogeneous in genetic makeup, lifestyle, diet, physiological status, and a host of other factors which influence our response to a toxicant. The same cannot be said for the inbred laboratory rat strains used in toxicological experiments.

Comment 19: P 7-9, ¶ 2, L 8: What is the basis for saying that benign lesions are similar to their malignant counterparts "except at cell junctions"? Where did this idea come from? What is the reference? Malignancy is inferred from several cellular characteristics. I have heard no pathologist make the distinction solely on the basis of "cell junctions". Moreover, the logic that squamous cysts would likely become squamous cell carcinomas if the rats lived longer sounds plausible, but is not well supported. For an example of benign and malignant forms of the same tumor type, we can examine the occurrence of adenomas and adenocarcinomas. In the Mauderly *et al*. (1987) study, the first tumors observed were adenocarcinomas in controls! The second adenoma in exposed rats was not observed until over 800 days (26 months) of exposure, yet four adenocarcinomas had been observed in exposed rats by 24 months. Adenomas were observed to the end of the study; indeed, just as many adenomas as adenocarcinomas were observed at the final sacrifice after 30 months of exposure. Current views hold that some benign tumors can become malignant, but that is not a sufficient basis for assuming that squamous cysts, lesions of unknown significance and peculiar to rats, should be counted as tumors because they would certainly become malignant with time.

***Response**: The reference to benign lesions being similar to their malignant counterparts "except at cell junctions" has been removed from Section 7.2.8. However, it should be noted that the progression potential of squamous cysts (or benign keratinizing cystic squamous cell tumors) is controversial. They have been described by various authors as being nonneoplastic (Mauderly et al., 1994), or as having the potential to progress to malignant tumors (Kittel et al., 1993). Some recent rat diesel exhaust carcinogenicity bioassays (Heinrich et al., 1995; Dasenbrock et al., 1996) have included squamous cysts in their total tumor incidence data, indicating a lack of consensus on this issue.*

Comment 20: P 7-12, ¶ 1: The suggestion that rat studies were not meant to model responses of children because their exposures did not begin until 17 weeks of age has little, if any, basis. The rat lung is still growing at that age, and continues to grow until well after one year of age. Since human lungs don't grow much after sexual maturity, you could just as well state that the rat studies were designed specifically to model risks in growing human lungs. It is worth noting that the study comparing noncancer responses of rat lungs to diesel exhaust between the ages of either 0-6 months or 6-12 months showed lesser effects in the younger than in the older rats (Mauderly *et al*., 1987b). It's hard to appreciate the point being made, especially since lung tumors from environmental agents in immature subjects is not an issue for either rats or children.

***Response**: The growth rates of the various organ systems (including the lungs) in the rat are significantly greater in neonates than in 17 week old animals. Given lifespans for rats and humans of 2 and 70 years, respectively, the ages of the rats used in the carcinogenicity bioassays generally correspond to a human age of 10-12 years of age. This indicates that diesel exhaust*

exposure of 15-17 week old rats would not adequately model the responses of children to diesel exhaust.

A large number of studies exists demonstrating that exposure to a wide variety of carcinogens including PAHs early in life (e.g., in utero and postnatal exposure) results in increased tumor yield than when exposures occur past sexual maturity (10 weeks for rats). Thus, the potential for greater carcinogenic effects of diesel exhaust exposure in very young humans is not adequately modeled in the currently available studies.

Comment 21: P 7-12, ¶ 2, L 20-23: This statement might be relevant if there were significant (or even (substantial) increases in lung tumors at the lower levels, but there weren't. Noting that the organic mutagens might be important for human risk at low levels is appropriate. Suggesting, in the face of all the available data, that the organics might be important for rat carcinogenicity at levels below their "particle-induced" carcinogenicity isn't credible.

***Response**: As noted in the response to Comment 1, Sections 6 and 7 of this document state that the mechanism of action by which diesel exhaust induces lung tumors in rats is not established. The clonal expansion carcinogenicity modeling data and the diesel exhaust-induced genotoxicity (including oxidative DNA damage) and Ah (dioxin) receptor binding data indicate that diesel exhaust-induced carcinogenicity may exhibit low dose linearity without the existence of a threshold. A recent report by Borm et al. (1997) indicates that incubating rat lung epithelial-derived cells with human polymorphonuclear lymphocytes (PMN) (either unactivated or activated by preexposure to phorbol myristate acetate) increases DNA adduct formation caused by exposure to benzo[a]pyrene; addition of more activated PMN in relation to the number of lung cells further increased adduct formation in a dose-dependent manner. The authors suggest that "an inflammatory response in the lung may increase the biologically effective dose of polycyclic aromatic hydrocarbons (PAHs), and may be relevant to data interpretation and risk assessment of PAH-containing particulates.". These data raise the possibility that low dose diesel exhaust exposure may result in levels of neutrophil influx which would not necessarily be detectable via histopathological examination as acute inflammation but which might be effective at amplifying any potential diesel exhaust genotoxic effect. WHO (1996) has noted that modeling of human cancer risk from rat lung tumor data should take into account the effects of both particles (carbon core) and extractable organic matter (PAHs, nitro PAHs).*

Comment 22: P 7-12, ¶ 3, L 7: Why speculate that there might not be a threshold for cell proliferation from a non-genotoxic carcinogen, when we have data showing that cell proliferation is increased in rats at carcinogenic exposure levels of diesel soot, but not at high (compared to environmental exposures), but non-carcinogenic levels? Cell proliferation rates were measured in rats and mice in the Mauderly *et al*. (1987) study after 18 months of exposure. These data were not included in the paper reporting the carcinogenicity results, but were reported in a technical report that is available to the public (Mauderly *et al*., 1983, copy attached). Epithelial cell proliferation was not increased in rats after 18 months of exposure to 350 $\mu g/m^3$ and was not observed histologically at that exposure level, even at the final sacrifice at 30 months. Epithelial proliferation was clearly elevated at the 7080 $\mu g/m^3$ level, in the focal areas of the lung where soot had accumulated, but not in areas devoid of soot. The no-threshold theory of cell proliferation

from diesel exhaust is not supported by these findings, and supporting data are not cited. It is also noteworthy that cell proliferation was not increased in the mice at either exposure level, although there was a slight, nonsignificant, increase in the alveolar interstitium of mice. The data showing this species difference were summarized in the Mauderly (1996a) publication (copy attached). All tumors observed in both species in the study were of epithelial origin. Again, it does not seem appropriate to invoke a theory that cell proliferation may not have a threshold, while ignoring actual data suggesting just the opposite.

Response*: As noted in the response to Comment 1, alveolar type II cell epithelial hyperplasia has been noted after diesel exhaust exposure, but the measures of cell proliferation used (histological examination) were relatively crude and unsuitable for use in a quantitative estimate of cell proliferation as would be required for biologically-based modeling. More appropriate measures for making quantitative comparisons of cell proliferation (e.g. labeling index determinations using bromodeoxyuridine (BrdU) DNA labeling) have not been employed, making it premature to state that a true threshold of diesel exhaust-induced lung cell proliferation has been determined. Additionally, the work by Gaylor and Zheng (1996) does not state that cell proliferation may not have a threshold; it states that 1) small increases in non-necrotic cell proliferation rates which may be undetectable may result in significant increases in tumorigenicity; 2) a nongenotoxic carcinogen that increases the cell proliferation rate via the cell division rate is not likely to have a threshold dose, and 3) dose response curves for cell proliferation and tumor incidence do not necessarily mimic each other.*

Comment 23: P 7-12, ¶ 3, L 1O: The statement that the "rat lung tumor data have not been shown to be mechanistically irrelevant" to human cancer risk presumably trades on the fact that it's nearly impossible to prove a negative. In this statement, and in their continued use of the rat data to derive estimates of human risk, OEHHA chooses to ignore the weight of evidence that the diesel-induced rat lung tumors (and thus their mechanism, whatever it is) are almost certainly irrelevant to human risk. I'll outline the current evidence.

1. The rat lung tumor response has an exposure and mechanistic threshold that is far above environmental exposure levels.

The rat lung tumor response is clearly associated only with heavy lung loading with soot, and the accompanying inflammatory and proliferative responses. Exposures to soot that do not result in an "overloading" of clearance and accelerated accumulation of soot in alveoli do not cause increased lung tumors in rats. Present data support the view that there is a threshold for this response, while current theory would suggest that chemical carcinogenesis from the soot-associated organic mutagens would not have an obvious threshold. Certainly OEHHA in its extrapolation from occupational exposures to environmental risks assumes that the human response would not have a threshold.

Figures from two publications illustrate the evidence for a threshold in the rat response, even if the threshold can't be proven statistically. Figure 5-1 in Mauderly (1992) (copied below, paper attached), illustrates tumor results from multiple rat studies with 30+ month observation times - the studies with the greatest ability to demonstrate carcinogenicity at low levels. Tumor incidence

is expressed as the net increase above control (i.e., exposed-control incidence), and the figure shows that the tumor incidence was not increased in four exposed groups with weekly exposure rates below 120 mg.hr.m^{-3}. While one could certainly draw a line through the origin (zero net increase in incidence) and models could be selected to do the same, it is also clear that there were no net increases in lung tumor incidence in the lowest four exposure groups among these studies. This is not just a statistical weakness, because a glance at the figure reveals that the three highest exposure groups among these four had net increases of zero or below.

Figure 5-1 From Mauderly, 1992

The second illustration of the threshold is Figure I from Heinrich, (1994) (copied below, paper attached). Heinrich demonstrated that diesel soot, carbon black, and titanium dioxide were equally potent in producing lung tumors in chronically-exposed rats when exposure was expressed as the cumulative exposure. Again, the absolute response varied with exposure rate, and the lowest diesel exposure rate did not cause an increase in lung tumors.

Figure 1 From Heinrich, 1994

The "mechanistic" threshold was described in part an earlier comment, The rat lung tumor response only occurs in groups exposed sufficiently to cause accelerated accumulations of soot in the lung, accompanied by a slowing of particle clearance, chronic, active inflammation, a sustained increase in epithelial proliferation, and progressive lung disease manifested both histopathologically and functionally. In the Mauderly *et al*. (1987) study, exposure to 350 $\mu g/m^3$ did not cause any of these effects, nor did similar exposures in other studies. That exposure concentration x time factor would approximate a 24-hour air concentration of 73 $\mu g/m^3$, 24-30 times OEHHA's estimate of continuous average environmental exposures. Again, this exposure did result in the retention of a measurable amount of soot in the lung, but caused no detectable adverse health effect using a wide array of biological evaluations (as described in detail in several publications).

2. The rat lung tumor response can not be extrapolated to other rodents.

It is well-documented from work at multiple laboratories that mice and Syrian hamsters do not express the lung tumor response to diesel soot observed in rats. It is difficult to understand how one could assume that the response could be extrapolated to humans, when it appears in only one species, and cannot be replicated in two other rodent species.

It is also well-demonstrated that the non-cancer responses of mice and Syrian hamsters to diesel soot are different from those of rats. All three species accumulate soot in the lungs to a similar extent and in predominantly alveolar locations. All have inflammatory and fibrotic responses of differing degrees to chronic, heavy exposure. The most striking difference is the much greater epithelial proliferative response of the rat than the other rodents. It is unarguably biologically plausible that the greater tendency of the rat lung epithelium to divide in areas of soot accumulation is associated with their greater tendency to form epithelial metaplasias and neoplasias at those sites. We don't know why the species differ in this way. It is reasonable to

assume that the difference has a genetic basis. If this is true and we understood the genetic basis, it might be possible to determine on a genetic basis whether human lung epithelium might behave more like rats or the other rodents. Regardless of mechanism, our observations point clearly toward a relationship between species differences in epithelial division and growth control and species differences in tumorigenesis from diesel soot and other solid particles.

3. There is growing evidence that the lung epithelial proliferative response of rats to diesel soot and other particles is very unlikely to model the response of the human lung epithelium to accumulations of soot or other particles.

The rat lung responds with similar proliferative and tumor responses to a range of solid, respirable particles, regardless of the chemical nature or mutagenic content of the particles. As OEHHA noted in the draft document, this is illustrated by the fact that rats have identical lung tumor responses to diesel soot and mutagen-poor carbon black. More extremely different particles have different potencies in producing these effects, but the rat appears to have a common pattern of response to diverse materials. Because the rat is commonly used in cancer bioassays, the proper interpretation of this response and its relevance to human lung cancer risk has a considerable importance that has been increasingly recognized by the scientific community over the past decade. Mauderly, (1996b) presented a recent overview of this issue, including current thinking about potential mechanisms of the rat lung tumor response.

Until recently, there was not much evidence to draw upon to determine whether the responses of rat lungs to heavy particle exposures were characteristic of nonrodent species. Mauderly (1994) (copy attached) described evidence demonstrating that large numbers of coal miners have been exposed to dust at levels loading their lungs to a degree' similar to that produced in rat studies, without evidence of a cancer response. That paper also described the evidence that rats exposed heavily to coal dust do develop lung tumors. More importantly, discussions with experienced pathologists reveal that the typical response of human lungs to dust loading differs from that of rats. Snipes (1996) found the published evidence for responses of the lungs of non-rodent species to loading with particles to indicate that lungs of dogs and non-human primates do not mount the type of cellular response typical of rats.

To examine the issue more closely, Nikula *et al*. (1997) (copy attached) studied cellular responses in rats and nonhuman primates exposed for two years by NIOSH to diesel soot and coal dust at 2000 $\mu g/m^3$. The exposures were sufficiently heavy to cause accumulations of particles in lungs of both species, but were not sufficient to cause significant lung tumorigenesis in rats, nor long enough to serve as a carcinogenesis study in monkeys. Nikula *et al*. found clear differences between the lung responses of the two species. First, diesel soot and coal dust accumulated similarly and caused similar tissue responses within each species. Second, the particle accumulations in lungs of monkeys were just as great (on a size-adjusted basis) as in the rats. Third, particles accumulated predominantly in the alveoli of rats (as observed in all previous particle studies), but predominantly in the interstitium of monkeys (as is characteristic of humans). Third, and most importantly, the characteristic epithelial proliferative response occurred in rats exposed to both particles, but virtually no epithelial proliferative response occurred in monkeys exposed to either material.

These findings strongly support the anecdotal evidence that the epithelial response of rats to dust accumulation is not representative of human responses. They also contribute to the growing body of scientific evidence that the rat lung proliferative and tumor responses to heavy particle loading should not be extrapolated to human lung cancer risk. The absence of controlled, matched human and rat exposures makes direct comparisons between the species difficult. Work underway at the Lovelace Respiratory Research Institute is extending the species comparison to human lungs exposed heavily to particles. Human lung materials from collections in the U.S., Europe, and Canada are being assembled for review by an international panel of human and experimental pathologists and comparison to lungs of rats exposed to different types of particles. The goal is to describe and compare, as well as available materials allow, the characteristic responses of the two species in terms of particle sequestration and epithelial abnormalities.

Summary: There is a substantial and growing body of evidence indicating that the rat lung tumor response to diesel soot under heavy exposure and lung loading conditions should not be used to derive mathematical estimates of human lung cancer risk from environmental exposures orders of magnitude lower. At this stage of our understanding, we can be confident that the increased tumor risk in rats is associated with their mounting a strong, prolonged epithelial proliferative response, and suggesting that human lungs exposed to environmental levels of soot might mount epithelial proliferative responses like those observed in rats is preposterous.

***Response**: OEHHA has decided, following peer review including suggestions made by the Scientific Review Panel, to provide calculations of human cancer risk using rat lung tumor data on an informational basis, but use only the human cancer risk estimates based on human data in the final range of unit risk estimates. Specific responses to the three parts of Comment 22 are listed below:*

1) "The rat lung tumor response has an exposure and mechanistic threshold that is far above environmental exposure levels."

