REPORT

on

PEER REVIEW WORKSHOP ON

PCBs: CANCER-DOSE RESPONSE ASSESSMENT AND APPLICATION TO ENVIRONMENTAL MIXTURES

May 1996

National Center for Environmental Assessment
Office of Research and Development
U.S. Environmental Protection Agency
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NOTICE

This report is intended as a general record of discussions held during the Peer Review Workshop on *PCBs: Cancer-Dose Response Assessment and Application to Environmental Mixtures.* Preparation of this material was coordinated by Eastern Research Group, Inc. (ERG), a contractor to the U.S. Environmental Protection Agency (EPA). The report is not a complete record of all details discussed, nor does it embellish, interpret, or enlarge upon matters that were incomplete or unclear.

Mention of trade names or commercial products does not constitute endorsement or recommendation for use. Statements in this report are the individual views of each meeting participant; none of the statements represents analyses or positions of the National Center for Environmental Assessment or EPA.

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FOREWORD

EPA's National Center for Environmental Assessment (EPA/NCEA) has prepared a draft assessment of PCB cancer potency estimates. The document, *PCBs: Cancer-Dose Response Assessment and Application to Environmental Mixtures,* also includes guidance for applying the potency estimates to PCB mixtures found in different environmental media. In keeping with Agency peer review policy requirements, EPA/NCEA subjected the draft assessment to external expert review. NCEA convened a workshop as an opportunity for expert reviewers to discuss the issues and offer recommendations on the draft assessment.

SECTION ONE

PANEL INTRODUCTION

In January 1996, the U.S. Environmental Protection Agency's (EPA's) National Center for Environmental Assessment (NCEA) issued a draft assessment of cancer potency estimates for polychlorinated biphenyls (PCBs). The draft document, *PCBs: Cancer-Dose Response Assessment and Application to Environmental Mixtures,* includes guidance for applying the potency estimates to PCB mixtures found in different environmental media.

In April 1996, Eastern Research Group, Inc. (ERG), under contract to EPA, selected a panel of external experts to peer review the draft PCBs document, prepare written comments, and convene to discuss review comments at a workshop. The external peer review panel included representatives from industry, academia, environmental groups, and state government. The selected reviewers have expertise in the following areas: carcinogenicity, public health, epidemiology, toxicology, pathology, immunotoxicology, mechanisms, and biostatistics/risk assessment modeling (see Appendix A for a list of expert reviewers).

Panel members were given 2 weeks to review the draft PCBs document and submit comments regarding "major scientific issues" as identified by EPA. (The list of review issues and premeeting comments are provided in Appendices B and C). The Peer Review Panel Workshop on *PCBs: Cancer-Dose Response Assessment and Application to Environmental Mixtures* was convened at Bethesda, Maryland, May 21 and 22, 1996. The agenda included presentations by several EPA officials, representatives of the General Electric Company, and the general public (see Appendix D for the workshop agenda and Appendix E for the presenter list and schedule for public comment).

For the workshop, the panel's charge was to critique the draft PCBs document and discuss issues and comments. Subsequently, the panel was to prepare a report summarizing all workshop discussions and highlighting suggestions and recommendations to EPA for drafting the final PCBs document. This report presents summaries of discussions held on each issue at the workshop.

Material was contributed by individual panel members and revised following review of an initial draft of the report.

The next step in developing the PCBs document calls for EPA to revise the PCBs document, based on consideration of issues raised both at the workshop and in premeeting comments, by about July 1. EPA will then submit the document to inter-Agency review and subsequently revise the document as necessary before submitting a final report to Congress by September 1, 1996. Given the time constraints, members of the peer review panel focused their attentions on the major issues raised by EPA and provided the Agency with a number of recommendations.

SECTION TWO

PANEL SUMMARY

The peer review panel discussed the following issues concerning the draft PCBs document during the 2-day workshop. The reviewers offered a number of recommendations, which are presented in the context of topic-specific discussion summaries.

GENERAL ELECTRIC DATA: APPLICABILITY TO THE ESTIMATION OF HUMAN CANCER RISKS PRESENTED BY ENVIRONMENTAL MIXTURES

The General Electric (GE) study is regarded as a superior experiment, providing the best available rat carcinogenicity data. The panel recommends that the tumor data be included in EPA's Integrated Risk Information System (IRIS), contingent on GE providing certification that the summary tumor data accurately reflect the raw data. It should be noted that the Aroclor 1254 sample may be atypical; however, data showing this have not been presented or published. The toxicokinetic data from this study are of great interest, but are difficult to evaluate without reviewing the entire study. In addition, unlike the pathology information, to date these results have not undergone external peer review. Likewise, information on non-neoplastic toxicity could provide valuable information on conditions underlying carcinogenicity and should be sought for future guidance on risk assessment methods. The observations presented in the draft report on the GE study are generally correct; however, the inference of a specific mechanism from the pattern of liver cancer warrants further research.

In the absence of congener-specific analytical data and an acceptable model for relating a Cancer Slope Factor (CSF) to PCB composition, the risk assessor may assume that the CSF of an environmental mixture may lie somewhere (1) within the range defined by the CSF values for Aroclor 1016 as a lower bound and Aroclor 1254 as an upper bound, as calculated by EPA from the GE/Battelle data, or (2) outside that range, reflecting the metabolic concentration of the more toxic and persistent congeners as PCBs pass up the food chain. Aroclor 1016 may contain congeners that

are somewhat more persistent than those in well water or heavily dechlorinated sediments. Nonetheless, it represents the commercial PCB mixture with the lowest CSF value that can be documented by available data.

In the absence of analytical data to the contrary, the assessor may presume that the PCBs in air or water samples have compositions, and hence presumptive CSF values, similar to that of Aroclor 1016 (low end of the range). Conversely, the assessor may assume that the PCBs found in soils, sediments, or biota have compositions, and hence presumptive CSF values, more like Aroclor 1254. If analytical data suggest that the more toxic and persistent PCB congeners are present in increased concentrations, the appropriate CSF for the environmental PCB mixture would be higher than that determined for Aroclor 1254.

When congener-specific analytical data and an accepted (i.e., peer reviewed) model linking a CSF to a PCB composition become available, that model may be used to calculate a CSF for the environmental composition that might fall above or below the above-defined range. For example, preliminary analysis of the GE/Battelle data suggests that the tumor incidence data for both sexes can be described ($r^2 = .897$) in terms of the combination of total adipose PCB concentrations and liver toxic equivalency (TEQ) loadings via a so-called unisex model. Once such relationships have been optimized, confirmed, and reviewed, they should be useful for defining CSF values for a wide range of environmental PCB compositions.

ISSUES OF ANALYTICAL CHEMISTRY

The draft PCBs document focuses on the hazard identification and dose-response issues for PCB-induced carcinogenicity. It also raises some issues concerning exposure to humans; in particular, how differences between various exposure scenarios are likely to result in exposure to different mixes of PCB congeners (i.e., because of environmental processes). The issue of Aroclor concentrations accurately reflecting PCB concentrations should be considered by risk assessors with regard to characterizing potential human exposures based on monitoring data.

Because of the environmental fate and transport processes, the analytical methods used to detect Aroclors may not always accurately reflect what is present. In particular, gas chromatography/mass spectrography (GC/MS) reveals a pattern of several peaks that are characteristic of the various Aroclor mixtures and that can be used to indicate the presence of PCB mixtures representative of specific Aroclors. As environmental transformation processes become more extensive, however, the presence of individual congeners in the original Aroclor mixture can change significantly. Perhaps at some point, the GC/MS spectra for such mixtures might suggest the absence of Aroclors based on the lack of a characteristic pattern of peaks. While there may in fact be no "Aroclors" present, this does not preclude the presence of high levels of individual PCB congeners. Thus, those developing or using monitoring data to estimate potential human exposure should be cautioned that data on Aroclor concentrations may not be reflective of the PCB concentrations. If an initial screening analysis suggests that the data on total PCBs and Aroclor mixtures are incongruous, the assessor may want to recommend that a more congener-specific analysis be conducted. Where data on both Aroclor mixtures and individual congeners are available, those most accurately reflecting actual site conditions should be used in the assessment.

ISSUES RELATED TO PRENATAL AND EARLY NEONATAL EXPOSURES

The accumulation of PCBs in human adipose tissue creates a store for subsequent release of PCBs into the bloodstream and then into the fetal circulation. During the postpartum period, PCBs are mobilized from adipose stores, transferred into breast milk, and delivered to the neonate via nursing (Dewailly et al., 1991). This source of exposure may account for a substantial fraction of dioxin-like compounds, and the same may be true for the dioxin-like PCBs as well as other PCBs. Sensitivity of fetuses and early neonates has not been assessed sufficiently in humans. The panel expressed some concern that EPA needs to acknowledge the role of exposures during early development because of the magnitude of the exposure pathway, the possibility that the fetus and neonate are more sensitive, and the likelihood of interactions among thyroid and hormonal development.

Normal fetal development depends on the timing and rate of release of T3 and T4. Some evidence indicates that PCBs can alter normal T3 and T4 metabolism, thereby disturbing thyroid function and provoking secondary impacts on organogenesis during development.

The present assessment uses lifetime bioassays in adult animals; EPA did not have data for prenatal and early neonatal exposure to incorporate in this assessment. The panel suggested that the assessment should acknowledge that individuals in early life stages may be more sensitive to carcinogenesis than in adulthood. The panel also offered the following additional suggestions:

- EPA should note and discuss the role of thyroid function and abnormalities in the carcinogenic action of PCB exposure during pregnancy and lactation.

- The risk assessment needs to identify the higher exposure that occurs during gestation and lactation. This exposure has been recognized for dioxin-like compounds, and Dewailly et al. (1991, 1994) extended the observations to PCBs. This exposure pathway, while technically ingestion (i.e., breast feeding), is sufficiently distinct that EPA should note it separately.

- Any estrogenic/anti-estrogenic, androgenic/anti-androgenic, or other hormonal activity of PCB mixtures has the possibility of altering the development of reproductive organs and/or the urogenital tract. The risk assessment should note that this mechanism of carcinogenesis is distinct from the liver cancer mechanism in exposure of adults and may be significant, but is not examined.

- Are cancer potency factors any greater for early life stages? EPA should comment on any evidence that cancer potency varies with life stage and include this information in the risk assessment. Three studies involving acute high dosing of pregnant mice or of neonatal mice with Aroclor 1254 found no increase in any tumor type in offspring exposed to PCBs alone, compared with oil-treated controls. This suggests that direct carcinogenicity to the fetus and neonate is not a major concern (Anderson et al. [1983] JNCI 71:157-163; Anderson et al. [1986] Int. J. Cancer 38:109-116; Anderson et al. [1994] Carcinogenesis 15:2245-2248). This finding is consistent with the general observation that chemicals that require extended exposure of adults to induce a tumorigenic effect (a presumed tumor promotion-like mechanism) are not perinatal carcinogens in rodents. In the three studies referenced above, the perinatally administered PCBs were effective in promoting tumors of liver and lung initiated by a nitrosamine. In another study (Beebe et al. [1993] Carcinogenesis 14:1545-1548), the PCB treatment was not given until 8 weeks of age and promotional effects were still observed; thus, it is not clear whether perinatally received PCBs are especially effective in promoting perinatally initiated tumors.

METABOLISM AND MECHANISMS OF ACTION

Although the rate of metabolism is slow (Mills et al. [1985] TAP 78, 96-104), PCBs may be converted by hepatic enzymes to hydroxylated metabolites. The relative rates of conversion are dependent on the number and placement of the chlorine atoms present. PCBs with fewer chlorines and with adjacent, unsubstituted carbon atoms are more readily susceptible to metabolic attack. Cytochrome P-450 isozymes may catalyze these hydroxylation reactions via an electrophilic arene oxide intermediate or via direct insertion mechanisms. Evidence for the intermediacy of arene oxides during PCB metabolism is found in the identification of (1) NIH-shift products, (2) dihydrodiol metabolites, (3) mercapturic acid products, and (4) sulfone metabolites (Sipes and Schnellman, Biotransformation of PCBs: Metabolic pathways and mechanisms. In: Safe and Hutzinger, eds. [1987] Polychlorinated Biphenyls [PCBs]: Mammalian and Environmental Toxicology, Springer Verlag, Heidelberg, pp. 97-110).

PCB metabolites with multiple hydroxyl groups also have been identified in animals and in microsomal incubations (McLean et al. [1996] Chem. Res. Toxicol. 9:158-164). Dihydroxy metabolites may be oxidized in vitro to o- or p-quinones by peroxidases. In vitro studies have demonstrated that adducts of PCBs and nucleotides (dGp and dAp) or exogenous DNA may be formed during the hydroxylation step (from electrophilic arene oxides) and during the peroxidase-catalyzed oxidation of PCB catechol and hydroquinone metabolites to the respective o- and p-quinones (McLean et al. [1996] Chem. Res. Toxicol. 9:165-171; Oakley et al. [1996] Carcinogenesis 17:109-114). Hydroxylated PCB metabolites may have estrogenic activity (Gierthy et al. [1995] Organohalogen Compounds 25:419-423).

Higher halogenated PCBs may be efficacious inducers of xenobiotic-metabolizing enzymes, although they are poor substrates. Several PCBs, possessing no or one ortho (2,2',6,6') chlorine, bind the Ah receptor with avidity (Bandiera et al. [1982] Chem.-Biol. Interact. 39:259) and induce cytochrome P-450 1A. Several di-ortho substituted PCBs induce cytochrome P-450s such as phenobarbital, while other congeneric PCBs may induce cytochrome P-450s from both subfamilies (Robertson et al. [1991] Environ. Toxicol. Chem. 10:715-726). Many of these PCBs may also induce epoxide hydrolase, glutathione transferases, and glucuronosyl transferases. Induction of xenobiotic

metabolism may be accompanied by an increase in hepatic cell size and number and a proliferation of the endoplasmic reticulum. The persistent induction of hepatic cytochrome P-450s, in the absence of an oxidizable xenobiotic substrate, may provide suitable conditions for the generation of reactive oxygen species (Portier et al. [1996] TAP 138:20-30).