The rat diesel exhaust lung tumor data discussed in Section 6 of this document are insufficient for the purposes of determining if an exposure threshold for diesel exhaust-induced carcinogenicity exists. As an example, in the study by Mauderly et al. (1987), rats exposed to 350 µg/m^3 diesel exhaust demonstrated a non-statistically significant increase in lung tumor incidence (1.3% compared to 0.8% for controls; relative risk of 1.4). Comment 9 correctly points out that the problem in this case is sample size. To determine if a difference in lung tumor incidence of that magnitude is significant at a 95% confidence level would require approximately 4000 (Dr. Mauderly estimated 15,000) animals/group. Another study (White et al., 1983) lists tumor incidences of 0/30, 1/30, and 3/30 at diesel exhaust concentrations of 0, 0.25 and 0.75 mg/m^3, respectively. The p value for the 0.75 mg/m^3 group is 0.12 (Fisher exact test); this value is less than the normal 0.05 cutoff, but comes close enough to significance to be suggestive. Thus, while the available data do not allow a conclusion that diesel exhaust induces increases in lung tumors at concentrations less than 2.2 mg/m^3, the data are insufficient for determining that there is a threshold for diesel-exhaust induced rat lung tumors.

Rat lung tumor induction due to high dose (2.2 mg/m^3 or higher) exposure to diesel exhaust may share some commonality of mechanism with other carcinogenic insoluble particles; this possibility is discussed in the document. However, some parameters of the "particle overload" hypothesis are incompletely characterized. Alveolar type II cell epithelial hyperplasia has been noted after diesel exhaust exposure, but the measures of cell proliferation used were relatively crude and unsuitable for use in a quantitative estimate of cell proliferation as would be required for biologically-based modeling. It should also be noted that uncertainties exist regarding the magnitude and biological importance of particle overload for diesel exhaust-induced rat lung carcinogenicity. Mauderly et al. (1994) included data from a rat bioassay on the number of neutrophils/mL present in bronchoalveolar lavage fluid from the exposed and control animals (males and females combined). Active oxygen species generated by activated neutrophils are one component of the inflammatory response to diesel exhaust exposure that might be mechanistically important to the induction of tumorigenesis. The number of neutrophils was increased approximately 50-75% for the high carbon black group compared to the low carbon black group; the increase for the high diesel exhaust group was 20-40% compared to the low diesel exhaust group. However, the tumor incidence (males and females combined) for the high carbon black and diesel exhaust groups were approximately 3-fold greater than that for the low carbon black and diesel exhaust groups, respectively. Similarly, the differences in the severity scores for alveolar macrophage hyperplasia and alveolar epithelial hyperplasia in rats that died or were killed after 18 months of exposure between the low and high diesel exhaust groups (approximately 25 and 20%, respectively) do not correlate well with tumor incidence. It would be expected that a better correlation between tumor incidence and indices of inflammation and cell proliferation would exist if diesel exhaust-induced rat lung tumors were solely due to particle overload.

Hattis and Silver (1994) examined lung burden data from diesel exhaust rat carcinogenicity studies and came to the conclusion that "there is continuing accumulation of diesel-derived dust in the lungs of rats throughout life, even at low doses". They also found that this was not predicted by models developed to represent diesel exhaust particulate matter accumulation under "overload" versus nonoverload conditions. Finally, they have found that at high diesel exhaust exposure levels, the increase in the ratio of internal diesel exhaust particulate matter burden to external exposure is not very large, being slightly larger than a factor of 2 at most, and state that "Although dust overloading is a real phenomenon, it is not a very large effect and thus would not be expected to give rise to dramatically lowered estimates of risk at low exposure levels.".

It is therefore premature to conclude that the carcinogenic response in rats to diesel exhaust is completely nonspecific, or that a mechanistic threshold exists.

2) "The rat lung tumor response can not be extrapolated to other rodents."

Section 6.1.1.1 of this document lists two studies (Pepelko and Peirano, 1983; Heinrich et al., 1986a) in which the authors describe statistically significant lung tumor induction in mice in response to diesel exhaust exposure. Takemoto et al (1986) reported increased tumor incidences in mice which were not statistically significant, but which IARC (1990) determined to be

statistically significant after the data were reanalyzed. Section 6.1.3 has been changed to include a study by Ichinose et al. (1997) which describes lung tumor induction in mice after intratracheal exposure to diesel exhaust particulate matter. Of the six mouse diesel exhaust inhalation or intratracheal instillation carcinogenicity studies described in this document, three definitely contain positive results, one probably contains positive results, and two are negative. This indicates that the description of the mouse carcinogenicity results as "mixed" is justified, and it cannot be accurately stated that the only rodent species susceptible to diesel exhaust-induced lung tumors is the rat.

The rat lung has been a relatively good predictor of known human lung carcinogens (e.g., beryllium compounds, cadmium compounds, chromium VI compounds, nickel compounds, asbestos, crystalline silica, radon, tobacco smoke, coal tar, bis(chloromethyl)ether). Based on bioassay data, the rat appears to be a better predictor of known human lung carcinogens than the mouse or hamster, especially for particulate carcinogens. For example, some cadmium and nickel compounds which test positive in the rat show mixed results in mice and are negative in hamsters. Certain forms of asbestos are positive in rat bioassays but negative or mixed in mouse and hamster. Likewise, crystalline silica is positive in rat bioassays but negative in mice. It is not unusual to see different results of carcinogenicity bioassays in different rodents or even in different strains of the same rodent. Thus, the lack of positive results in hamsters and mixed results in mice does not of itself preclude extrapolation of results in rats to humans.

3) "There is growing evidence that the lung epithelial proliferative response of rats to diesel soot and other particles is very unlikely to model the response of the lung epithelium to accumulations of soot or other particles."

The only available study comparing lung inflammatory and proliferative responses to diesel exhaust in rats and non-rodent species is that of Nikula et al. (1997). As noted above, the conclusions that can be drawn from this study are limited by the techniques used (histological examination) to evaluate lung inflammation and proliferation parameters (e.g. alveolar macrophage hyperplasia and alveolar type II cell epithelial hyperplasia). Data for such evaluations needs to be acquired using more sensitive and quantitative measures (e.g., labeling index determinations). Additionally, in some respects, the inflammatory and proliferative response of rats and monkeys to diesel exhaust is different in degree, but not in kind. As an example, in the Nikula et al. (1997) study, increased alveolar epithelial hyperplasia in noted in 15 of 15 rats examined, but only in 4 of 15 monkeys (27%) examined. However, the severity scores for rats and monkeys were 1.7 and 1.5, respectively. Similarly, the incidences for alveolar macrophage hyperplasia were 15/15 for both rats and monkeys, while the severity scores were 1.7 and 1.2, respectively. This suggests that monkeys are less sensitive than rats to diesel exhaust-induced lung proliferation, but do respond in a similar fashion.

Comment 24: P 7-12, ¶ 4, L 6: Again, it would be useful to compare this estimate to one derived from the actual measured lung burdens. The real data suggest that the time-averaged lung burden would have been less than this.

***Response**: The lung burden for the rat from Table 7-1 is the time-averaged value obtained from the actual measurements. As the comment suggests, this time average for the experiment does seem appropriate to use for the comparison of the human value, which is predicted for a steady state. This is the ratio in the draft.*

Comment 25: P 7-13, ¶ 3: First, saying that the engine used in the Mauderly *et al*. 1987 study was "popular" is a considerable overstatement. It was used because it was the only current manufacture U.S.-made light-duty engine widely available at that time, but it was hardly "popular"! On a more serious note, the (1989) reference at mid-paragraph needs an author.

***Response**: We removed the word "popular" from the description of the automobile engine. Also on a more serious note, we have fixed the missing reference.*

Comment 26: P 7-25, ¶ 1: Inferring on the basis of the Ishinishi study alone that heavy-duty engines produce exhaust that is more carcinogenic than light-duty engines is an unwarranted and unscientific posture. It would be more plausible to cite compositional data for light vs heavy-duty engines, if such data support the idea that there would be a difference in toxicity. Moreover, if you want to make the inference from the Ishinishi results, then you'd better make the case that the fuel, engine type, and combustion characteristics of the heavier of the Ishinishi engines is similar to engines prevalent in heavy vehicles in California today.

***Response**: The text does not infer from the Ishinishi study that heavy duty engines produce exhaust that is more carcinogenic than light duty engines but rather notes that Ishinishi et al (1986) found no statistically significant difference in the carcinogenicity of diesel exhaust from a truck engine versus a light duty engine. The qualitative reasoning about relative toxicity following that statement was premature as well as apparently not being clear; so it has been removed from the text.*

Comment 27: P 7-26, ¶ 2, L 8: Do you really mean "diesel exhaust" here, or do you mean carbon black? It's not clear.

***Response**: This section has been removed from the chapter in response to an earlier comment on the relevancy of issue in carbon black carcinogenicity to diesel exhaust carcinogenesis.*

Comment 28: P 7-30, Table 7. 1: It should be noted that the particles in the control exposure were not diesel soot, but were the particles normally present from the animals themselves. It would be appropriate to subtract this amount from the mass concentrations of the diesel-exposed groups.

***Response**: Comment noted.*

Comment**: **29: Comments on Appendix C. P C-1, ¶ 3: This paragraph is a disclaimer that the evaluation of the carbon black literature is: 1) not comprehensive, and 2) not intended as an evaluation of the carcinogenicity of carbon black. Despite that disclaimer, with which I agree, OEHHA goes right ahead and reviews the literature with emphasis on suggestions that

occupational exposures to carbon black might cause lung cancer, and uses that inference in the document as supporting evidence that small amounts of diesel soot carbon might pose a cancer risk for humans exposed to environmental levels of soot. The argument is not impressive. You can't have it both ways. Either you do a -good job of analyzing the carbon black data (one that would withstand scrutiny), or you don't bring that new information into this draft as support for the carcinogenicity of **a** few micrograms of diesel soot.

Carbon black, like diesel soot, has not been shown to cause significant lung carcinogenicity in rats except those exposed to high concentrations sufficient to result in overload of clearance and prolonged, strong inflammatory and proliferative responses. The studies in rats present strong evidence that the organic fraction of diesel soot is not important in the lung tumor response to heavy exposures. This does not mean that the organics have no importance for human risk. Citing the presently very weak evidence that carbon black could possibly be associated with increased lung cancer risk in workers doesn't strengthen OEHHA's case that environmental exposures to diesel exhaust pose a cancer risk.

***Response**: Appendix C was not meant to be a treatise on the tumorigenicity of carbon black. It was generated in response to a comment on an earlier draft. We have removed Appendix C from the document and amended Part C by including the material as a response to comment.*

Comment 30: P C-5, ¶ 2: The meaning and significance of this conclusion are not clear.

***Response**: Appendix C was not meant to be a treatise on the tumorigenicity of carbon black. It was generated in response to a comment on an earlier draft. We have removed Appendix C from the document and amended Part C by including the material as a response to comment.*

Comments of Roger McClellan, Chemical Industry Institute of Toxicology, letter dated June 30, 1997 to Peter Venturini

Comment 1: Let me start by commending the staffs of the Air Resources Board and the Office of Environmental Health Assessment on the very substantial effort they expended in preparing the various parts of this voluminous report. As an active participant for over two decades in the planning, conducting, analyzing, and interpreting research to understand and assess the potential human health risks of diesel exhaust, I know firsthand the complexity of this issue. I also know the substantial changes that have occurred in the relevant data base and its interpretation over that time and especially during the last five years. As I will relate, my primary concern with the report is the inadequate coverage and inappropriate interpretation of the findings of the past five years as presented in Part B - Health Risk Assessment -for Diesel Exhaust. My primary concerns are in three interrelated areas.

In my professional opinion, the report does not adequately report and interpret the latest findings on the carcinogenicity of diesel exhaust in rodents. The report does accurately relate the findings of the key studies including those my colleagues and I conducted at the Inhalation Toxicology Research Institute in Albuquerque, New Mexico. Those studies did show that exposure of rats to high levels of diesel exhaust particles for two years or longer did increase the incidence of lung tumors. However, it is now known that a similar increase in tumors is seen when rats are exposed to carbon black and other relatively insoluble particles. Further, it is now well established that the mechanisms by which the lung tumors are produced is a high concentration, long-term exposure phenomena. Thus, the rat data are not relevant for use in assessing human lung cancer risk of ambient exposure to diesel exhaust or other particulate matter. Enclosed is a review paper I have published on this subject [McClellan, R.O. (1996), Lung Cancer in Rats from Prolonged Exposure to High Concentrations of Carbonaceous Particles: Implications for Human Risk Assessment, *Inhalation Toxicology* 8:193-226].

***Response**: The OEHHA document reviewed the inhalation rat cancer bioassays with carbon black and titanium dioxide and discussed their implications with respect to the interpretation and use of the results obtained with diesel exhaust in the rat lung. The OEHHA staff have also reviewed the submitted paper. Following peer review, including a public review and discussion of the issue by the Scientific Review Panel, OEHHA has decided to base the range of unit risks derived for diesel exhaust only upon the epidemiological data.*

Comment 2: In my professional opinion, the report overinterprets the findings of the various epidemiological studies including the key studies by Garshick *et al.* Taken collectively, these studies at best suggest a weak association between exposure to diesel exhaust and increased risk of lung cancer. For some time, I and others working in this field interpreted the Garshick *et al.* studies as providing the strongest evidence for a positive association between work involving diesel exhaust exposure and increased risk of lung cancer. My earlier confidence in these studies led me to use them for developing quantitative estimates of the potency of diesel exhaust.

However, a rigorous reanalysis of the Garshick *et al.* data raises serious questions about the validity of the conclusions drawn by Garshick *et al.* The detailed reanalysis was conducted by

Crump and associates [Crump, K.S., Lambert, T., and Chen, C. (1991). Assessment of Risk from Exposure to Diesel Engine Emissions: Report to the U.S. Environmental Protection Agency for Contract 68-02-4601 (Work Assignment no. 182). Washington, DC: Office of Health and Environmental Assessment, U.S. Environmental Protection Agency].

In my professional opinion, the flawed interpretations noted above have led to the inappropriate development of a cancer potency estimate for exposure to diesel exhaust. The present scientific data do not support the development of quantitative estimates of the potency of diesel exhaust exposure for causing human lung cancer. Although I and my colleagues have previously published such estimates and they are referenced in the report, these estimates are no longer valid because of the more recent findings noted earlier.

***Response**: In evaluating the epidemiological literature, OEHHA identified 47 potentially relevant studies. Of these, 31 met the objective inclusion criteria. The Garshick case-control and cohort studies of railroad workers were both included. The OEHHA meta-analysis weighed each of the 31 epidemiological studies according to standard criteria. By excluding one study at a time, the influence analysis did not reveal any single study (including either of the two Garshick studies) as being particularly influential to the overall estimate of relative risk from the meta-analysis. The results of the meta-analysis were similar across different occupational categories and study designs. Taken collectively, these studies found consistent evidence of a weak association between exposure to diesel exhaust and increased risk of lung cancer. The relative risk estimates obtained in the Garshick case-control and Garshick cohort studies were close to many of the summary relative risk values derived in the meta-analysis. Therefore, the meta-analysis supports reliance upon the Garshick studies.*

OEHHA acknowledges that uncertainty as to the study population's exposure histories affects the overall confidence in the study results. However, where exposure data are alleged to be uncertain, reanalyses of epidemiological studies which rely upon such uncertain data can not convincingly support the rejection of the validity of the epidemiological studies involved. Furthermore, OEHHA notes that Dr. Crump performed analyses of the Garshick et al. (1988) cohort study and, that the commentator, without more support, uses those analyses to challenge the validity of not only the cohort study but also the Garshick et al. (1987) case-control study with its very different study design.

OEHHA does not accept the major premise that supports the commentator's conclusion opposing use of the Garshick cohort study. OEHHA's analyses and interpretation of the Garshick et al. (1988) cohort study differ from those of Dr. Crump, including the substantial corrections to the 1991 report that he has transmitted to us. The nature of those differences are described in detail in Appendices E and F of the revised document. We have reevaluated the exposure data for the railroad workers and have identified a range of exposures for use in the quantitative risk assessment.

Comment 3: The results of neither the rat nor the epidemiological studies provide a basis for developing quantitative estimates of potency. As discussed earlier, the mechanism by which lung tumors were produced by high level, prolonged exposure to diesel exhaust are not relevant for

assessing human cancer risks from ambient exposures to low concentrations of diesel exhaust and other particulate matter. The epidemiological findings, suggestive of a positive association between diesel exhaust exposure and increased cancer incidence, cannot be used for developing quantitative estimates of potency because of the crudeness of the estimates of past exposure. This point is emphasized by Dr. Eric Garshick in his May 30, 1995 memo to the EPA commenting on the EPA's health assessment document for diesel emissions. A copy of the cover page and pages 7 and 8 of his memo is attached. He concludes by stating that "it is not possible to use the human epidemiologic data that was reanalyzed to assign a unit risk with confidence due to the uncertainty of the exposure data." I concur with his opinion concerning the use of the findings he has published.

In summary, it is my opinion that the report does not adequately discuss and use the most recent findings on the health effects of diesel exhaust. This results in the report overstating the cancer risks of exposure to ambient levels of diesel exhaust.

***Response**: With respect to concerns regarding the use of the animal data, OEHHA presented similar concerns to the Scientific Review Panel on October 16, 1997. There was a thorough discussion of the issue of using the rat data for quantitative human risk assessment at the meeting. The sense of the panel was that rat data and calculations provide useful information and should be left in the document, however, since human epidemiologic evidence was available on which to base the human risk estimate, that the human data should be used to form the range of risks. Therefore, OEHHA now bases the range of unit risk estimates only on the epidemiological information.*

The crudeness of the estimates of past exposure does contribute more than usual to the overall uncertainty of the dose response risk assessment for diesel exhaust based upon the epidemiological findings. The overall magnitude of that uncertainty is not unduly large and is substantially offset by the much smaller than usual range of extrapolation from the occupational exposures of interest to the ambient levels of concern here. Nevertheless, OEHHA did not assign a single unit risk to which reduced confidence would attach. Instead, OEHHA presented a broad range of risk so as to fairly capture the scope of the uncertainty in these analyses, including the uncertainty related to the lack of exposure data.

Comments from Mercedes-Benz of North America, Inc., letter to Mr. Robert Krieger, dated August 22, 1997 from Harold Polz

Outlined below are the concerns of Mercedes-Benz. These comments focus on California's finding of carcinogenicity, the weight of evidence supporting this finding, and the quantitative risk assessment presented in the documents.

Comment 1. These documents should be amended to present the full range of views, regarding the interpretation of rat studies and the Garshick studies, particularly regarding their use to derive unit risk factors.

At the July 1 workshop, Richard Becker of OEHHA stated that the risk assessment analysis was "intended to capture the entire range of thought" on the health effects of diesel exhaust. The current draft, however, does not accomplish this goal. This fact was dramatically demonstrated at that workshop when Dr. Greenbaum, head of the Health Effects Institute (HEI) and a former state director of environment, expressed his concern that the documents, particularly the executive summary which is most likely to be read by the public and key decision makers, do not provide any sense of the level of controversy and degree of disagreement within the scientific community surrounding the findings in the risk assessment analysis. Mercedes-Benz shares this concern.

For example, the Executive Summary dated May 9 is 14 pages long but has only a few general sentences addressing the scientific appropriateness of the agency's risk estimate. The summary never even acknowledges the concerns expressed by various diesel effects experts regarding the relevance of rat studies when drawing conclusions about human cancer risk nor the inappropriate use of the Garshick data to derive a unit risk factor for human exposures. If the agency is truly interested in airing the full range of views then the executive summary should dearly state that:

> "Dr. Mauderly, the author of the rat study being used by OEHHA to assess human risk, has gone on record as saying that his research is not relevant for assessing effects in humans."

Dr. Crump, as co-author of an EPA diesel risk analysis and continuing analyst of the Garshick data, has found that "there is no convincing evidence for an effect of diesel exhaust exposure upon lung cancer in this cohort;"

Dr. McClellan has retracted his past derivation of a risk factor from the Garshick data and now believes that "The results of neither the rat nor the epidemiological studies provide a basis for developing estimates of potency" and, finally;

> Dr. Garshick, author of the Garshick studies in question, has taken the position that "It is not possible to use the human epidemiologic data that was reanalyzed to assign a unit risk with confidence."

In other words, the risk assessment analysis should clearly acknowledge that every scientific investigator relied upon to support the development of a carcinogenic risk factor for diesel

exhaust has clearly and unequivocally stated that their research is not appropriate for reaching such a scientific finding. The body of the OEHHA risk assessment analysis, without such an acknowledgment, does not address the findings of key researchers. Mercedes-Benz is aware that Dr. Crump, Dr. Mauderly, and Dr. McClellan have also completed new research and will file this information in their written comments. Mercedes-Benz requests that the findings of this research and analysis from these key researchers be presented in their own words in the body of the report so that the reader can truly understand the "entire range of thought" on the interpretation of evidence of cancer effects of diesel exhaust.

***Response**: The document does provide a full range of views. Part C provides the views of every commentator and the OEHHA response.*

As a practical matter, where the subject matter is as complex as diesel exhaust quantitative risk assessment and the different entities' positions are so very closely nuanced at each level of analysis, it is not feasible to fully and reliably distinguish those positions in an Executive Summary. However, while necessarily abbreviated, the Executive Summary does not gloss over major differences in scientific opinion in the interpretation of the evidence. For instance, with respect to controversies in the use of the epidemiological data, the Executive Summary has a Section (1.4.3.1) titled, Differing Analyses of the Garshick et al. (1988) Cohort Data. This section is primarily devoted to describing the issues raised by Dr. Crump and refers the reader to those parts of the document treating the issue at length. The document states that "these differences in the analyses and interpretation of the Garshick cohort study underlie some of the differences amongst these authorities [USEPA, HEI] in the overall quantitative assessment of the human risk based upon the epidemiological data. Furthermore, where multiple parties as here have relied upon the work of Dr. Crump to reach the same or similar conclusions, it clarifies the scope of the disagreement to give precedence and emphasis to the work of Dr. Crump. In any case, we have revisited the Executive Summary to address the issues raised by Mr. Greenbaum of the HEI.

With respect to the comments of Dr. Garshick to the effect that his railroad worker cohort epidemiologic data could not be reanalyzed to assign a unit risk with confidence, the draft presented a very broad range of unit risks and not a single value of unit risk to which such confidence would attach.

With regard to the "retraction" by Dr. McClellan of his calculations of potency based upon epidemiological data, our draft document antedates his "retraction" and could not have addressed it or its merits. However, we are currently evaluating the exposure range of the case-control study to provide an additional analysis.

With respect to the use of the animal data, the Executive Summary describes and emphasizes the major uncertainties and assumptions identified by whatever source. With respect to those concerns and the comments of Dr. Mauderly, we shared them with the Scientific Review Panel on October 16, 1997. There was a thorough discussion of the issue of using the rat data for quantitative human risk assessment at the meeting. The sense of the panel was that rat data and calculations provide useful information and should be left in the document, however, since

human epidemiologic evidence was available on which to base the human risk estimate, that the human data should be used to form the range of risks. Therefore, OEHHA now bases the range of unit risk estimates only on the epidemiological information.

Comment 2: There is substantial scientific evidence that the effects research on rats exposed to diesel exhaust should not be used to assess human health effects. The OEHHA risk assessment analysis should be revised to reflect the latest research and present the views of key researchers regarding this point.

Dr. Joe Mauderly is recognized worldwide as one of the leading researchers on the effects of diesel exhaust on rats. It is Dr. Mauderly's work which was specifically referenced by OEHHA to support the conclusion that "diesel exhaust causes lung cancer in animals" (page ES-12 of the executive summary) and to derive a specific risk estimate for humans. Dr. Mauderly has published his findings in peer reviewed literature and testified at each of the OEHHA workshops. Dr. Mauderly's conclusions are, however, in stark contrast to those contained in the report, even though they are supposedly based on his research. Dr. Mauderly has determined that the rat research relied upon by OEHHA does not demonstrate that a similar effect will occur in humans at current human environmental exposure levels (if at all). (See Reference 1 attached) Dr. Mauderly has also concluded that it would be bad scientific judgment to use such rat studies to even attempt to estimate human risk. As the originator of the data used to estimate human risk, Dr. Mauderly's views should be presented fully in this document and, either OEHHA should refute his findings in detail or follow his advice.

Dr. Roger McClellan of the Chemical Industry Institute of Toxicology recently published a paper entitled "Lung Cancer in Rats from Prolonged Exposure to High Concentrations of Carbonaceous Particles: Implications for Human Risk Assessment." (See Reference 2 attached) In this paper, Dr. McClellan makes several important findings which are directly at odds with the agency's draft. For example, Dr. McClellan finds that the organics on the diesel particle are not contributing to observed cancer in the rats. In stark contrast to this conclusion, OEHHA relies specifically on the prospect of chemical contribution in rat studies as the scientific basis for suggesting there is no threshold for the observed effects in rats. This conclusion is in direct conflict with Dr. McClellan's conclusion that "the observed effects in rats appear to be threshold phenomena that occur only with prolonged exposure to high concentrations of particles. Thus the rat lung cancer findings at high concentrations should not be extrapolated to low concentrations using the linearized multistage model typically used as a default assumption for assessing the cancer risk of chemicals." Even with this direct conflict in conclusions, highlighted by the similar conclusions of HEI and Dr. Mauderly, the OEHHA risk assessment analysis fails to present any scientific justification for its no threshold effect conclusion. Mercedes-Benz requests that the report either be conformed to the findings of Dr. McClellan or a detailed analysis be presented by OEHHA regarding why such research is incorrect and can be ignored.

***Response**: With respect to the comments of Dr. Mauderly, we shared them with the Scientific Review Panel on October 16, 1997. There was a thorough discussion of the issue of using the rat data for quantitative human risk assessment at the meeting. The sense of the panel was that rat data and calculations provide useful information and should be left in the document,*

however, since human epidemiologic evidence was available on which to base the human risk estimate, that the human data should be used to form the range of risks. Therefore, OEHHA now bases the range of unit risk estimates only on the epidemiological information. However, it was also clear that the information presented to justify the existence of a threshold response in the rat data were not considered convincing by the Panel.

Sections 6 and 7 of this document state that the mechanism of action by which diesel exhaust induces lung tumors in rats is not established. One proposed mechanism for diesel exhaust-induced rat lung tumors is that exposure to diesel exhaust particulate matter at high concentrations exceeds pulmonary clearance capabilities and causes chronic inflammation. This inflammation leads to macrophage and/or neutrophil-induced oxidative DNA damage resulting in mutations which are instrumental in the induction of lung tumors, and also to cell proliferation which may be mechanistically important to the promotion of the rat lung tumors. This mechanism has also been invoked for carcinogenicity caused by other insoluble particles (e.g. carbon black, titanium dioxide). Rat lung tumor induction due to high dose (2.5 mg/m^3 or higher) exposure to diesel exhaust may share some commonality of mechanism with other carcinogenic insoluble particles; this possibility is discussed in the document. Several authors (e.g. Driscoll, 1996; Nikula et al., 1997) have hypothesized that this mechanism may have an exposure threshold of action, and tumor induction due to this mechanism would also have a threshold. Gaylor and Zhang (1996) have suggested using the Moolgavkar-Venzon-Knudson clonal expansion carcinogenicity model that small increases in non-necrotic cell proliferation rates which may be undetectable may result in significant increases in tumorigenicity. They also state that 1) a nongenotoxic carcinogen that increases the cell proliferation rate via the cell division rate is not likely to have a threshold dose; 2) dose response curves for cell proliferation and tumor incidence do not necessarily mimic each other. These increases in cell proliferation may be effected either by a stimulated increase in cell division or by an inhibition of apoptosis (programmed cell death).

The rat diesel exhaust carcinogenicity studies included in this document that have evaluated diesel exhaust-induced lung cell proliferation (Heinrich et al., 1995; Nikula et al., 1995; 1997) used an insensitive measure of cell proliferation (histopathological comparison to controls). More appropriate measures for making quantitative comparisons of cell proliferation (e.g. labeling index determinations using bromodeoxyuridine (BrdU) DNA labeling) have not been employed, making it premature to state that a true threshold of diesel exhaust-induced lung cell proliferation has been determined. Also, lung cell necrosis has not been noted in any of the rat diesel exhaust carcinogenicity studies. The studies by Driscoll et al. (1996, 1997) did not study diesel exhaust but rather utilized other insoluble particles (a quartz, carbon black) which, unlike diesel exhaust, have no directly genotoxic component which would have implications for low-dose response and therefore limits their applicability to explaining mechanisms of diesel exhaust-induced rat carcinogenicity. The work of Gaylor and Zheng (1996) is therefore useful in illustrating that cell proliferation, which is one of a number of potential components of the mechanism of rat lung tumor induction by diesel exhaust, may not exert a threshold effect on carcinogenicity. This information does not prove that diesel exhaust-induced carcinogenicity exhibits low dose linearity. However, the clonal expansion carcinogenicity modeling data and the diesel exhaust-induced genotoxicity (including oxidative DNA damage) and Ah (dioxin)

receptor binding data indicate that diesel exhaust-induced carcinogenicity may exhibit low dose linearity without the existence of a threshold. A recent report by Borm et al. (1997) indicates that incubating rat lung epithelial-derived cells with human polymorphonuclear lymphocytes (PMN) (either unactivated or activated by preexposure to phorbol myristate acetate) increases DNA adduct formation caused by exposure to benzo[a]pyrene; addition of more activated PMN in relation to the number of lung cells further increased adduct formation in a dose-dependent manner. The authors suggest that "an inflammatory response in the lung may increase the biologically effective dose of polycyclic aromatic hydrocarbons (PAHs), and may be relevant to data interpretation and risk assessment of PAH-containing particulates.". These data raise the possibility that low dose diesel exhaust exposure may result in levels of neutrophil influx which would not necessarily be detectable via histopathological examination as acute inflammation but which might be effective at amplifying any potential diesel exhaust genotoxic effect. WHO (1996) has noted that modeling of human cancer risk from rat lung tumor data should take into account the effects of both particles (carbon core) and extractable organic matter (PAHs, nitro PAHs).