Several PCBs tested as promoters in rat two-stage hepatocarcinogenesis were efficacious when they were administered at doses that caused liver hypertrophy and the induction of cytochrome P-450s (Silberhorn et al., 1990). Promotor activity has been observed among groups of PCB congeners that have been characterized as having widely different kinds of biological activity, including congeners that are Ah receptor agonists, congeners that induce cytochrome P450 1A and 2B isozymes, and congeners that have a pattern of enzyme induction similar to that of phenobarbital. This may indicate multiple mechanisms of action for promotion (Buchmann et al. [1991] TAP 111:454-468). Congeneric PCBs may interfere with gap-junctional intercellular communication via structure-specific mechanisms. Mono- and di-ortho chlorine substituted PCBs were more active (Swierenga et al. [1990] Carcinogenesis 11:921-926).

Aroclor 1254 is active as a promoter in the mouse lung initiated with methylating nitrosamines. The lung is also a potential target for cancer risk following PCB exposure. Relevant mechanistic information includes: (1) persistent induction of cytochrome P-450 1A in the lung (up to several months after a single PCB dose), (2) promotion by congener 138, not by congener 153, and partial abrogation of 138's effects by 153, suggesting a role of the Ah receptor, (3) correlation of the promotion effect with body burden of congener 99, and (4) selective retention of PCB congeners, especially congener 105, in mouse lung (Anderson, Experimental Lung Research 17:455-471; Anderson et al. [1993] J. Environ. Pathol. Toxicol. Oncol. 12:3-16).

EPIDEMIOLOGY AND ANIMAL DATA

Human Studies

A summary of the human epidemiologic data regarding PCB exposures and cancer risk should be included in the draft document. It was noted (not a consensus) that increased risks reported thus far, even where statistically significant, are rather small compared with what might have been predicted from the rat liver tumor studies. In interpreting these findings, however, as a reality check the following should be noted:

- There are inadequacies in the epidemiologic data with regard to limited cohort size, problems in exposure assessment, lack of data on confounding factors, and the fact that occupational exposure may be to different congener mixtures than found in environmental exposures.

- Most researchers think that PCBs act mainly as tumor promoters. Thus, at nontoxic doses, PCBs might be expected to increase cancer risk mainly in humans that have sustained cancer initiation due to exposure to genotoxicants or to the presence of a mutant gene. For common cancers that have complex and multiple etiologies, promotive effects will be seen by epidemiology only if specifically looked for. Epidemiologic studies have not thus far tested this hypothesis.

- It might also be noted that PCBs can have a tumor-reducing effect in some cases. PCB exposure before genotoxic carcinogens also results in a reduction in tumors, due to induction of detoxification enzymes. A suppressive effect on mammary tumors has been noted in some studies. This possibility greatly complicates interpretation of human data.

Animal Studies

In view of the considerable species, strain, and target organ differences in response to carcinogenic stimuli, and given the fact that chronic toxic-dose lifetime exposure studies on PCBs alone do not model probable human risk situations, it seems too limited to predicate all PCB risk assessment on the number of rat liver tumors arising after such exposure. It would be desirable to

have datasets on different tissues of at least two species, including initiation-promotion, to arrive at the range of probable-risk doses.

There is nearly a complete lack of tumor promotion dose-response data that could be used. Suggested experimentation could be neonatal initiation with two doses of representative carcinogens (tobacco-specific nitrosamine, polycyclic aromatic hydrocarbon, aryl amine, mycotoxin), followed by two or more doses of chronic PCBs. The highest dose in each case would be the lowest known effective dose. Lower doses would go down by 10-fold, to within range of human values. Exposure would be for at least 18 months and tumors would be the endpoint.

Since PCB mixtures may have complex tumor promotive and suppressive effects, and particular congeners may show tissue-specific retention patterns, more animal work is needed on the in vivo actions of individual congeners and of defined mixtures.

Sex Differences

Liver tumors were much more prominent in female than male Sprague-Dawley rats exposed to lifetime PCBs. This differential was not seen in Fisher rats, however, and in mice males are more susceptible than females. The possibility of sex differences should be considered in analysis of human data, but not built into differential risk values at present.

Research has shown that this class of compounds can affect estrogen metabolism causing a pattern of toxicity that differs between females and males. In comparing observed Standard Mortality Ratios (SMRs) and like measures to animal-based predictions, measurements grouped across sexes might underpredict responses derived from animal-based estimates that are drawn from studies on females. This is especially true for liver carcinogenesis where the most recent animal studies (predominantly the GE study) show large differences between the sexes.

Tumor Sites Besides the Liver (Thyroid)

In view of the causation of thyroid tumors in male rats exposed to PCBs, incidences of cancers at sites other than the liver should be scrutinized closely in human studies. EPA should consider the following:

- Existing human data may be analyzable for interactions, particularly for factors such as smoking and other occupational exposures.
- New epidemiologic studies could build in current concepts on initiation, such as dietary factors and relevant genetic polymorphisms.

Internal Dose

EPA should be commended for suggesting an assessment method that goes beyond the use of administered doses as the basis for evaluating risk. The review panel encourages EPA to continue to develop a method using internal dose or another form of dosimetry. The data are not yet available, however, to determine the appropriate dosimetric for PCB carcinogenicity. Given this and other uncertainties, it is inappropriate at this time to include this adjustment for internal dose.

One reason not to adjust for internal dose is that the mechanism by which PCB causes carcinogenicity in rats is unclear. Although a number of researchers have suggested that the area under the curve measurements is useful for predicting adverse effects associated with persistent chemicals, such a hypothesis requires the assumption that the components of the PCB mixture that can cause cancer persist in the body and are available to initiate or promote cancer.

The proposed quantitative approach involves accounting for internal exposure after external exposure ceases. This method has some fundamental problems, including:

- The omission of a negative impact on the area under the curve resulting from the slow buildup of the toxicant.

- A valid approach should vary depending on the period of life when exposure occurs (early versus late in life).
- A necessary assumption for the method to be valid (i.e., extended exposure duration will be predictive of the human body burden versus time curve) does not hold.
- A valid approach should vary with exposure duration.
- The dose used to calculate cancer potency is external dose; this is then applied to an assumed internal dose. The dosimetries should be consistent.
- The approach should consider the temporal exposure profile.
- If potencies are expressed in terms of internal dose, the body weight 3/4 ($BW^{3/4}$) interspecies translation of dose should be omitted.