Additionally, some parameters of the "particle overload" hypothesis are incompletely characterized. Alveolar type II cell epithelial hyperplasia has been noted after diesel exhaust exposure, but the measures of cell proliferation used were relatively crude and unsuitable for use in a quantitative estimate of cell proliferation as would be required for biologically-based modeling. It should also be noted that uncertainties exist regarding the magnitude and biological importance of particle overload for diesel exhaust-induced rat lung carcinogenicity. Mauderly et al. (1994) included data from a rat bioassay on the number of neutrophils/mL present in bronchoalveolar lavage fluid from the exposed and control animals (males and females combined). Active oxygen species generated by activated neutrophils are one component of the inflammatory response to diesel exhaust exposure that might be mechanistically important to the induction of tumorigenesis. The number of neutrophils was increased approximately 50-75% for the high carbon black group compared to the low carbon black group; the increase for the high diesel exhaust group was 20-40% compared to the low diesel exhaust group. However, the tumor incidence (males and females combined) for the high carbon black and diesel exhaust groups were approximately 3-fold greater than that for the low carbon black and diesel exhaust groups, respectively. Similarly, the differences in the severity scores for alveolar macrophage hyperplasia and alveolar epithelial hyperplasia in rats that died or were killed after 18 months of exposure between the low and high diesel exhaust groups (approximately 25 and 20%, respectively) do not correlate well with tumor incidence. It would be expected that a better correlation between tumor incidence and indices of inflammation and cell proliferation would exist if diesel exhaust-induced rat lung tumors were solely due to particle overload.

Hattis and Silver (1994) examined lung burden data from diesel exhaust rat carcinogenicity studies and came to the conclusion that "there is continuing accumulation of diesel-derived dust in the lungs of rats throughout life, even at low doses". They also found that this was not predicted by models developed to represent diesel exhaust particulate matter accumulation under "overload" versus nonoverload conditions. Finally, they have found that at high diesel exhaust exposure levels, the increase in the ratio of internal diesel exhaust particulate matter

burden to external exposure is not very large, being slightly larger than a factor of 2 at most, and state that "Although dust overloading is a real phenomenon, it is not a very large effect and thus would not be expected to give rise to dramatically lowered estimates of risk at low exposure levels.". It is therefore premature to conclude that the carcinogenic response in rats to diesel exhaust is completely nonspecific.

Furthermore, the rat diesel exhaust lung tumor data discussed in Section 6 of this document is insufficient for the purposes of determining if a threshold for diesel exhaust-induced carcinogenicity exists. As an example, in the study by Mauderly et al. (1987), rats exposed to 350 µg/m^3 diesel exhaust demonstrated a non-statistically significant increase in lung tumor incidence (1.3% compared to 0.8% for controls; relative risk of 1.4). Comment 9 correctly points out that the problem is a case of sample size. To determine if a difference in lung tumor incidence of that magnitude is significant at a 95% confidence level would require approximately 4000 (Dr. Mauderly estimated 15,000) animals/group. Thus, while the available data do not allow a conclusion that diesel exhaust induces increases in lung tumors at concentrations of less than 2.5 mg/m^3, the data are also insufficient for determining that there is a threshold for diesel exhaust-induced rat lung tumors.

Comment 3: Extensive analyses by Dr. Kenny Crump have shown that the data in the Garshick studies cannot be used to establish unit risk factors for diesel exhaust. Dr. Crump's basic concerns with that data base are shared by several key researchers, including Dr. Garshick. Dr. Crump's findings and conclusions should be presented and addressed both in the body of the effects document and in the executive summary, as this is the most critical element of the agency's findings.

For years the work of Dr. Garshick on railroad workers has been viewed as the most comprehensive data base and epidemiological analysis of the effects of diesel exhaust on humans. For this reason, Dr. Kenny Crump was hired by the U.S. EPA to determine whether the Garshick data could be used to derive a unit risk factor. Dr. Crump found that the data could not be used to support any unit risk factor. Since that original analysis, Dr. Crump has continued to apply a wide range of analytical tools to the Garshick data to determine its implications for a relationship between diesel exposure and human lung cancer risk. His views were expressed clearly and compellingly at the July workshop and in the written comments he has filed on the pending draft documents. Dr. Crump has pointed out that:

- "The significant dose response trends found by OEHHA appear to be solely a consequence of the fact that train riders had a higher risk than clerks and signalmen."

- "Lung cancer mortality was not significantly elevated among shopworkers in comparison to clerks and signalmen, despite the fact that the shopworkers likely had the most intense exposure of any group"

- "Relative risk of lung cancer tended to decrease with increasing duration of exposure."

- "This lack of evidence for a dose response leads me to conclude that there is no convincing evidence for an effect of DE exposure upon lung cancer in this cohort."

Since the Garshick data is the cornerstone of the draft health effects report and the unit risk factor presentation in it, Mercedes-Benz believes it is extremely important that the report presents Dr. Crump's findings in detail and explains in detail whether the agency agrees with his findings or why the agency has found that Dr. Crump's analyses are in error or that they are being misinterpreted. To date, the agency documents and the comments at the various meetings at which Dr. Crump has spoken, have not indicated why Dr. Crump's views should be dismissed.

In the agency's 1994 draft, Dr. Crump's work was not addressed and interested parties were told that it was because his work had not been peer reviewed. The agency can no longer rely on this excuse. Dr. Roger McClellan of CIIT has reviewed Dr. Crump's work and has concluded in his recent publication in Inhalation Toxicology that the Crump work clearly indicates that the Garshick data "should not be used to derive quantitative estimates of lung risk for diesel exhaust exposure". Dr. S. Moolgavkar of the University of Washington reached the same conclusion after reviewing the Garshick data and has presented those findings at the December, 1996, VDA conference in Germany. Both Dr. McClellan's and Dr. Moolgavkar's findings should be presented in the agency's report and addressed as to whether the agency agrees or disagrees with these expert opinions.

***Response**: Responses to quotations from the comments of Dr. Crump are found with his comments. His comments, presenting his most recent results, are quoted in full.*

The 1997 draft TSD did address the Crump et al. (1991) report. The 1997 draft TSD pointed out that an error in the Crump et al. (1991) reanalysis invalidated the results for cumulative exposure in that report. The 1997 draft also pointed out that Dr. Crump's more recent results, outlined in correspondence with OEHHA, corrected the error. However, in his own comments on the 1997 draft, cited in this comment, Dr. Crump continues to hold the same general view of the data, "This lack of evidence for a dose response leads me to conclude that there is no convincing evidence for an effect of DE exposure upon lung cancer in this cohort." The current draft TSD points out that, after considerable dialogue, including improving some hypotheses as mentioned in Apppendices E and F, the insignificant results obtained by Crump (1995, 1996, 1997) for the ramp exposure pattern appear to depend at least in part upon his assumption of unexposed workers having, in effect, a backgound level of diesel exhaust. The current draft TSD, Part B, also points to potential explanations of the up-and-down trends obtained for most categorical analyses in this draft.

McClellan cited the uncorrected Crump et al. (1991) report in a more complete version of the quote in the comments: "The Crump et al. (1991) finding suggests caution in the utilization of the Garshick et al. (1988) finding of a weak association and most certainly suggests that the data should not be used to derived quantitative estimates of lung cancer risk for diesel exhaust exposure.".

This comment characterizes Dr. Moolgavkar's findings presented at a December 1996 conference in Germany in a way that is inconsistent with Dr. Moolgavkar's comments on the 1997 TSD. Dr. Moolgavkar's comments on the TSD encourage OEHHA to pursue further analyses of the Garshick et al. (1997) cohort data. This is inconsistent with the comment cited above that the Garshick data should not be used to derive quantitative estimates of lung cancer risk for diesel exhaust exposure.

Comment 4. Finally, Dr. Garshick himself in a letter to the EPA written over two years ago, cautioned against using his data to derive a unit risk factor. Dr. Garshick's caution should be presented in the agency's report and an explanation of the agency's view relative to that caution should be presented in the body of the assessment of human cancer risk and in the section on the feasibility of deriving a risk factor from currently available data.

***Response**: Dr. Garshick has not disavowed his study results or the underlying data. Rather, Dr. Garshick's comments largely address the uncertainties in the exposure history reconstructions and modeling assumptions which must reduce the confidence attached to any unit risks to be derived from his study data. OEHHA acknowledges that uncertainties as to the study population's exposure histories and the applicable modeling approaches must reduce the degree of confidence in the results. OEHHA has presented a broad range of unit risk values which capture much of the uncertainties due to the limited exposure data and choice of modeling assumptions. OEHHA also provides additional related information in its responses to Dr. Garshick's comments.*

Comment 5: When reviewing the findings of other parties, including researchers and effects assessment organizations, the report should be careful to note the relevant reports and research which were not available at the time of the finding and the extent to which a past finding has been retracted by the source.

The scientific community's perspective on the rat exposure studies and the Garshick railroad worker study has evolved significantly over the last ten years. As a result, it is important to place any findings by other individuals, agencies or health effects organizations, in careful perspective. For example, several parties have suggested that the IARC findings on diesel effects made in 1989 would be very different today. One researcher, Dr. McClellan has specifically withdrawn his past work on unit risk factors. Mercedes-Benz recommends that the agency contacts all parties whose past work will be cited to see if they still support those past findings and report their response in the final document.

***Response**: We have revised the report and the Executive Summary to include recent information provided by scientists in the field. In general, we have endeavored to provide such information in the health assessment document. However, IARC has not changed its conclusions that diesel exhaust is a probable human carcinogen. Whether or not it would change its position based upon the information available today is conjecture. There are numerous mutagenic and carcinogenic substances in diesel exhaust supporting biological plausibility of carcinogenic activity. There are numerous epidemiologic studies indicating significant association between diesel exhaust exposure and lung cancer risk.*

Where parties have retracted positions, it is important to address the underlying evidence and rationale. When study authors retract a finding or argue that there is limited or no application of their findings to a problem at hand, there is a strong presumption they may be right, particularly when the source of error lies largely within the body of their work. Here, however, according to Dr. McClellan's letter, the analyses of Dr. Crump have served as a premise for Dr. McClellan to recommend limited or no reliance upon the Garshick et al. (1988) cohort study in estimating the carcinogenic potency of diesel exhaust. OEHHA has closely examined the scientific data and been in communication with Garshick and Crump to discuss technical issues numerous times. Yet, in conducting its own analyses of the Garshick et al. (1988) cohort study and its own review of the related work by Dr. Crump, OEHHA has come to different conclusions regarding the dose-response of the Garshick et al. (1988) cohort study. Therefore, OEHHA continues to use the Garshick et al. (1988) cohort study data in our analyses to develop a plausible range of unit risk factors for diesel exhaust.

In the scientific process, authors whose work is cited are generally not contacted prior to inclusion of their studies in other scientific documents. In this case, OEHHA has made an effort more extensive than the usual scientific process to incorporate information from previous authors. Furthermore, OEHHA co-sponsored a scientific workshop in 1996 to bring the scientists together to discuss the issues. Thus, we believe that we have made an extraordinary effort to obtain information published by key scientists and any further information they may have.

Comment 6: There is currently no credible basis to derive a unit risk factor for either diesel exhaust or diesel particulates and the report should be revised to reflect that.

Mercedes-Benz has been involved in the research of diesel effects over the last ten years and continues to support significant health effects research in this area. Based on the results of the most current research, it is clear that the two pieces of research upon which the entire OEHHA draft relies are no longer valid. Research in the last five years has shown that rat cancer studies are not relevant to human exposures to ambient levels of diesel exhaust and that the Garshick data, when looked at objectively and from various perspectives, does not indicate a dose response relationship between diesel exposure and lung cancer incidents in railroad workers. As a result, the California EPA documents should be revised to present the current understanding of both of these data sources. In addition, the agency should not attempt to derive a unit risk factor when such a factor can not credibly be derived from all currently available information.

***Response**: After reviewing all the available information, OEHHA is providing a range of unit risks for humans from exposure to diesel exhaust based only upon the epidemiological evidence. This range captures the substantial uncertainties involved in the exposure data and modeling assumptions.*

As noted in the response to comments 1 through 5 above, OEHHA has expended much time and energy evaluating others' interpretations of the rat data and the human data. We agreed that the uncertainties in the mechanism of rat lung tumorigenesis limit its use in extrapolating to

humans. The uncertainty in such an extrapolation coupled with the availability of human data on which to base unit risk estimates has resulted in the OEHHA decision to focus on the human studies. We respond in the document as well as in responses to comments above and elsewhere to the issues raised by Crump in the analysis of the Garshick data with regard to the exposure-response relationship. We stand with our interpretation of the findings. We present a range of unit risks based on alternative exposure and modeling assumptions which we believe captures the uncertainties in the study populations' exposure histories.

Comment 7: Finally, Mercedes-Benz recommends that the agency revise its finding of carcinogenicity to acknowledge that current data are insufficient to classify diesel exhaust as a human carcinogen. The following chart summarizes the basic positions within the draft report and how they are contrary to the stated positions of researchers in each area and significantly undercut any attempt to set a unit risk factor or range.

OEHHA - Conclusions vs. Researcher's (Author's) Findings	
OEHHA's Conclusions	Author's Actual Findings
OEHHA considers Dr. Mauderly's work as the "most suitable study for QRA" [pp. 1-6 HRA paper, March 1997] interpreting it to indicate a relevance of rat test results to human beings.	Dr. Mauderly has gone on record stating that his research is not relevant for assessing effects in humans and that it would be bad scientific judgment to use such rat studies to even attempt to estimate a human risk.
OEHHA developed a QRA from the Garshick study (1988) by referring to an analysis by Dr. McClellan [p 1-6 HRA paper, March 1997] (in its analysis, OEHHA specifically relies on the prospect of chemical contribution in rat studies as the scientific basis for suggesting a dose-response relationship for Diesel PM and lung cancer in rats, consequently rejecting a potential threshold theory)	Dr. McClellan --has specifically studied lung cancer in rats after prolonged exposure to high concentrations of carbonaceous particles -- found that: "… organics on the diesel particle are not contributing to observed cancer in rats" and that ". . . the observed effects in rats appear to be a threshold phenomenon."
OEHHA concludes from the Garshick study that it allows for establishing a unit risk factor (i.e., dose-response relationship) for diesel exhaust	Dr. Garshick himself has taken the position that". . . it is not possible to use the human epidemiologic data that was re-analyzed to assign a unit risk with confidence." Dr. Crump, Dr. Moolgavkar, and Dr. McClellan share this position. EPA and HEI state that ". . .the Garshick study would not be useful for predicting human risk" [pp. 1-8 HRA 1997]

***Response**: The comment discusses the qualitative issue that diesel exhaust should not be considered a human carcinogen. The comment compares the statements in the OEHHA document with those of other individuals and organizations. However, the three statements selected from the OEHHA document for quotation do not pertain to whether or not diesel exhaust should be considered a human carcinogen. Instead, they pertain to the potential size of the cancer risk posed by diesel exhaust. Consequently, we will briefly address both the qualitative and quantitative issues generally raised in the comment.*

With respect to a qualitative relationship between occupational exposure to diesel exhaust and lung cancer, OEHHA's present conclusions can be compared to the conclusions of the USEPA, the HEI, and the WHO:

The March 1997 OEHHA draft states "The epidemiological studies concerning lung cancer risk and exposure to diesel exhaust provide evidence consistent with a causal relationship. The many associations found between lung cancer and diesel exposure are unlikely to be due to chance. Also, with the possible exception of the studies that did not take smoking into account, the findings reviewed above are unlikely to be due to confounding or bias. The results of various studies are consistent in the direction of an effect and are even somewhat similar in magnitude of effect. For example, all the studies of diesel railroad workers with adequate latency and more than 50 cases show evidence of an effect. Although the strength of the associations is weak, with low relative risk estimates being reported, several studies show clear exposure-response relationships. Finally, it is biologically plausible because of its mutagenic and carcinogenic constituents, that exposure to diesel exhaust would increase the risk of lung cancer. Therefore, a reasonable and likely explanation for the increased risks of lung cancer observed in the epidemiologic studies is a causal association between diesel exhaust exposure and lung cancer."