The review panel recommends that the new GE data—particularly the stop experiments, plus other stop studies (e.g., Kimbrough/Linder, 197) and other mechanistic data—be fully explored to provide qualitative support for developing a method to account for internal dosing after exposure ceases. These data, along with data on human and animal internal doses, should be used to determine the appropriate dosimetric and, if necessary, a quantitative correction for internal dosing.

POTENCY ISSUES

Background

The draft PCBs document establishes two ranges of carcinogenic potencies. One is based on the range of 95-percent upper confident limits (95-percent UCL) observed in the different studies available to EPA at the end of 1995. A second range was developed for the maximum likelihood estimates (MLEs) from the same studies. In developing these ranges, EPA excluded the estimated potencies from two studies of PCBs in male rats.

The two ranges reflect the effects of study-specific factors on the estimated 95-percent UCL values and the MLE values for potency. The study-specific factors include: gender, strain,

commercial mixture tested, and inter-assay variation. The draft document also offers guidance for selecting values from either end of the two ranges. This guidance is based on differences in PCB composition as determined by the route of exposure. Finally, the draft offers guidance on which of the two ranges to use in different types of risk assessments.

Comments

The review panel strongly supported many aspects of the proposed approach. In particular, panel members supported the use of a range versus a single value of potency, including zero as the lower end of the range. This support was based on the panel members' belief that differences in the composition of PCBs received by individuals exposed at different locations and by different routes of exposure would result in differences in the carcinogenic potential of the mixtures. Therefore, a similar approach should be used when the results of the new Battelle study are considered. In addition, panel members agreed that the range should predominantly reflect changes in potency attributable to changes in the composition of the mixtures. Four issues, however, were raised by one or more panel members. These are discussed below.

Issue 1. ***The Proposed Ranges Do Not Appropriately Account for the Sources of Uncertainty in PCB Potency Estimates***

The suggested approach presents a conceptual problem in that EPA uses the proposed ranges as a measure of the differences in potency that could occur as the result of being exposed to mixtures of PCBs with different compositions. As the Agency acknowledges, however, much of the variation in the ranges is not due to differences in PCB composition but to interstudy variation and differences in the strain and gender of the animals tested. Because of these differences, the low-end estimate of potency for PCBs is taken from the assay of a highly chlorinated mixture, Aroclor 1254.

The uncertainty resulting from noncompositional factors are equally applicable to mixtures of higher and lower chlorinated PCBs. If EPA wishes to characterize the variation in PCB potency

that arises from differences in the composition of PCBs, then the Agency should base the range on the results of studies where data are matched by strain and gender (the GE/Battelle and Schaeffer et al. studies) and the differences in potency are only due to differences in mixture composition.

A second and related problem is that the ranges of the MLE values and the 95-percent UCL values (roughly a factor of 3) do not capture the total uncertainty in the potency ranges. The difference between the two ranges is only a function of the uncertainty that occurs from the limited number of animals in the assays and does not take into account the uncertainty attributable to variation in the strain and gender tested, variation in Aroclor tested (i.e., the composition of Aroclor 1254 in one study is not identical to the composition of Aroclor 1254 in a second study), or inter-assay variation. Inter-assay variation may be significant if the assays differed in number of dose groups, choice of dose rates, and laboratory practices used. Most important, the range of dose does not consider the uncertainty attributable to the choice of the dose-response model used in the low-dose extrapolation. Because of these additional sources of uncertainty, the differences between the two ranges might be greater than the current draft document suggests. For example, because of the decision to rely on data from female rats in the Battelle study and the use of a conservative dose-response model, the true range of central tendency estimates might be orders of magnitude below the range of 95-percent UCL values.

Finally, several members of the review panel questioned EPA's decision to exclude the results of the male rats in the Norback Aroclor 1260 study and the Schaeffer Clophen A 30 and 60 study. Given that the purpose of the analysis was to present the range of potency estimates observed in studies of various strains, in either gender, and in different mixtures, the exclusion of these two sets of data seems inappropriate.

Conclusion and Recommendation: EPA needs to develop a range (or ranges) of potency in a fashion that is meaningful to risk assessors and decision-makers and that clearly communicates the impact of all sources of uncertainty in the potency estimates. One approach would be to develop estimates of the range of potency that occurs as a function of PCB composition and then to quantitatively or qualitatively characterize the uncertainty around that range. Whatever manner is

chosen, however, the Agency needs to clearly state which sources of uncertainty are included in the potency ranges and which are not.

Because of the limited time available and the difficulty in quantitatively characterizing many of these sources of uncertainty, EPA may wish to deal with certain sources of uncertainty in a qualitative or semiquantitative fashion. Information in Figure 1 and Table 1 may be helpful in this regard.

Figure 1. Sources of Uncertainty and the Relative Magnitude of Their Impact on PCB Carcinogenicity

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Low-dose extrapolation:

The uncertainty in potency estimates from low-dose extrapolation cannot be quantified at this time. Other studies have indicated that this source of uncertainty can affect potency estimates by several orders of magnitude.

Gender:

Gender differences are seen in all studies, with males usually, but not always, less responsive than females.

Strain:

Four strains of rat have been used to evaluate PCBs. Sprague-Dawley rats appear to fall in the middle of the range of sensitivity to PCBs.

Inter-assay variability:

Two assays used the same Aroclor and strain (Battelle and Norback). The studies differ in the response rates and in the study design. These differences can result in a 3- to 6-fold change in potency (see premeeting comments by Dale Hattis, Appendix C).

Variation in composition:

Estimates of potency vary by a factor of 20 for different mixtures of PCBs.

Limitations in sample size:

In all studies, the ratio of the MLE to the upper confidence limit was within a factor of 3.

Human variability:

Current understanding of the uncertainty in potency due to gender, age, and other types of human variation cannot be quantified at this time.

Table 1. Variation in Responses Due to Differences in STRAIN

Males given 100 ppm of Aroclor 1260 or A60:

Study	Strain	Response
Battelle	Sprague-Dawley	0.21
Schaeffer	Wistar	0.91
Norback[a]	Sprague-Dawley	0.13

Females given 100 ppm of Aroclor or A60:

Study	Strain	Response
Battelle	Sprague-Dawley	0.48
Kimbrough	Sherman	0.73
Norback[a]	Sprague-Dawley	0.89

Females given 100 ppm of Aroclor 1254:

Study	Strain	Response
Battelle	Sprague-Dawley	0.59
NCI	Fischer	0.04

a. Doses to rats reduced to 50 ppm during the study

Issue 2. ***Selection of Single Values of Potency From the Two Potency Ranges***

A majority of the panel members who voiced an opinion on this issue suggested that consideration be given to reflect the composition of the PCB mixture. Possible options for such an approach include the use of analytical information on the nature of the parent PCB mixture associated with the exposure. Panel members agreed that the proposed use of exposure pathways as a default approach for the selection of a specific potency estimate in a specific risk assessment is reasonable.