The 1994 USEPA draft document states "Collectively, the epidemiology studies show evidence of an association between inhalation of diesel exhaust and lung cancer in humans. Although, the evidence for carcinogenicity in humans was in most cases positive, it is judged to be limited according to the EPA's weight of evidence guidelines, because the observed increases in risk were quite low and the influence of confounding factors could not be completely accounted for." Similarly, the HEI concluded in 1995 that "The available evidence suggests that occupational exposure to diesel exhaust from diverse sources increases the lung cancer rate by 20% to 40% in exposed workers generally and to a greater extent among workers with prolonged or intense exposure or both. These results are not readily explicable by confounding due to cigarette smoking or other known sources of bias." While the WHO in 1996 found "The relative risks for lung cancer as a result of exposure to diesel exhaust are generally low, and risks of this magnitude are more susceptible to chance and to the effects of unmeasured confounding factors and imprecision for adjusting for known confounding factors. As discussed above, the elevated risk for lung cancer in the four most informative studies is unlikely to be due to confounding by cigarette smoking and is probably due to exposure to diesel exhaust. Other studies, although limited primarily by the exposure ascertainment, support this assessment."

With respect to dose response assessment, OEHHA does consider Dr. Mauderly's work "as the most suitable study for QRA" based upon rat data. However, following review of the available scientific information and input from the Scientific Review Panel, OEHHA is basing its range of unit risk only upon the epidemiological information. OEHHA is therefore not using the rat data to estimate risks in humans.

D*r. Mauderly's comments and OEHHA's responses to them are included elsewhere in Part C. With respect to a dose response threshold and the role of the adsorbed organics in the rat cancer response, Dr. Mauderly stated* "Meaningful increases in lung tumors in diesel soot-exposed rats only occur at exposure rates overwhelming particle clearance

defenses and inducing a strong prolonged and progressive inflammatory, and cell proliferative response. Although soot particles accumulate in small amounts at lower exposure levels, there appears to be a threshold exposure rate for triggering progressive lung disease in rats. The apparent threshold for induction of this progressive rat response is at least two orders of magnitude above the rates of environmental exposures to diesel soot. This threshold behavior is not characteristic of current models of chemical carcinogenesis; moreover, it has been shown that soot-associated organic mutagens are not important in the rat lung tumor response to diesel soot. Syrian hamsters and mice do not develop sustained cell proliferation or lung tumors at soot exposure rates carcinogenic in rats; thus it is well-proven that cancer risk estimates for these other rodent species cannot be derived from the rat data. Chronic exposure of nonhuman primates to diesel soot does not induce the cell proliferative response associated with development of lung tumors in rats. Although lifetime cancer studies of diesel exhaust have not been conducted in nonrodent species, there is no scientific basis for assuming that lung tumors would be induced through the same mechanisms producing tumors in rats, and there is a growing body of evidence that they would not.

The above findings do not prove that: a) there is no lung cancer risk for humans; b) that there is no lung cancer risk for humans from soot-associated organic mutagens; or c) that the risk for humans, if it exists, has a threshold. These are open issues that will have to be resolved on the basis of other information. The findings do indicate that human lung cancer risk from environmental exposures diesel exhaust, if it exists, almost certainly occurs by mechanisms different from those resulting in the rat lung tumors; thus, the rat lung tumor response is not an appropriate basis for quantitative estimates of human lung cancer risk. If this is true, and there is a growing consensus among the scientific community that it is, then it is true regardless of the numerical value, or intended use, of the estimates. It is simply inappropriate to generate human cancer risk estimates from the present rat data for any purpose. However, if the rat lung cancer data are not relevant to the assessment of human lung cancer risk (according to the quoted position of Dr. Mauderly), then the rat findings could not imply a threshold in human dose response."

With respect to Dr. Garshick's concerns regarding our use of his study, Dr. Garshick's comments and the OEHHA responses to them are more fully discussed elsewhere in Part C. Dr. Garshick has not retracted his studies. His comments reflect the absence of exposure measurements in his study. They also relate to the uncertainties in the exposure history reconstructions and modeling assumptions involved in performing a quantitative risk assessment using his data. It is important to note that the information referred to by Drs. Garshick, Crump, and McClellan specifically applies to the cohort analyses. OEHHA acknowledges that uncertainties as to the study population's exposure histories and modeling approaches must reduce the degree of confidence in the results. OEHHA has presented a broad range of unit risk values which capture the uncertainties involved in the exposure data and modeling assumptions.

To the extent to which the positions of the HEI, the USEPA, Dr. Garshick, and Dr. McClellan have been influenced by the reanalyses of the Garshick et al. (1988) cohort study which were conducted by Dr. Crump, the merit of those positions turns on the validity of the work by Dr.

Crump. In conducting its own analyses of the Garshick et al. (1988) cohort study and its own review of the related work by Dr. Crump, OEHHA has come to question the basis for their positions. Therefore, OEHHA continues to use the Garshick et al. (1988) cohort study data in our analyses to develop a plausible range of unit risk factors for diesel exhaust.

Comments of Dr. Suresh H. Moolgavkar,
received by Air Resources Board, August 18, 1997

Comment 1: Quantitative Cancer Risk Assessment. The risk assessments reported in the document are based both on empirical statistical approaches and on biologically-based modeling of experimental and epidemiologic data. I will restrict my remarks to the latter approach. Biologically-based approaches, based on multistage models of carcinogenesis, provide a rich and flexible family of incidence (hazard) functions for analyses of experimental and epidemiologic time-to-tumor data. In this approach, time- and age-dependent covariates, such as age at start of exposure, age at end of exposure, detailed patterns of exposure, can easily be accommodated in analyses for single or multiple exposures. This is of particular importance in analyses of the Garshick railroad cohort data, in which different methods for controlling age-related covariates in empirical statistical models have led to radically different interpretations of the data with regard to the carcinogenicity of diesel exhaust. In addition to the ease of incorporation of time-related factors, biologically-based analyses may offer insights into mechanisms of action of the putative carcinogenic agent under investigation. CALEPA is to be commended for undertaking biologically-based analyses of the Garshick data in response to the comments made at the San Francisco workshop held in 1996. I believe, however, that the current analyses presented in the document do not exploit the full power of this approach. In particular, failure to explore carefully the shape of the exposure-response curve could have led to biased estimates of the unit risk for diesel exhaust.

A. Experimental Data. The document describes analyses, using the two-mutation clonal expansion (MVK) model, of lung cancer in rats following exposure to diesel exhaust. Time-to-tumor analyses require information on tumor lethality. Generally, tumors are classified as either being rapidly fatal or incidental (do not cause death of the animal). The construction of the likelihood function for statistical analyses depends upon which assumption is adopted. Rat lung tumors are generally considered to be incidental. The document does not explicitly state whether the assumption of incidental tumors was made for the analyses. From the description in the document it appears as if the assumption of incidental tumors was used, which is appropriate for these data.

There are a number of ways in which the analyses could be improved. The single most important deficiency of the analyses is that a linear effect of diesel exhaust was assumed for promotion. There is considerable evidence of non-linearity (threshold-like behavior) in this data, and a thorough exploration of the shape of the exposure-response curve should be undertaken. Moreover, the analyses should not assume that diesel exhaust acts as a promoter. Rather, various assumptions regarding the action of diesel exhaust (initiation, promotion, or both) should be tested within the framework of the two-mutation clonal expansion model.

The CALEPA analyses used an approximate form of the hazard function of the two-mutation model. The use of the approximation could lead to biased estimates of the parameters. The exact solution is available and has been widely used for the analysis of data. I would suggest that CALEPA repeat the analyses using this solution. Finally, the document does not make it clear how confidence intervals were computed. The usual method based on standard errors derived

from the Information Matrix, which depend upon asymptotics, may yield intervals with poor coverage properties in small data sets, particularly with complicated likelihoods. Since the ultimate goal of the analyses is to obtain estimates of unit risk (which is a function of the model parameters) together with the confidence interval it would be best to use methods that sample the likelihood directly, such as Markov chain Monte Carlo. This procedure allows confidence intervals for derived statistics to be constructed without appealing to the delta method, which is a first order approximation and also dependent upon asymptotics. In order to check on the asymptotics, at the very least, profile likelihood based intervals should be constructed for the parameter estimates and compared with those based on standard errors.

***Response**: Following public and peer review, including comments made by the Scientific Review Panel, OEHHA has decided not to include the unit risk estimates from the rat lung tumor data in the final estimate of risk to humans from diesel exhaust exposure. While the commentator's suggestions are interesting, we cannot justify expending further resources on the analysis of the animal tumor data at this point. We think that results generated by the more sophisticated analyses would not differ substantially from the current range of risk for animal.*

In regard to the comment on tumor classification, we noted under Table 7.1, page 7-30 that all tumors were considered to be incidental.

We have clarified the method for determination of confidence interval in the text. The current draft TSD now explains that the confidence intervals cited in the results of the calculations are determined by likelihood ratio test. See, for example, ICF Kaiser (1993).

Comment 2: Epidemiologic Data. In response to comments made at the San Francisco workshop in 1996, this revised CALEPA document presents analyses of the Garshick data based on the Armitage-Doll multistage model. There are a number of ways in which these analyses could be extended and improved. As in the analyses of experimental data, the CALEPA analyses of the Garshick data set makes the assumption that the exposure-response relationship is linear. Perhaps the single most important task that should be undertaken is a careful exploration of the shape of the exposure-response curve.

Although information in the Garshick cohort is available on an individual basis on each member of the cohort, the CALEPA analyses were based on cross-tabulated summaries of this detailed information. One of the advantages of using biologically-based models is that such a collapsing of the data is not required. Individual level information can be explicitly considered. Since human malignant lung tumors (in contrast to malignant lung tumors in the rat) are rapidly fatal, any statistical time-to-tumor analysis requires the density and survival functions of the time-to-tumor distribution. In particular, these need to be known (or derived) for any biologically-based model that is to be used for the analyses. Moreover, since exposure in the Garshick cohort is time-dependent, these quantities need to be derived with time-dependent parameters. The EPA approach, which uses cross-tabulated data and Poisson regression, requires the hazard function (with time-dependent parameters), which is available from the published literature, but only as an approximation to the exact hazard function. My suggestion would be to use the exact solutions, whether cross-tabulated data (requires the hazard function) or individual level data (requires the

density and survival functions) are being analyzed. For reasons given below, I would prefer to analyze the data using the two-mutation clonal expansion model.

Given the composition of diesel exhaust, it is reasonable to assume that, if diesel exhaust is a human carcinogen, then it has both initiating and promoting activities. Within the context of the Armitage-Doll model this means that at least two stages could be affected by diesel exhaust. The CALEPA analyses assume a seven stage Armitage-Doll model, with one stage linearly affected by diesel exhaust. I believe that these analyses should be extended to consider two or more stages affected by diesel exhaust and with non-linear effects of diesel exhaust on the transition rates. With a seven stage model a comprehensive analysis is a formidable task because of the large number of possible combinations of two or more stages that could be affected. Moreover, I believe that it is biologically more realistic to model promotion as the clonal expansion of initiated cells. Therefore, I believe that the Garshick data set should be analyzed using the two-mutation clonal expansion model. Within the framework of this model, the effect of diesel exhaust could be tested on the rates of the two mutations (initiation and conversion) and on the rate of clonal expansion of initiated cells (promotion). Further, the shape of the exposure-response curves for these effects should be explored. Traditional statistical analyses (reported in the document) indicate that birth cohort effects are important in these data. I believe that it is important to incorporate birth cohort effects in biologically-based analyses of these data as well, particularly because smoking information is not available. Birth cohort effects are surrogates, albeit crude, for smoking effects. Even if smoking is a balanced covariate (i.e., has the same distribution in exposed and unexposed subcohorts) in these data, it may well be an effect modifier. The comments made above regarding the construction of confidence intervals apply here as well, i.e., either MCMC or profile likelihood based methods should be used for construction of confidence intervals. Finally, the estimate of unit risk should be adjusted for competing causes of mortality. An example of such an adjustment and details of the procedure can be found in the USEPA's 1984 risk assessment of coke oven emissions.

In summary, I would suggest the following: 1) Analyze the Garshick cohort using the exact solution of the two-mutation clonal expansion model and individual-level information, 2) test for effects of diesel exhaust on the parameters of the model and explore the shape of the exposure-response curves; 3) test for the effects of birth cohort in this model, and incorporate birth cohort effects if they appear to be important 4) estimate unit risks without and with adjustment for competing causes of mortality, 5) construct confidence intervals for unadjusted and adjusted unit risks using MCMC methods.

***Response**: With respect to the summary:*
1) The analysis of the Garshick et al. cohort using the exact solution for the two-mutation clonal expansion model and individual information would be a large undertaking. Apparently no one has taken on such an analysis. Results from such an analysis would provide a useful extension of and check on the present results, which already provide a substantial range of risk. However, the large undertaking seems unwarranted for this stage of the risk assessment because it would undoubtedly delay the identification process substantially and seems unlikely to provide very different risk numbers. The greatest uncertainty is not in the choice of model, but in the exposure. These risk numbers would likely be near the values for the same exposure pattern in

the TSD's approximate 7-stage model with a very late stage dependent on diesel exhaust. This is because the effect in the clonal expansion model would probably be dominated by the second (final) stage and thus have mathematical characteristics close to those of the 7-stage model with a very late stage dependent on diesel exhaust.

2a) Description of the effect of diesel exhaust on the parameters of the clonal expansion model could be very interesting scientifically, but the specific value of the parameters does not have a clear bearing on the quantitative risk assessment.

2b) The TSD explores the shape of the exposure response curves by using the categorical analysis for the approximate 7-stage model, with results depicted in Figures D- 4, D-5 and D-6 of the revised draft TSD.

3) Using the approximate 7-stage model, the analysis in the revised draft TSD now tests for the effect of birth cohort, which is equivalent to age at start of study. The deviance test shows the effect to be marginal, and the risk relationship is little changed. The results are now presented in the revised draft TSD, Appendix D.

4) The unit risks are now all calculated using a standard California life table for lung cancer, taking competing causes of mortality into account. The comment does not give a reason for estimating unit risks both with and without adjustment for competing causes of mortality. For simplicity in the face of many alternative calculations, we consider one of the two forms to be sufficient, and the one adjusted for competing causes appears most useful to risk managers.

5) The TSD reports confidence intervals based on standard Wald statistics (standard error). These values are asymptotic approximations; so the values have been spot checked by likelihood ratio test and found to agree closely. Both methods are part of the AMFIT program in EPICURE. Markov chain Monte Carlo (MCMC) methods would add a degree of sophistication to the work but have not been used in the work because they would require extensive reprogramming and because the agreement of the Wald statistics with the likelihood method is assumed to offer sufficient assurance of the approximate correctness of the values for confidence intervals.

Comments of Dr. Suresh H. Moolgavkar in letter dated September 25, 1997 to Genevieve Shiroma

Comment 3: From the reports I have had of the September 16 meeting on diesel exhaust, I believe that my comments on the meta-analysis contained in the Cal EPA report "Proposed Identification of Diesel Exhaust as a Toxic Air Contaminant" have been misunderstood. Let me clarify. I believe that the meta-analyses were technically well conducted. In particular, a number of important issues, such as publication bias, were thoughtfully addressed, and the statistical methods used were sound. I point out, however, that no meta-analysis can correct for the deficiencies of the individual studies, which remain a real concern with epidemiologic studies of diesel exhaust. In particular, meta-analyses cannot correct for biases that may have arisen through inadequate control of confounders in individual epidemiologic studies. I also noted that some of

the results of the meta-analyses were rather unexpected. For example, the level of risk in different occupational categories was rather similar, which is surprising in view of the different levels of exposure to diesel exhaust in the different occupations. Finally, since there is no dose-response information in the meta-analyses, it would be totally inappropriate to use these analyses for quantitative risk assessment.