Conclusion and Recommendation: The panel affirmed the approach proposed by EPA in the draft PCBs document as an acceptable default option; namely, the upper end of the CSF range is used to quantify risks associated with soil ingestion and food intake exposures, while the lower end of the range is used for dermal contact, water ingestion, and inhalation exposures in the absence of congener specific or Aroclor data.

Issue 3. ***Should the Range of Potencies Be Limited to Those of the Tested Aroclor Mixtures?***

In the draft PCBs document, EPA proposes to use the range of PCB potencies derived from existing studies of Aroclor mixtures. Implicit in the proposal is the decision not to consider potencies outside of this range, even though the Agency acknowledges that the "range observed for commercial mixtures may underestimate the true range for environmental mixtures" (p. 35). The draft also includes warnings such as "For exposure through the food chain, risks can be higher than those estimated in this assessment" (p. 44).

Conclusion and Recommendation: After considerable discussion, the panel concluded that it was premature to offer any quantitative adjustment of the upper end of the range of potencies. The group suggested that EPA retain the qualitative warning. Also, panel members were in general agreement that the issue deserves further study as a research topic.

Issue 4. The Need for Age and Gender-Specific Potency Estimates

Several panel members suggested that EPA provide guidance on age- and gender-specific potency estimates. The finding that PCBs cause different carcinogenic responses in male and female Sprague-Dawley rats and the finding that age effects the ability of PCB congeners to act as promoters raised considerable discussion during the workshop.

Conclusion and Recommendation: The panel concluded that more data are needed on this issue and recommended additional research.

HOW TO APPLY DATA TO RISK ASSESSMENT? TECHNIQUES USED IN CALCULATING SLOPE FACTORS, UNCERTAINTY ANALYSIS OF EXPOSURE PATHWAY-BASED FACTORS

On this topic, the panel found four points of general agreement:

- "Undetermined" models have fewer data points than estimated parameters and should not be used for risk assessment. When data are processed in this way, an infinite number of perfect-fit solutions are possible, and the choice of any one combination of parameter values must necessarily be arbitrary.

- Where possible, nonlinearities due to pharmacokinetic processes, cell killing, cell proliferation, enzyme induction, and similar "high dose" phenomena should be incorporated into the definition of effective dose prior to entering the dose and response data into a multistage model. If it is possible to express internal dose in terms of an internal concentration of putatively active agent, then the $BW^{3/4}$ scaling factor used for translating animal mg/kg-day dosage into human equivalents can be eliminated for "central tendency" risk projections.

- Likelihood techniques or other analogous weighting systems should be used where feasible to integrate information from multiple studies in order to appropriately describe the range(s) of parameter values that is compatible with the data and the value(s) that is most compatible with the data.

- Where indicated by the combined data from multiple available studies, a model with both linear and higher order terms may be chosen as the "best fit" description of the dose-response relationship. Whether to incorporate one or more higher order terms, however, should be determined in specific cases by weighing the merits of improving the fit to the data versus maintaining reasonable parsimony in the model.

DATA GAPS AND RESEARCH NEEDS

The panel identified a number of areas where research could help resolve important issues in the assessment of cancer risks for PCB environmental mixtures. These areas are listed below along with general recommendations pertaining to each of them.

- Measurement of levels of PCB congeners in environmental samples
  - — Recommend standard analytical methodologies (including sample preparation) for measuring congener-specific PCB residue levels in environmental samples. Recommend sample preparation methods for environmental samples such as soil, sediment, air, water, human breast milk, dairy products, foodstuffs (especially eggs, poultry, fish, and shellfish), and for marine, freshwater, and terrestrial biota as needed.
  - — Develop a database on congener-specific PCB residue levels in environmental samples.
- Dose
  - — Determine the appropriateness of the $BW^{3/4}$ scaling factor for PCB environmental mixtures used as a default when extrapolating animal cancer data to humans.
  - — Explore structure-activity relationship (SAR) methods to predict physiologically based pharmacokinetic (PBPK) parameters for PCB environmental mixtures.
  - — Develop an appropriate dose metric for PCB environmental mixtures.
- Cancer Slope Factors
  - — Develop mechanism-oriented dose-response data for PCB environmental mixtures (e.g., examining promotional, hormonal, gender-related, genotoxic, or secondary effects of the mixtures).
  - — Using a broader range of test animals, quantify CSFs for commercial and environmental PCB mixtures.
  - — Identify significant carcinogenic PCB congeners in commercial and environmental PCB mixtures.
  - — Describe the mode(s) of action for each of these congeners.
  - — Quantify CSFs for these significant PCB congeners.

- — Use experimental and modeling approaches to generate these data.

- — Evaluate the consistency of existing and future epidemiology results with the carcinogenic potencies from the rodent studies.

- — Develop quantitative uncertainty distributions to express key sources of uncertainty.

- Age susceptibility: prenatal and early post-natal effects

  - — Determine sensitivities of fetuses and early neonates to the carcinogenic effects of PCB environmental mixtures.

  - — Determine risks for thyroid and urogenital/reproductive tract cancers in neonates and adults from exposures to PCB environmental mixtures.

- Mechanisms

  - — Develop mechanism-oriented dose-response data for PCB environmental mixtures (e.g., examining promotional, hormonal, gender-related, genotoxic, or secondary effects of the mixtures).

  - — Determine the mechanism(s) of PCB-induced liver cancer in rats, its similarity to the mechanism(s) of rat liver cancer induced by "nongenotoxic" carcinogens, its activities at low dose levels, and its relevance to humans.

APPENDIX A

FINAL REVIEWER LIST

EPA United States Environmental Protection Agency
National Center for Environmental Assessment

Peer Review Workshop on *PCBs: Cancer-Dose Response Assessment and Application to Environmental Mixtures*

Holiday Inn Bethesda
Bethesda, MD
May 21-22, 1996

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APPENDIX B

ISSUES CONSIDERED DURING REVIEW

Peer Review Workshop on *PCBs: Cancer Dose-Response Assessment and Application to Environmental Mixtures*

List of Issues To Be Considered During Review

Your review should focus on, but not be limited to, the following major scientific issues:

- Are the studies fairly represented? Are any studies pertinent to dose/response assessment missing?
- Is a range (instead of a single value) appropriate to represent the cancer potency of PCB mixtures? If so, is the exposure pathway a reasonable default indicator of which end of the range is appropriate?
- Is it important that exposure assessments include internal exposure that persists after external exposure stops? If so, is half-life in the body a reasonable way to do this given the information currently available to risk assessors?
- Is the assessment correct in identifying food chain exposure as highest risk? Is it sufficient to say "risks may be higher than those estimated in this assessment" or should the risk estimates be multiplied by an explicit food-chain adjustment factor?
- Is this assessment's approach of a range indexed by exposure pathway likely to be useful for incorporating new information? Should it be considered for non-cancer assessments?

Compared with past assessments, this assessment reflects some changes in guidelines and approach. Any reaction to the new features in this assessment would be welcome.

APPENDIX C

PREMEETING COMMENTS

EPA

U.S. ENVIRONMENTAL PROTECTION AGENCY

PEER REVIEW WORKSHOP ON *PCBs: CANCER-DOSE RESPONSE ASSESSMENT AND APPLICATION TO ENVIROMENTAL MIXTURES*

Premeeting Comments

Bethesda, MD
May 21-22, 1996

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LIST OF ISSUES TO BE CONSIDERED DURING REVIEW

Peer Review Workshop on *PCBs: Cancer Dose-Response Assessment and Application to Environmental Mixtures*

List of Issues To Be Considered During Review

Your review should focus on, but not be limited to, the following major scientific issues:

- Are the studies fairly represented? Are any studies pertinent to dose/response assessment missing?

- Is a range (instead of a single value) appropriate to represent the cancer potency of PCB mixtures? If so, is the exposure pathway a reasonable default indicator of which end of the range is appropriate?

- Is it important that exposure assessments include internal exposure that persists after external exposure stops? If so, is half-life in the body a reasonable way to do this given the information currently available to risk assessors?

- Is the assessment correct in identifying food chain exposure as highest risk? Is it sufficient to say "risks may be higher than those estimated in this assessment" or should the risk estimates be multiplied by an explicit food-chain adjustment factor?

- Is this assessment's approach of a range indexed by exposure pathway likely to be useful for incorporating new information? Should it be considered for non-cancer assessments?

Compared with past assessments, this assessment reflects some changes in guidelines and approach. Any reaction to the new features in this assessment would be welcome.

REVIEWERS' PREMEETING COMMENTS

Lucy Anderson

PCBs: Cancer Dose-Response Assessment and Application to Environmental Mixtures
L.M. Anderson, May, 1996: Comments

Summary. As a general comment, a risk-assessment data base consisting of liver tumors caused in rats by lifetime exposure to high doses of PCBs seems too limited, for a class of chemicals that act primarily through a tumor-promotion mechanism, and which may target other tissues in addition to liver. Epidemiology has not yet addressed the possibility of tumor promotion in humans by PCBs. Data are presented and summarized showing that, on a per-dose basis, PCBs can be more effective as tumor promoters, than as complete carcinogens after chronic lifetime exposure. While more initiator-dose/promoter-dose assays are needed, the greater likelihood of initiating events in humans vs laboratory rats argues for integration of promotion effects into risk analysis at the present time.

Additional pertinent studies are suggested for inclusion in the document, relating to specific congener effects, interations, correlations with neoplasia endpoints, tissue retention, and toxicokinetics.

Introduction and general comments. The document under consideration addresses quantitative cancer dose-response assessment for PCBs, including application to environmental mixtures and incorporation of factors such as partitioning, chemical transformation, bioaccumulation of congeners, route of exposure, and half-life in the body. Thus an objective is to develop an approach that is more complex and more accurate than the current, single, dose-response slope guideline. The quantitative data used as the basis for the calculations are numbers of liver and stomach tumors and leukemias/lymphomas in rats fed varying concentrations of commercials PCBs in diet for all or a significant fraction of their lifetime. Human data have not been utilized because of their current inadequacy.

Two general questions can be raised about the data base that is used. The first is that the quantitative evaluation is based almost entirely on the responses of rat liver, which may or may not be informative as to human effects, qualitatively in terms of target organ, or quantitatively in terms of sensitivity. Epidemiology has indicated that liver may be a target also in humans, but in addition melanoma is mentioned (p. 4, l. 10), data for other tissues

have been suggestive in various studies, and rumor has it that important new findings will be shortly forthcoming. It would be useful to have data from another animal model system for comparison with the female rat liver results.

Secondly, the epidemiological studies have not considered, through nested or co-variant analyses, synergistic or interactive effects *re* other exposures (e.g., smoking, consumption of well-done meats containing arylamine mutagens, etc.), an approach that seems essential for a group of chemicals that probably act through a tumor-promotion mechanism primarily, at least in the sense that prolonged exposure is required. In the absence of tumor initiation, liver tumors arise in rodents only after chronic treatment with relatively high doses of PCBs over a significant fraction of their lifetime. This exposure situation may have limited relevance to humans, whose exposure is much lower and usually intermittent. It could be proposed that, if PCBs are carcinogenic in humans, it is often because they are promoting cancers that have been initiated by genotoxic carcinogens (nitrosamines, polycyclic aromatic hydrocarbons, arylamines, mycotoxins, etc.) and/or by endogenous processes as a function of aging (reactive oxygen species, loss of DNA repair capacity, etc.). Therefore, dose-response determination for PCBs should be based not only on chronic, high-dose PCBs-only studies, but also on tumor promotion dose-response data. Ideally these should include all of the known human-exposure genotoxic carcinogens, and also a comparison of life stages at the time of start of PCB treatment. Some of this data is already available for liver, and could be readily obtained for mouse lung and hairless mouse skin as well. Other animal target tissues that have been shown to be subject to tumor promotion by assorted agents include breast, stomach, large bowel, thyroid, and kidney. Whether these are responsive to PCBs remains to be determined.

This issue is particularly important because PCBs can have a tumor-promoting effect at doses, including single doses, much lower than the total doses required for carcinogenesis by PCBs given alone. This obviously could have a major impact on quantitative risk assessment. As an example, I will offer some of our data, on promotion of mouse lung tumors initiated by N-nitrosodimethylamine. The graph shown below is from Anderson, *et al.*, 1991. It can be seen that, 28 weeks after *a single i.g. dose* of Aroclor 1254, there was a significant, dose-responsive tumor promotion effect. This was of statistical significance with

500 mg/kg, and the small increase after 50 mg/kg is probably real.

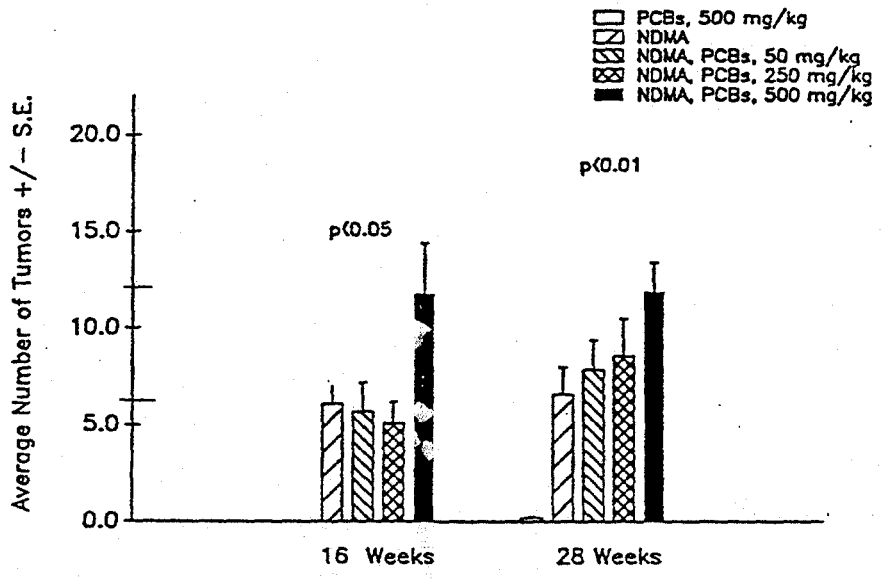


Figure 1 Infant male Swiss mice received NDMA, 5 mg/kg, i.p. on postnatal day 4 and/or PCBs in olive oil i.g. at the dose indicated on day 8. Data are from ref. 11, with permission. P values indicate statistical significance by the 2-tailed Student's t-test. The numbers of lung tumors are higher than those found after the same dose of NDMA given to Swiss mice in the new study shown in Fig. 2 and Table 1 and those reported in ref. 37. Current efforts suggest that the difference is not attributable to technical error but may reflect diurnal variation in susceptibility to tumor initiation. This hypothesis is under test. In any case, it is clear that the promotive effect of the PCBs occurs to the same extent with either a low or a high degree of initiation of lung tumors.