Response: *The commenter is correct that a meta-analysis cannot correct for individual study deficiencies. In fact, one of the purposes of meta-analysis is to explore a body of research to identify which study characteristics generate heterogeneity in the pooled results. For example, in the meta-analysis, the pooled estimates for the cohort studies contained significant heterogeneity. The presence of such heterogeneity undermines the validity of the pooled risk estimate. When stratified on the presence or absence of a healthy worker effect (HWE - one form of selection bias), however, those studies with a HWE continued to show heterogeneity, while those without HWE showed a dramatic reduction in heterogeneity, along with a large increase in the estimate of the relative risk. So, while the meta-analysis could not correct the bias involved in those studies characterized by a HWE, it was able to identify that the HWE was a source of significant heterogeneity that produced lower pooled risk estimates.*

Though the pooled level of risk is similar in different occupational categories, historical exposure data, and exposure patterns could not be found or estimated for all populations in question. That is, in some industries, recent and past estimates may be available, while in others only recent estimates may be available. In some cases exposures may be expressed as elemental carbon while in others they may be expressed as respirable particulate. To make a useful comparison of exposures and relative risks among industries would require an analysis beyond the scope of this health assessment. Thus, while one might intuitively postulate that the exposures were likely to have been quite different, this is not possible to verify. Moreover, given the diversity of occupations, the potential for misclassification of exposure, differences in length of follow-up (for cohort studies), the presence of various biases, confounders, and effect modifiers across studies, it should not be surprising that an exposure-response relationship corresponding to one's intuitive notion of lighter versus heavier diesel exposures is not starkly evident.

OEHHA will not be using the meta-analysis as the basis for quantitative risk assessment. Rather, the pooled relative risk estimates for the epidemiological studies that adjusted for smoking have been used, in conjunction with the upper and lower bounds of the estimated range of historical exposures in diesel exhaust-exposed populations, in order to provide a basis for comparison with the quantitative risk assessment developed using the Garshick et al. (1988) study. The exposure analysis was conducted for us by Dr. Hammond of U.C. Berkeley. The information she provided focused on those occupations considered in the smoking-adjusted pooled analysis, and the exposures were adjusted to a single exposure metric.

Comments on behalf of the American Health Foundation, from Joshua Muscat, dated July 23, 1997.

In this submission, comments were limited to the meta-analysis of occupational diesel exposure and lung cancer.

Comment 1: "Multiple effect measures were published in several individual studies. The effect measure(s) chosen for the meta-analysis was not the most relevant one in several instances.

a) Only elevated SMR were listed in Table I of the report by Menck and Henderson (1). The CARB/OEHHA report selected the effect measures for truck drivers and mechanics from this table. Cranemen were not listed in the table, but the text describes a decreased SMR for cranemen of below 70. Clearly, the SMR for cranemen should be incorporated into the meta-analysis."

Response: *The commenter is correct that only statistically elevated SMRs (standardized mortality ratios) are presented in Table 1 of the paper by Menck and Henderson, and that the text indicates that the SMR for cranemen was below 70, as were estimates for 15 other occupations. However, the text gives neither a precise value for the cranemen SMR nor any indication of data sufficient to calculate a standard error for the estimate (i.e., a confidence interval or a notation of the statistical significance of the SMR). Without this information, one cannot incorporate the cranemen's mortality experience into either the fixed- or random-effects models used in the meta-analysis. Therefore, OEHHA staff respectfully disagree with the commenter on this issue.*

Comment 2: "b) Coggon *et al.* ... reported an unadjusted risk of 1.3 (95% CI 1.0-1.6). However, occupations classified as having a high exposure to diesel showed a unadjusted risk of 1.1 (95% CI 0.7-1.8). This latter measure was not chosen for the meta-analysis although the criteria for end-point selection was "the highest level of exposure." The Coggon report was also one of the few studies where the degree of exposure was classified by an industrial hygienist. The effect measure of 1.1 should be substituted for the effect measure of 1.3 based on the criteria established by CARB/OEHHA.

Response*: The Coggon study is a case-control study of lung cancer occurring in males younger than 40 years, in which death certificates alone provided information on the decedents' last occupation before death. An industrial hygienist then classified the occupational information by likelihood of exposure to diesel exhaust and a variety of other exposures. There was no information on duration of exposure in any given occupation. This method is likely to result in substantial misclassification of exposure, which would tend to produce a bias towards the null hypothesis of no effect. The estimate used in the OEHHA analysis involved the mortality experience of 453 individuals, whereas that suggested by the commenter was based on only 89 deaths, which will tend to downweight the influence of the Coggon study in the pooled RR estimates. However, in the recalculation of the pooled estimates undertaken since the last draft, OEHHA staff have used the estimate suggested by the commenter.*

Comment 3: "The effect measure selected from the study by Boffetta (3) using American Health Foundation data is based on ≥ 30 years of diesel exposure (OR, 1.49 (CI 0.7-3.1) although the meta-analysis protocol is based on ≥ 20 years of diesel exposure. The OR for 16-30 years of diesel exposure in the Boffetta study, which presumably included mostly 20-30 years of diesel exposure and was based on larger numbers of subjects, is 0.7 (0.34-1.44). The more appropriate analysis would therefore be to combine the 0.7 and 1.49 risk estimates. Alternatively, the American Health Foundation agrees to reanalyze the data for CARB/OEHHA for subjects exposed only to ≥ 20 years of diesel."

Response: *OEHHA staff contacted the commenter, who has left the American Health Foundation and was unable to reanalyze the data as indicated in the comment. Though he indicated that the stratum with 16-30 years of exposure was based on larger numbers of subjects than the 30+ years stratum, the actual data (from Table V of the Boffetta (1990) report) indicate that there were 15 cases occurring in 25 subjects in the former stratum and 17 cases in 19 subjects in the latter. To be consistent with the data extraction procedures used throughout the rest of the meta-analysis, which involved using the estimates derived from the group with the longest or most intense exposure to diesel exhaust, OEHHA staff left the estimate from this study as is. However, for several sensitivity analyses we pooled the data from these strata as suggested by the commenter. This resulted in minor modifications of the pooled risk estimates. For example, the pooled estimates for the case control studies went from 1.44 (95% C.I. = 1.33-1.56) using only the 30+ years stratum, to 1.42 (95% C.I. = 1.30-1.55) using the combined strata (random effects model).*

Comment 4: In occupational epidemiology, when comparing the rates of disease in a target population to a referent population, the use of national rates as the referent group can be grossly inappropriate. Local geographic rates provide a more appropriate reference (4). Gustafsson (5) reported an increased rate of lung cancer in dock workers compared to all Swedish men (70 observed, 52.9 expected). This yields a risk of 1.23. The more appropriate comparison is based on the expected number of deaths in the counties of the docks. Here the expected number of deaths is 55.3. We recommend that the effect measure be recalculated using 55.3 deaths as the expected number.

Response: *Table 2 of the report by Gustafsson et al. indicates that the SMR for lung cancer among Swedish dock workers compared to the Swedish male population was 1.32 (95% C.I. = 1.05-1.66) (not 1.23, as indicated by the commenter). Table 2 also indicates that all-cause mortality for dock workers was significantly lower than that for the Swedish male population: SMR = 0.89 (95% C.I. = 0.84- 0.94). The latter is an indication that the estimates in this study showed a clear healthy worker effect (HWE), which was one of the study characteristics used in the meta-analysis. Table 3 of this report provides observed and expected numbers of lung cancer cases in the 17 counties and city regions in Sweden in which the docks were located. The latter table provides no confidence intervals or p-values, but one can use Byar's approximation to calculate a confidence interval around the point estimate. Doing so indicates that the SMR for lung cancer would be 1.27 (95% C.I. = 0.99 - 1.60). However, Table 3 does not provide an estimate for all-cause mortality based on regional rates and, therefore, were we to include this estimate in the analysis, any subset analysis involving the presence or absence of the HWE could*

not include the Gustafsson report. Although the SMR for lung cancer using the regional rate is about 3.8% lower than the estimate using national rates, which is enough of a change to affect the statistical significance of the estimate, the difference between the two does not appear large enough to warrant losing the HWE subanalysis. However, in some of the other subset analyses, we have included the lower estimate for comparison purposes. This resulted in minor changes in the risk estimates and confidence intervals: For instance, dropping the Gustafsson study from the pooled estimate for the cohort studies using national lung cancer rates for comparison changed the random effects pooled estimate from 1.14 (95% C.I. = 1.00-1.31) to 1.11 (95% C.I. = 0.96-1.29). Incorporating the regional Gustafsson relative risk estimate to the pooled estimate for the cohort studies using regional or state rates for comparison changed only the confidence interval (0.83 - 2.39 to 0.83 - 2.36), while the point estimate of 1.40 remained unchanged.

Comment 5: It is unclear how an RR of 2.39 for transportation equipment operators was derived from the study of Wegman and Peters (6). Using Table 3 of that study, an unadjusted RR of 1.26 is calculated for this group. We request clarification of this calculation.

***Response**: Using only Table 3, which provided occupational history groupings used by the Bureau of the Census, the commenter is correct that the unadjusted RR estimate is 1.26. However, our estimate was calculated based on information not only from Table 3, but also from Table 4. Because the Bureau of the Census occupational groupings are not intended for the purposes of determining work exposures, the authors of this report undertook a review of the study subjects' job classifications without regard for case/control status. Their review resulted in changes in the redesignation of several transportation operatives in Table 4. Combining data from Tables 3 and 4 resulted in the following 2 x 2 table:*

	Cases	*Controls*
Exposed	*9*	*4*
Unexposed	*82*	*87*

This table yields an odds ratio of 2.39 (95% C.I. = 0.71 - 8.05, calculated by Woolf's approximation).

Comment 6: Several studies with null findings were excluded from the analysis because of insufficient latency or follow-up. The criteria for exclusion as defined by OEHHA is a latency of less than 10 years. The study by Kauppinen *et al*. (7) of Finnish woodworkers was included in the analysis. The authors calculated a smoking-adjusted risk associated with diesel exposure of 2.21. In a number of calculations, the authors allowed for an induction period of 10 years when available. However, for the analysis of diesel-exposed workers, the study clearly states that the authors did not allow for the 10 year latency. The results from this should be excluded from the meta-analysis based on CARB/OEHHA criteria.

Response: *We agree with the commenter, and have excluded the Kauppinen study from the revised meta-analysis. It should be noted, that some of the sensitivity analyses in the earlier version of the meta-analysis also excluded the Kaupinnen study, which made little difference in the results. However, the commenter is correct that the estimate extracted from this study was characterized by inadequate latency; thus, the appropriate course of action is exclusion.*

Comment 7: Unadjusted risk estimates yield lower pooled estimates than smoking-adjusted risk estimates. Presumably, the unadjusted estimates are confounded by smoking. For example, Balarajan (8) found a significant increased SMR of lung cancer for lorry drivers but also a significantly elevated SMR for asthma, bronchitis and emphysema. The unadjusted risk estimates calculated by Howe (9) were presumably confounded by smoking because the cohort experienced higher than expected death rates from other smoking-related cancers and emphysema although not bronchitis. While smoking-adjusted estimates are considered higher quality if the risks are elevated, the fact that smoking-adjusted risk estimates yield higher risks than smoking adjusted risks (sic) could also be interpreted as a systematic bias in an upward direction.

Response: *The commenter implies that adjustment for smoking should result in a lower RR estimate for lung cancer, and where this does not occur, there may be some other unidentified systematic bias. While such a speculative scenario is certainly possible, it is clearly not necessary. For example, if the diesel-exposed population had a lower prevalence of smoking than the control or reference population, adjustment for smoking would result in a higher estimate of diesel exhaust-related relative risk. If the proportions of smokers, ex-smokers, and nonsmokers in the exposed and reference populations are relatively close, then adjustment for smoking would not have much of an effect on the estimates of relative risk. Thus, it is unnecessary to posit some other unspecified bias merely because some of the smoking-adjusted pooled risk estimates in the meta-analysis are higher than the unadjusted estimates,*

Comment 8: Publication bias. This is acknowledged by OEHHA although no formal attempt has been made to investigate sources of negative findings or sources not within the 3 electronic databases. Included in the meta-analysis was the study by Swanson *et al*. (10) which showed increased risks in male drivers in the Detroit Metropolitan area. Using the same database to examine occupation and lung, cancer incidence in women but not included in the meta-analysis, the authors found no smoking-adjusted association between driving occupation and increased lung cancer incidence (II). The numbers of drivers are not stated but the lack of an association is clearly not due to small numbers of drivers since a significant positive association was observed between drivers and eye cancer, a very rare tumor. The second study is significant in that it is the only published study of diesel exposure and lung cancer in women and perhaps deserves separate mention.

An analysis of mortality patterns by occupation in US Veterans was based on 6369 lung cancer deaths (12). The SMR's used internal comparisons to reduce the healthy worker effect. No excess smoking-adjusted lung cancer deaths were found for railroad conductors (n= 233), railroad and car shop mechanics (n= 192), railroad brakemen (n= 208), railroad switchmen (n= 255), cranemen, derickmen, hoistmen (n= 119), carpenters (n= 510), automobile mechanics and

repairmen (n= 2327), other mechanics (n= 2937), and firemen (n= 902). Excess risks were observed for locomotive engineers (n= 629, RR= 1.9) and locomotive firemen (n= 86, RR= 2.6). Engine exhaust or asbestos exposure may have accounted for the increased risks in locomotive workers (12). Other occupations with increased lung cancer rates were mine operatives (n= 918, RR= 1.4) and truck and tractor drivers (n= 1453, RR= 1.4). The Veterans Study includes large numbers of diesel-exposed workers. Exclusion of this and other data may have biased the overall meta-analysis.

Response: *Publication bias refers to the increased likelihood of publication of statistically significant results compared to nonsignificant or null results, which may potentially distort a pooled risk estimate. Publication bias is generally attributed to journal editorial policies that prefer "positive" results, so that small, statistically insignificant studies are less likely than large, statistically insignificant studies to be published or even submitted for publication. One way to assess graphically whether publication bias is likely to have affected the results of a meta-analysis is to construct funnel plots of the logarithms of the relative risk (log RRs) versus sample size. If there is no publication bias, the plot should resemble an inverted funnel with the apex located approximately over the mean log RR. Publication bias is discussed on pp. D-7, D-10 and D-11 and addressed graphically by Figures D-6 and D-7 of the March 1997 draft.*

In the revised version of the OEHHA meta-analysis, we have modified how publication bias is addressed. First, we have included one funnel plot combining the results for case-control and cohort studies by plotting the inverse of the standard error versus the logarithm of the individual study estimates of relative risk. As Pettiti (1994) notes, plotting such a measure is preferable to plotting total sample size for case-control and cohort studies, as the latter metric does not provide information about the variability of the effect measure. This plot, as was true of the separate plots by study design, also indicates a relatively less dense distribution of individual study estimates in the lower left corner of the graph. Second, we have addressed the issue of publication bias with a separate sensitivity analysis, examining the effect of pooling progressively larger studies (i.e., with smaller standard errors). Finally, there is a methodological paradox in the commentator's implications about the effects of publication bias. The implication is that there should be a roughly equivalent number of null or negative studies, but for the past editorial policies favoring publication of "positive" studies. However, there are many reasons why specific studies do not get published. Inadequate sample size in a given study decreases the power to detect an association between an exposure and a health effect: this represents a serious study-design flaw. Thus, in a sense, it is not clear that underpowered studies, particularly those that are not published, should be accorded the same weight as studies that have passed the scrutiny of peer review.