In a later study, we examined tumor promotion in liver also. As the table below indicates, a 10-fold increase in liver tumor incidence was seen at one year of age, with a single dose of 250 mg/kg given after NDMA (Anderson, *et al.*, 1994). For comparison, the largest carcinogenic effect in rat liver attained with Aroclor 1254 given alone in the diet seems to be in the new GE study with female Sprague-Dawley rats, where 100 ppm caused a 49-fold increase in liver tumor incidence compared with untreated controls. If this was a lifetime study, the total dose was about 3500 mg/kg. Thus, a 14-fold increase in total dose (3500/250) related to a 4.9-fold increase in effect (49/10). It must of course be remembered that rats and mice are being compared here.

Table I. Incidences and sizes of lung and liver tumors initiated by NDMA with or without PCB treatment

Treatment	n	Age (weeks ± SD)	Lung tumors			Liver tumors[a]		
			No. with tumors	Average No.[b] (± SD)	Average size[c] (mm ± SD)	No. with tumors	Average no.[b,d] (± SD)	Average size[c] (mm ± SD)
NDMA	76	16±0	26 (34%)	0.6±1.0	0.6±0.2	0		
NDMA−PCB	66	16±0	27 (41%)	0.7±1.0	0.6±0.3	0		
PCBs	75	16±0	1 (1.3%)	0.01±0.12	0.8	0		
Saline/oil	60	16±0	1 (1.6%)	0.02±0.12	0.5	0		
NDMA	23	28±0	7 (30%)[f]	0.5±1.1[g]	0.8±0.3	3 (13%)	0.1±0.3	0.4±0.1
NDMA−PCB[e]	27	28±0	19 (70%)[f]	1.9±2.9[g]	0.8±0.4	2 (7%)	0.1±0.3	0.3±0
NDMA	25	52±0	12 (48%)	0.6±0.8[h]	1.4±1.3	1 (4%)[i]	0.04±0.2	0.6
NDMA−PCB	23	51.8±0.1	15 (65%)	2.7±3.8[h]	1.4±0.4	9 (39%)[i]	0.6±0.8	0.6±0.5
PCBs	24	50.7±5.7	4 (17%)	0.17±0.38	1.1±0.6	0		
Saline/oil	27	51.2±4.0	6 (22%)	0.26±0.40	0.9±0.7	0		
NDMA	23	67.8±6.9	21 (91%)	5.1±4.5	3.1±2.7[j]	16 (70%)	1.5±2.2	0.7±0.5
NDMA−PCB	25	64.5±9.7	17/23 (74%)	3.9±4.3	2.4±1.6	14 (56%)	1.5±2.0	0.8±0.4
PCBs	25	66.1±9.7	17 (68%)	0.9±0.8	1.6±0.8	0		
Saline/oil	39	69.3±8.6	17 (44%)	0.6±0.7	1.5±1.1[j]	0		

[a]Liver tumors were adenomas, except for one carcinoma at 52 weeks with NDMA−PCB and at 72 weeks eight carcinomas with NDMA-only and three carcinomas with NDMA−PCB. Also at 72 weeks, four livers after NDMA and one liver after NDMA−PCB had coalesced tumors.
[b]Average number per mouse at risk.
[c]Average width in the largest dimension in the gross fixed (lung) or gross fresh (liver) tissue was calculated for each mouse. These mean values were then averaged to obtain the data given.
[d]Average for all mice with countable (not coalesced) liver tumors.
[e]At this time point there were no tumors after PCBs only (13 animals) or saline/oil (16 animals).
[f–j]Numbers with matched superscripts are significantly different.
[f]$P = 0.01$, Fisher's exact test.
[g]$P = 0.0033$, Wilcoxon test.
[h]$P = 0.0496$, $P = 0.032$ with zero values excluded, Wilcoxon test.
[i]$P = 0.004$, Fisher's exact test.
[j]$P = 0.049$, Student's t-test; $P = 0.0024$, Wilcoxon test.

Few studies have carried out dose-response studies on rat liver tumor promotion by PCBs, with tumors as endpoint. The most systematic data pertain to preneoplastic liver foci and are contained in three publications by Deml and Osterle (1982 and 1987, and Osterle and Demle, 1984). Liver foci were initiated in Sprague-Dawley rats of different ages and promoted with Clophen 50. A total dose of 350 mg/kg caused a 9- to 14-fold increase in number of foci (depending on the marker enzyme), after 7 weeks of exposure (1982). A more detailed dose-response study was reported in the 1984 paper (see tables below), showing a significant 2-fold effect of the lowest dose tried, 14 mg/kg, in adults initiated with a total of 96 mg/kg NDEA, and a significant 2.5-fold effect with 70 mg/kg PCBs total, in females initiated as weanlings with a single 8 mg/kg NDEA dose. In the adult-initiated rats, a 10-fold increase in number of foci resulted from a total PCBs dose of 350 mg/kg. The weanling model was pursued further (1987), and it was found that a total of 33 mg/kg over 11 weeks was marginally promotive (2-fold effect), given over 11 weeks (1987). This would be

equivalent to ~1 ppm lifetime exposure to A1254, wich might extrapolate to ~6% tumor incidence from the data in Table 5-1. A large 5.5-fold effect was attained with 165 mg/kg. It may be noted that relatively short follow-up times were utilized in these studies. Our findings with single-dose treatment suggest that much larger effects and/or lower minimal doses might have been demonstrated, had the animals lived a larger fraction of their lifespan.