The U.S. Veterans Study referred to by the commenter is, however, a large study in which a variety of diesel-exposed occupations are represented. This is the only such study that OEHHA staff members are aware of. We were unable to obtain a copy of this report locally and therefore ordered it from the U.S. government. A copy of this report has arrived recently, but too late for supplemental analyses to be included in these responses to comments. However, we will conduct such analyses prior to the meeting of the Scientific Review Panel in April 1998.

Comment 9: In summary, although the patterns of relative risk in the meta-analysis are generally elevated, this is partly due to inappropriate choice of effect measure selected from some individual studies and possibly publication bias. Because lower risk estimates are found in studies that did not adjust for cigarette smoking than in studies that did adjust for smoking, these findings are inconsistent with a causal inference of diesel exhaust and lung cancer.

Response: *See responses to prior comments of the American Health Foundation. Calculations in the meta-analysis have been re-done incorporating some of Mr. Muscat's suggestions, which has resulted in minor adjustments to the risk estimates. The basic conclusions of the meta-analysis remain unchanged, however. The issue of the effects of adjusting for cigarette smoking has been addressed in the response to his comment # 7, above.*

Comment 10: Validity of risk-assessment using the data of Garshick *et al*. Workers exposed to high concentrations of industrial polycyclic aromatic hydrocarbons have increased rates of lung cancer. However, studies of workplace atmospheric measurements of polycyclic hydrocarbons levels show no correlation with various measures of biological exposure. For example, polycyclic aromatic hydrocarbons were measured in an aluminum reducing plant. The mean inhalation in an 8-hour shift was estimated at 1000 μg (13). However, the amount excreted in the urine was only about 2 μg. In contrast, smokers of 20 cigarettes inhale about 1 - 4 % of this amount but excrete twice as much. While the polycyclic aromatic hydrocarbon-DNA adduct levels in white blood cells from smokers are significantly higher than in nonsmokers, no correlation was found between PH-DNA adduct levels with PAH-concentrations in the workplace atmosphere of several industries (14). Similarly, the ambient measures made by Garshick *et al*. (15) may have little relationship to the bioavailability of diesel particulates in the lung. Industrial PAHs such as diesel are adsorbed onto particles such as carbon black and are not readily bioavailable. If PAH's or nitro-PAHs are the presumed carcinogenic component of diesel, ambient measures may not reflect exposure to target organs.

In conclusion, although the Garshick *et al*. study is recognized as the study with the most accurate ambient exposure measurement, it provides no information on the relationship between biological dose of diesel exposure and lung cancer risk. Because the meta-analysis of studies of diesel and lung cancer, at least as presented, do not provide convincing evidence (that is, beyond the point where findings can be explained by study biases, publication bias) of a carcinogenic effect of diesel, risk assessment based on any one individual study may be insufficient to draw any conclusions. It should be noted that the airborne asbestos health assessment conducted by the Environmental Protection Agency (16) and other groups was based on up to a dozen studies of asbestos-exposed cohorts.

Response: *The comment implies that the PAHs in diesel exhaust are not bioavailable. However, information on genotoxicity studies, measurements of PAH metabolites in urine, and presence of DNA adducts in animals and humans exposed to diesel exhaust indicate that PAHs in diesel exhaust are bioavailable.*

The data discussed in Section 5.4 of the OEHHA document indicate that workers in occupations which involve maintenance or use of diesel-powered equipment may have increased levels of

lymphocyte DNA adducts, that such increases can be demonstrated in workers without likely dermal exposure to either diesel fuel or used lubrication oil, and that lymphocyte hprt mutations may be associated with increased lymphocyte DNA adducts. These data support the results of epidemiologic studies which describe a positive correlation between human diesel exposure and the induction of lung cancer. In addition, several studies of rats and one in monkeys exposed chronically to diesel exhaust by inhalation discussed in Section 5.4.1 demonstrate elevated DNA adducts in the lung. Finally, in vitro exposure of rat tissue to diesel exhaust particles induces unscheduled DNA synthesis.

Kanoh et al found increased levels of urinary 1-hydroxypyrene in rats exposed by inhalation to diesel exhaust relative to controls. Scheeper et al. (1994) reported that although not statistically significant, urinary 1-AP levels obtained from single day urine collections were consistently 1.7 - 2-fold higher for diesel train-engine mechanics compared to office clerks. These data strongly suggest that PAHs and nitroPAHs contained in diesel exhaust particulate matter may be bioavailable in humans.

Section 5.1.2.6 describes attempts to determine if data from in vitro genotoxicity tests concerning bioavailability of the genotoxic component of diesel exhaust can be generated which would aid in determining if in vivo genotoxicity occurs as a result of exposure to diesel exhaust. Several investigators (Brookes et al., 1981; King et al., 1981; Siak et al., 1981; King et al., 1983) found that extraction of diesel exhaust particulate matter with simulated physiological fluids such as saline, bovine serum albumin, dipalmitoyl lecithin and fetal calf serum resulted in little or no mutagenic activity being present in the extract supernatant after filtration. However, it should be noted that King et al. (1981) also found that excitation and emission fluorescence spectroscopy data indicated that incubation of diesel exhaust particulate matter with both serum and lung cytosol extracted a substantial portion (79 - 85%) of the solvent-extractable mutagens. Although the serum-associated mutagens did not induce significant mutagenicity in Salmonella, incubation of the serum with protease increased the mutagenic activity of the serum, suggesting that the serum-extracted mutagens were bound to proteins and therefore unavailable to bind to Salmonella DNA under the assay conditions used by the authors. Sun et al. (1988) stated that the studies by Brooks et al. (1981) and King et al. (1981, 1983) "suggest that particle-associated organics become "bioavailable" to respiratory tract cells, allowing metabolic processes to occur".

Additionally, direct exposure of Salmonella to a diesel exhaust stream resulted in mutation induction (Courtois et al., 1993). Finally, diesel exhaust particulate matter suspended in dipalmitoyl lecithin, a major component of pulmonary surfactant, also induced mutations in both Salmonella and mammalian cells (Wallace et al., 1987; Keene et al., 1991; Gu et al., 1992). These studies indicate that solubilization of the genotoxic component of diesel exhaust particulate matter is not required for that component to exert a genotoxic effect in in vitro test systems, and suggests the same for in vivo genotoxicity.

Comments from Natural Resources Defense Council, provided July 1, 1997 at the California Air Resources Board Presented by Janet S. Hathaway, Senior Attorney with assistance from Todd R. Campbell, Science and Policy Analyst

Comment 1: The Cal-EPA/ARB 1997 study probably understates the cancer risk associated with diesel exhaust. IT is critical to ensure that the risk from diesel exhaust is rigorously evaluated. Cal EPA and ARB have done an outstanding effort to account for the cancer risk diesel poses. Cal EPA/ARB have been scrupulous to avoid any exaggeration of the risk. However, it is equally important not to understate the magnitude of the public health risk posed by diesel exhaust. There are many factors NRDC raised in comments on the 1994 draft (most of which Cal EPA/ARB has acknowledged in the 1997 report) which make this proposed risk assessment conservative - that is, this report likely understates the cancer risk posed by diesel exhaust in California.

In the context of a debate that may be dominated by industries with financial interests in retaining current uses of diesel, it is important to reiterate: there are many reasons to conclude that the 1997 risk assessment probably underestimates the public health risk from diesel exhaust.

Cal EPA and ARB report a range for the diesel exhaust risk for exposure to one microgram of diesel exhaust particulate matter per cubic meter (this is termed the "unit risk." but it is only one-third of the average ambient level of diesel exhaust in California) inhaled over a lifetime of from 10 to 2000 excess per million people exposed. The low end of the range is derived from animal (rat) data. NRDC urges that the higher end of the range, which is derived from worker studies, is enormously more relevant to human cancer risk.

Using the worker studies' diesel potency estimate, together with ARB's current calculation for time-weighted population exposure to diesel in 1990 at 2.2 microgram diesel particles per cubic meter, and the current California population of approximately 34 million people, we can calculate the excess cancer posed by diesel exhaust for the state. That result is a staggering 150,000 lung cancer cases just in the state of California over a 70 year human lifetime. This number indicates the magnitude of the task before us in reducing diesel risk. This stark number, one hundred fifty thousand usually fatal lung cancers, only hints at the enormous human tragedy due to diesel exposure. In fact, the real-world cancer risk from diesel exposure is likely larger than that estimated in the careful study done by Cal EPA/ARB.

***Response**: Following public and peer review, including suggestions from the Scientific Review Panel, OEHHA is now basing the range of risk estimates only on the epidemiological studies.*

Comment 2: Are diesel particle concentrations adequate surrogates for the cancer risk of whole diesel exhaust? Whole diesel exhaust is composed of both gaseous and particle phase compounds. The gas, or vapor, phase contains typical combustion products, including compounds such as aldehydes (e.g., formaldehyde and acetaldehyde), and aromatic compounds (e.g., benzene, 1,3-butadiene, polycyclic aromatic hydrocarbons (PAHs) and PAH-derivatives), many of which are probable or known carcinogens. A wide spectrum of gas- and particle-phase

PAH and PAH-derivatives are emitted in diesel exhaust. More than 50 nitro-PAH have been identified in diesel exhaust. The nitro-PAH compounds are recognized mutagens.

The 1997 diesel risk assessment for diesel exhaust is based on the presumption that diesel particle concentrations serve as an adequate surrogate of risk from whole diesel. In NRDC comments on the 1994 draft, NRDC suggested that the vapor phase might increase the potency of the cancer risk associated with diesel particles, and that it would be prudent to assume additivity of the vapor and particle risks while investigating possible synergy. OEHHA now acknowledges that assuming particle risk adds to the risk of the vapor phase would be proper if exposure were based on including gaseous diesel components as well as particle components. That has not been done, apparently because of the costs and complexity involved in collecting adequate data on the exposures to the vapor-phase carcinogens from diesel exhaust. However, in light of the many hazardous constituents of the vapor phase, it is reasonable to assume that future refinements of this diesel risk assessment which could include the vapor phase risk would increase the total risk associated with exposure to diesel exhaust.

Further investigation is needed concerning possible synergy of gaseous diesel components (and other air toxics) with diesel particles. NRDC is pleased that diesel speciation studies have been undertaken at the University of California, Riverside and the University of California, Davis. These studies will further our understanding of the complex mixture which results from diesel combustion. However, these studies, which will help identify the constituents of diesel combustion using different engines and fuels, are only a beginning. When considering a complex, variable mixture such as diesel exhaust, synergy between the constituents deserves further study.

Response*: The comment makes the point that we are not considering the carcinogenic effects of the gaseous portions of diesel exhaust. While that may have been true for risk assessment values from animal data it is not the case for the epidemiology-based values. In these cases the individual workers were exposed to whole diesel exhaust. The particulate matter-based value is simply a marker of exposure.*

Comment 3: Human work studies likely understate the cancer risk for the general population.

I. The worker studies only evaluated the cancer risk to males. All of the worker studies evaluated by U.S. EPA and by Cal EPA/OEHHA were studies of men. To extrapolate from the male worker studies to the general population may not provide adequate protection for women, children, the infirm and the elderly. While caution must always be exercised in extrapolating from worker populations to the public generally, the animal evidence suggests that it is especially warranted in the case of diesel exhaust.

In 1994 Cal EPA reviewed some animal studies which appeared to reveal higher diesel-induced cancer rates in females. Cal EPA/OEHHA stated, “Several studies also suggest that female animals are more responsive to tumorigenic activity (associated with diesel exhaust) than males.” A newly published animal study adds support to the earlier finding of higher cancer rates in diesel-exposed female animals.

In response to NRDC's comments raising concern about the potential for higher risk for women, Cal EPA/OEHHA acknowledges, "These [male worker] studies do not provide information bearing on the possible greater or lesser sensitivity of other groups such as women and children." Cal EPA/OEHHA also states, "[The diesel exposure assessment] now acknowledges the possibility that female humans may be more susceptible to cancer from diesel exhaust than the male workers whose experience is the main basis for risk assessment" This is an important admission that the current risk assessment may understate risk for a major portion of the population: women.

Women, as well as men, deliver packages, drive trucks, serve as conductors on trains, operate buses for public transit systems, and are otherwise frequently exposed to diesel at work. Even aside from potentially high occupational exposure levels, women are routinely exposed to diesel exhaust as urban residents. Prudent public health policy should ensure that levels of diesel exhaust are protective of even the most vulnerable segments of the population, and should assume, based on the animal evidence, that women may be more susceptible to cancer from diesel exhaust than men.

***Response**: It is not possible to quantify any difference in response of women and men to exposure to diesel exhaust because of a lack of epidemiological data in females. It is a concern to OEHHA and is one of the reasons for using the 95% upper confidence limit on the slope of the dose-response curve in male workers. Use of the upper confidence limit may help account for potential increased sensitivity of women to the carcinogenic effects of diesel exhaust. Use of the upper 95% confidence interval instead of the maximum likelihood estimate provides a 2-to 3-fold higher estimate of risk.*

Comment 4: Human worker studies likely understate the cancer risk for the general population due to the "healthy worker effect". Cal EPA/OEHHA appropriately places considerable weight on two studies of railroad workers conducted by Garshick et al. Although these studies are important indicators of the cancer risk, they cannot be assumed to directly reflect the cancer risk for the population at large. The general public may well have a greater cancer risk than the workers analyzed by Garshick. Garshick limited his studies to workers with 10 or more years in the railroad industry who qualified for railroad retirement benefits. Workers generally have been documented to be in better health than the general public; this "healthy worker effect" has been long noted in epidemiological studies. Better health among workers is probable particularly for workers in an industry with a relatively successful history of collective bargaining for benefits, such as the railroad industry.

Furthermore, worker studies are inherently limited in providing little information about possible effects for children, for the elderly, and for people with chronic illnesses (such as emphysema, asthma, bronchitis, etc.) or depressed immune systems. To the extent that these groups are slower to remove particles from deep lung tissues, they are likely to have greater risk from diesel exhaust exposure compared to long-term workers, because the time needed to clear small particles from the lungs appears to be a factor increasing the risk of tumor development.

In response to NRDC's 1994 comments about the "healthy worker effect," and how the generally better health of studied workers may induce us to understate the real risk posed by diesel exhaust for the general public, OEHHA's 1997 risk assessment report now includes a substantial discussion of this effect. Nevertheless, it is important to remember that the 1997 quantitative risk assessment does not (and cannot) reflect this effect, because the magnitude of the effect is unknown. Therefore, the quantitative estimate is likely to understate the true population risk from diesel exposure.

Response: *The commentator correctly points out that it is not possible to quantify accurately the magnitude of the "healthy worker" effect. The meta-analysis conducted indicates that the healthy worker effect does contribute to heterogeneity in the calculations. It is yet another reason why OEHHA chooses to use the 95% upper confidence limit on the slope of the dose-response curve in workers to reflect the risk to the general population. It is not possible to state with certainty that this practice accounts fully for the healthy worker effect, but it is more appropriate than using only a Maximum Likelihood Estimate, which is the estimated average risk value for the study population. It is possible that the healthy worker effect may be reducing the risk estimate. It is also possible that the healthy worker effect may be the cause of the decreased relative risk estimate in the longest exposures in the Garshick cohort study. This could happen if the sensitive individuals are leaving the cohort through death or to reduce exposure.*

Comment 5: The general population begins exposure to diesel fuel at a very early age - which may increase the lifetime risk of disease. Diesel exhaust exposure is virtually universal. Therefore, the carcinogenicity of diesel exhaust may present an especially serious public health problem. Although the general public experiences lower exposures than workers in diesel-related industries (e.g., trucking, mining, shipping, rail, transit, and agriculture), exposure to diesel exhaust for nearly every human being begins at birth and lasts throughout a lifetime.

Cancers induced by diesel smoke involve a latency period of a number of years between damaging exposure and development of cancer. The 1987 railroad worker study by Garshick as well as a truck driver study by Steenland show an increase in lung cancer risk with increased duration of exposures.

Exposure to diesel exhaust begins for many people very early in life, often in infancy. Neither animal studies nor worker studies examining the effects of diesel exhaust adequately capture this feature of the general public's exposure. Most of the animal studies involving diesel exhaust inhalation begin exposure with "adolescent" rats. OEHHA has responded to NRDC's comments about the early initiation of human exposure by acknowledging the validity of this point.

Even apart from the likelihood that infants may have slower pulmonary clearance times, increasing their effective exposure to diesel exhaust, the very early onset of exposure itself may significantly increase cancer risk for the public relative to experimental animals and relative to adult workers. For these reasons, direct extrapolation from traditional animal exposure studies as well as extrapolation from worker studies are likely to underestimate the risk to the human public, whose

exposure to ambient diesel exhaust begins in early childhood.

***Response**: The commentator correctly points out that we have no way of quantifying the effect of exposure at an early age given the existing data. This is again another reason to use the upper 95% confidence limit of the slope of the dose-response curve in workers. We cannot say with certainty that this accounts for any effect of early exposure, but this practice is more protective of public health and attempts to account for more of the uncertainty than using a maximum likelihood estimate of the slope of the dose-response line.*

Comment 6: The general population is exposed to diesel exhaust and a variety of carcinogens simultaneously. Human beings do not have the good fortune of being exposed only to one carcinogen at a time. Most of us have routine, involuntary exposure to carcinogens: we ingest cancer-causing substances from pesticide residues on our food, household products, furniture and carpets, tobacco smoke (even if we are not smokers), ambient air toxics (such as benzene and formaldehyde), and air-borne particles (such as asbestos). The impossibility of eliminating exposures to these ubiquitous, though usually low-level, carcinogens makes it difficult to argue that direct extrapolation from most animal or worker studies can be adequately protective of the public.

Human risk from diesel exhaust may be amplified by the routine, low-level exposures to other carcinogens which can themselves act to initiate or promote tumor growth. Animal studies in which diesel exhaust was the only known carcinogenic exposure may not be properly representative of the complex exposures to which most people are subject. Therefore, a direct extrapolation from the animal studies may underestimate the risk to the public.

Worker studies are more likely to reflect the multiplicity of exposures to carcinogens which real people routinely receive than controlled animal studies, but extrapolation from the worker studies may still underestimate public risk to the extent that it excludes smokers or asbestos-exposed workers. Because the diesel exhaust risk assessments derived from the best occupational studies reveal a higher risk than those based on animal data, it is paramount that Cal EPA/ARB continue to use worker studies as the basis of human risk assessment.

Additionally, a safety factor should be introduced to account for the many chronic carcinogenic exposures which occur simultaneously throughout a lifetime.

Many members of the general population are exposed to low levels of some carcinogens, but those who are smokers (or who reside with smokers), drinkers, steelworkers or farmers may have relatively high level of exposures to carcinogens. A protective policy for the diesel exhaust risk assessment should consider some form of safety factor in order to account for multiple carcinogenic exposures.

OEHHA acknowledges that the 1997 risk assessment is unable to account for additional environmental exposures to carcinogens or their potential to enhance the potency of the risk associated with exposure to diesel exhaust. This provides yet another reason to view the current diesel-risk assessment as conservative, probably underestimating the human health risk.

***Response**: After public and peer review, including some suggestions by the Scientific Review Panel, OEHHA has decided not to use unit risk factors from the animal studies in the range of risks for humans. Rather, OEHHA will focus on the human epidemiologic studies to generate a range of risks for humans. At this time, based on available data, we do not see that it is warranted to use an additional safety factor in order to account for multiple carcinogenic exposures. The workers were undoubtedly exposed to multiple carcinogens. In addition, in a site-specific risk assessment, the risks of other carcinogenic exposures would be added in to the risk for exposure to diesel exhaust particulate matter.*

Comment 7: Atmospheric transformation may increase the mutagenicity and the carcinogenicity of diesel exhaust. Diesel exhaust undergoes changes when emitted into the atmosphere. In an effort to better understand how atmospheric processes may affect the risks associated with mobile source exhaust, EPA conducted simulations in smog chambers. A 1988 EPA study, which was confirmed by another smog chamber study in 1991, indicated that volatile organics from combustion greatly increased in mutagenic potency after being irradiated. Furthermore, the increased potency in inducing mutations in bacteria is especially profound for the gas phase of diesel exhausts. If genetic mutation is in fact a step in the multi-phase process leading to cancer from diesel exposure, a more potent mutagen would result in a more potent carcinogenic effect.

EPA's smog chamber studies suggest that the gas phase is activated into a more potent mutagen by atmospheric processes, including irradiation. EPA states, "atmospheric transformation may greatly exacerbate the risk from mobile sources, since the contributions of VOCs (volatile organic compounds) to mutagenicity of ambient samples increases dramatically following irradiation." Unless the effects of atmospheric transformation of diesel exhaust are considered, OEHHA's analysis may underestimate the true cancer risk from diesel.

In response to NRDC's 1994 comments on atmospheric transformation potentially increasing diesel risk, OEHHA concurs:

> "Because of lack of data on diesel exhaust, the risk assessment is not specifically intended to address risks posed by atmospheric transformation of gaseous components of diesel exhaust. In this regard, the analysis may indeed represent something of an underestimation of the true cancer rate from diesel. This could be a useful area of future research."

***Response**: OEHHA recognizes that the risk assessment does not deal with atmospheric transformation products of diesel exhaust components. At this point, data are still limited in terms of quantifying both the atmospheric transformation products and rates of transformation, as well as the toxicological properties of the products of atmospheric transformation.*

Comment 8: In addition to cancer, there are other health risks from diesel exhaust particles, ranging from respiratory irritation to premature death, which justify listing diesel exhaust as a toxic air contaminant. Great advances have been made in the 1990s in

understanding the health effects of fine particles as well as in cancer risks posed by diesel exhaust. Recently, two extremely extensive epidemiological studies (by Harvard University in 1993 and the American Cancer Society in 1995) presented further evidence that people living in more PM-polluted U.S. cities had an increased risk of premature death compared to those in cleaner cities. The Harvard "Six City Studies" followed 8,111 adults for 14 years in six U.S. eastern cities, and included evaluation of various potentially confounding factors, including smoking, social/economic status, and occupation. The American Cancer Society Study (also called the ACS study) was a prospective cohort study of over 500,000 adults in 151 U.S. cities, evaluated over the seven years from 1982-1989, with information about such risk factors as sex, race, smoking, passive smoking and occupation. Both of these studies found a strong relationship between $PM_{2.5}$ levels and deaths from respiratory and cardiovascular causes, even after correcting for smoking, occupational exposures and body mass.

Less attention has been devoted to the non-cancer health effects of chronic exposure to other diesel exhaust constituents. Further investigation is clearly warranted because many of the constituents of diesel exhaust are known to harm exposed animals even aside from cancer effects. Benzene, for example, is known to cause disorders of the blood and the blood-forming tissues, especially the bone marrow. Formaldehyde can cause irritation of the eyes, nose and throat and is suspected of interfering with human immune function. Acetaldehyde has many of the irritating properties of formaldehyde, and there is evidence that the chemical may be the causative factor in birth defects associated with fetal alcohol syndrome. 1,3-Butadiene can cause death through respiratory paralysis at high levels, but at lower levels can adversely affect blood-forming tissues and interfere with successful reproduction. Respiratory tract irritations and diminished resistance to infection are associated with long-term exposure to diesel particulate matter.

Obviously, the non-cancer effects of diesel exhaust can also be serious and damaging. The extent to which these effects are already occurring in the population is unclear. Efforts to reduce diesel cancer risk would also reduce these potential health effects.

***Response**: Comment noted. There is a discussion in Chapter 4 of the document of the noncancer health effects associated with diesel exhaust exposure in humans and animals. The discussion of immulogical and asthma-inducing effects has been updated based on data published in the past year. We were alerted to the availability of the data at the October 1997 SRP meeting.*

Comment 9: The cancer risk posed by diesel exhaust is significant, even for the general public. It is seven times higher for diesel workers and other routinely exposed to high levels of diesel exhaust. Even though there are currently state and federal standards restricting respirable particles, those standards were not designed to protect the public from the risk of cancer posed by diesel exhaust. Despite the many ways in which Cal EPA's risk assessment is likely to underestimate the true risk for at least some segments of our population, the cancer risk described by Cal EPA/ARB, even for the average person today, should elicit serious concern.

The US EPA suggests that a cancer risk may be "acceptable" or "negligible" if it induces increased cancers over a lifetime for one person in an exposed population of one million. In 1994

Cal EPA and ARB calculated the cancer risk for the average Californian from diesel exposure to be between 3×10^{-5} and 2×10^{-3} (micrograms per cubic meters)$^{-1}$, -- between ten and a thousand times above EPA's "negligible risk" level. And this is the risk for the theoretically average person who happens to breathe only the statewide "average" concentration levels of diesel exhaust.

In 1997 Cal EPA and ARB suggest that the unit lifetime risk is between 1×10^{-5} and 2×10^{-3}. The higher exposure estimate is the one derived from worker studies. Using the cancer risk estimate derived from the human worker studies and the daily population average exposure, we would expect on the order of 150,000 additional lung cancers in California over a 70 year period. The risk is even higher for urban residents who frequent areas near heavy bus, rail and truck corridors. The risk is also greater for people who have respiratory problems or who smoke, people who regularly exercise strenuously in diesel-polluted areas, and people who work or live near diesel exhaust sources. There is evidence that the risk may be greater for women. This is not a situation which should continue unaddressed. The public deserves prompt action to reduce diesel exhaust in order to prevent needless suffering from cancers and other diseases.

Response: *Comment noted. However, even for the human studies there is a range of risk. In the March 1997 document, the human range of risk reported was 2×10^{-4} to 2×10^{-3} (lifetime-$\mu g/m^3)^{-1}$ for the upper 95% UCL. In the revised document we have revised the range of risk based upon improved estimates of exposure.*

Comments from Western States Petroleum Association, letter dated August 22, 1997, from Jeff Sickenger to Michael Kenny

Comment 1: Cal-EPA should fully disclose its justification for using inadequate worker exposure data. As part of its risk characterization, Cal-EPA should acknowledge that HEI, IPCS, and the lead author of the railroad worker study, Dr., Eric Garshick, have stated that the available exposure data on railroad workers is too limited to develop quantitative cancer potency estimates. Cal-EPA should disclose to the public, the SRP, and Board that the only available actual exposure data is from the mid-1980's and that those data were extrapolated back to 1946. Of course, prior to full dieselization of the railroads in 1959, workers were subject to other exposures to varying degrees (e.g., coal smoke, bunker fuel smoke). OEHHA has made no attempt to distinguish these exposures from it's diesel exhaust exposure estimates- Furthermore, OEHHA excluded shop workers, generally recognized as one of the most highly exposed subpopulations from its analysis without fully disclosing its rationale for rejecting extrapolated shop worker exposure estimates, or the affect of this action on the dose-response relationship between diesel exhaust exposure and lung cancer.

***Response**: As noted in the comments, with respect to the quantitative assessment of risk based upon the railroad worker studies, OEHHA's position differs to different extents from those of a number of individual scientists, including Dr. Garshick, and also from the positions of the HEI and the WHO who generally have found that the inadequacy of exposure information limited the ability to conduct a quantitative risk assessment. However, in reaching their conclusions, they each relied to varying extents upon the results of the dose response analyses of the Garshick et al. 1988 cohort study data conducted by Dr. Crump. In those analyses, Dr. Crump did not obtain significantly positive dose response relationships between the incidence of lung cancer and diesel exhaust exposure. In its own later analyses using different methods, OEHHA obtained positive dose response relationships. This difference between the conclusions of OEHHA and these other authoritative bodies therefore depend in important part upon the comparative merits of the approaches of OEHHA and Dr. Crump. OEHHA, having characterized and reviewed the sources of this difference, finds its approach to be more appropriate. The differences are the subject of Appendix F of Part B, as well as Part C here and previously, and have been highlighted for independent review by the Scientific Review Panel. Additionally, elsewhere OEHHA provides responses to related comments from Dr. Garshick and the HEI. Therefore, OEHHA has included the study as one of several useful ways to estimate the risk from diesel exhaust.*

With respect to its exclusion of the shop workers, OEHHA has provided its rationale in Section 7.3.3. paragraph 5. As indicated by Dr. Garshick (1991), the shop workers who worked in the diesel repair shops shared job codes with workers in non-diesel shops where there was no exposure to diesel exhaust. In addition, the scientific presentations and discussion at the January 1996 meeting affirmed that shop workers were heterogeneously exposed. Some shops entailed diesel exhaust exposures in their operations, others did not. In the original Garshick et al. (1988) cohort study, the investigators reported results including and excluding the job classifications of shopworkers and hostlers. They reported that with both shopworkers and hostlers excluded from the analysis, the effect of diesel exhaust exposure remained significant

and of comparable magnitude to the whole cohort. Similarly, in the Crump et al. (1991) analysis the presence of an effect did not seem to be affected much by the inclusion or exclusion of shopworkers.

With respect to the possibility of confounding by exposures to bunker fuel or coal smoke prior to dieselization, any exposure to these agents was decreasing as diesel exhaust exposure increased. Therefore, these agents could only account for the positive association between cumulative exposure to diesel exhaust and lung cancer if they protected against lung cancer. The ramp and roof patterns of exposure incorporate information on dieselization.

Comment 2: Cal-EPA should acknowledge that the positive association reported in OEHHA's risk assessment is, at best, a very weak positive association. Cal-EPA should acknowledge that changing simple assumptions about worker age and length of employment eliminates the weak positive association between diesel exhaust exposure and lung cancer. Cal-EPA should also acknowledge that the model-fit after changing these assumptions is better than the model-fit obtained using the OEHHA assumptions. Certainly, OEHHA's analysis of the epidemiological data does not support its contention that a causal association is a "reasonable and likely explanation". By contrast, both IARC (1989) and USEPA (1994) have concluded that the epidemiological data is not adequate for use in establishing a cancer potency for diesel exhaust.

Response: *Appendix G addresses the impact of varying assumptions on the association between diesel exhaust exposure and lung cancer. OEHHA has found that the association holds for a variety of reasonable assumptions and approaches.*

With respect to IARC, from our review of their 1989 document, it does not appear that they took any position with respect to the use of epidemiological information in a quantitative risk assessment for diesel exhaust. IARC generally does not conduct quantitative risk assessments.

With respect to the USEPA, OEHHA has developed new analyses whose import greatly differs from those relied upon the USEPA in developing its 1994 draft position. Appendix G clarifies the bases for the difference between the 1994 draft USEPA position and ours. We have discussed these analyses with USEPA staff and they are generally supportive of our work. At the July 1, 1997 workshop Dr. Koppikar indicated that the USEPA would be using epidemiological data in the next quantitative risk assessment it presents to the Clean Air Scientific Advisory Committee. Dr. Koppikar also stated that the resulting risk values were "pretty consistent and very similar with what Cal-EPA has presented here today." (Transcript of the Public Workshop for the Diesel Exhaust Identification Report, p.75.)