Table II. Effect of Clophen A 50[a] on DEN[b]-initiated, enzyme-altered islands in livers of adult female Sprague-Dawley rats and percentage of coincidence of ATPase-deficiency, emergence of GGTase, and glycogen storage

Group	Treatment Clophen A 50 (mg/kg body weight)	Island number/cm² ATPase (−)	GGTase (+)	Glycogen (+)	Total area (mm²/cm²) ATPase (−)	GGTase (+)	Glycogen (+)	Coincidence
1	DEN –	13 ± 3	10 ± 1	4 ± 2	0.20 ± 0.05	0.08 ± 0.03	0.05 ± 0.02	11%
2	DEN 2	26 ± 11[c,d]	13 ± 2	6 ± 2	0.5 ± 0.2[c,d]	0.22 ± 0.16	0.12 ± 0.06	29%
3	DEN 10	50 ± 7[c,d]	15 ± 5	11 ± 4	1.0 ± 0.2[c,d]	0.45 ± 0.38	0.26 ± 0.09	33%
4	DEN 25	62 ± 9[c,d]	40 ± 5	28 ± 10	1.3 ± 0.2[c,d]	0.95 ± 0.12	0.41 ± 0.18	51%
5	DEN 50	130 ± 24[c,d]	82 ± 26	42 ± 9	1.5 ± 0.3[c,d]	0.63 ± 0.23	0.28 ± 0.11	46%
6	DEN 100	150 ± 28[c,d]	89 ± 19	85 ± 42	2.4 ± 1.4[c,d]	1.72 ± 1.00	1.70 ± 1.64	74%
2a	2	0.2 ± 0.2	2.0 ± 2.2	0.6 ± 0.4	0.003 ± 0.002	0.003 ± 0.003	0.004 ± 0.004	n.d.
3a	10	0.7 ± 0.3	1.5 ± 1.0	1.8 ± 0.4	0.01 ± 0.007	0.002 ± 0.002	0.004 ± 0.003	n.d.
4a	25	1.5 ± 0.3	1.3 ± 0.9	2.0 ± 0.1	0.17 ± 0.002	0.002 ± 0.001	0.008 ± 0.003	n.d.
5a	50	4.1 ± 1.1	1.6 ± 1.1	0.8 ± 0.6	0.10 ± 0.050	0.057 ± 0.060	0.020 ± 0.020	n.d.
6a	100	2.0 ± 0.5	2.3 ± 1.0	n.d.	0.05 ± 0.040	0.064 ± 0.080	n.d.	n.d.
Control (olive oil)		0.06 ± 0.03	0.2 ± 0.1	0.5 ± 0.5	0.008 ± 0.002	0.004 ± 0.001	0.002 ± 0.001	

[a]Adults: 12 x 8 mg/kg body weight; 4 animals per group.
[b]Clophen A 50 was applied for 7 consecutive weeks. Further experimental details are given in Materials and methods.
[c]Significantly different from group 1 ($p \leq 0.01$, t-test).
[d]Significantly different from the respective lower dosed group ($p \leq 0.01$, t-test).
n.d., not determined.

: III. Effect of Clophen A 50[a] on DEN[b]-initiated, enzyme-altered islands in livers of weanling female Sprague-Dawley rats and percentage of coincidence of ase-deficiency, emergence of GGTase, and glycogen storage

p	Treatment Clophen A 50 (mg/kg body weight)	Island number/cm² ATPase (−)	GGTase (+)	Glycogen (+)	Total area (mm²/cm²) ATPase (−)	GGTase (+)	Glycogen (+)	Coincidence
	DEN –	12 ± 4	5 ± 2	5 ± 2	0.2 ± 0.01	0.03 ± 0.002	0.03 ± 0.01	18%
	DEN 2	13 ± 2	4 ± 1	4 ± 1	0.1 ± 0.04	0.02 ± 0.007	0.02 ± 0.01	15%
	DEN 10	29 ± 5[c,d]	7 ± 1	8 ± 3	0.4 ± 0.09[c,d]	0.07 ± 0.03	0.06 ± 0.04	13%
	DEN 25	22 ± 4[c]	15 ± 2	9 ± 1	0.4 ± 0.05[c]	0.16 ± 0.02	0.08 ± 0.03	44%
	DEN 50	87 ± 13[c,d]	39 ± 7	33 ± 6	1.5 ± 0.60[c,d]	0.97 ± 0.30	0.53 ± 0.17	36%
	DEN 100	89 ± 10[c]	60 ± 13	31 ± 3	2.3 ± 0.50[c]	0.90 ± 0.30	0.40 ± 0.07	67%
	2	0.4 ± 0.3	0	0	0.004 ± 0.004	0	0	n.d
	10	0.4 ± 0.3	0	0	0.003 ± 0.003	0	0	n.d.
	25	0.7 ± 0.5	1.6 ± 1.3	2.5 ± 0.6	0.005 ± 0.005	0.001 ± 0.001	0.010 ± 0.004	n.d.
	50	2.2 ± 0.4	0.6 ± 0.4	0.9 ± 0.3	0.02 ± 0.007	0.006 ± 0.004	0.005 ± 0.002	n.d.
	100	2.1 ± 0.9	1.7 ± 0.4	0.8 ± 0.5	0.02 ± 0.010	0.030 ± 0.010	0.012 ± 0.009	n.d.
:rol (olive oil)		0.3 ± 0.1	0	0	0.005 ± 0.001	0	0	

anlings: 1 x 8 mg/kg body weight; 4 animals per group.

The experiments of Nishizumi (1976) utilized male Wistar rats, again NDEA as the initiator, and followed grossly-visible tumors (adenomas and carcinomas), up to 24 weeks. Kanechlor 500, at an approximate total dose of 26 mg/kg, resulted in a doubling of numbers of large (> 5mm) tumors at 24 weeks. Numbers of smaller tumors (>2 mm) showed an increase which depended on the initiating dose of NDEA, 13-fold for the higher dose.

These studies together confirm that in rat liver PCBs act more effectively on initiated foci or tumors, on a per-dose basis, even with short endpoints in time, than do lifetime PCBs alone on uninitiated liver, but that dose of initiator is probably an important variable. Thus, more detailed dose-of-initiator/dose-of-promoter studies, with several models and including lifetime treatment, would be needed before this promotion component could be factored quantitatively into risk evaluation. However, since most human beings are more likely to have sustained tumor-initiating events than laboratory rats, it is suggested that, for conservative risk estimates, the possibility of tumor promotion should be taken into account.

Specific questions to be discussed:

I. Presence of all pertinent studies

It would seem that, for the proposed detailed approach, more information about the properties of specific congeners and their interactions should be included. The most relevant would be those related to neoplasia as an endpoint, and to toxicokinetics. Some data of which I am aware, including some from this Laboratory, are shown below.

A. Promotion of mouse lung and liver tumors by PCB congeners (*re*: Section 2.3.)

In a study of promotion of NDMA-initiated lung tumors in mice, we tested congeners 2,2',4',4',5,5'-HCB (BZ #153) and 2,2',3,4,4',5'-HCB (#138), singly and in combination, at concentrations equivalent to their amount in a 500 mg/kg A1254 dose. These are the congeners that are retained in largest quantity in the bodies of mice after Aroclor 1254. The results are shown in the table below (from Anderson, *et al.*, 1991). Congener #138 caused a 2.4-fold increase in lung tumor multiplicity at 16 weeks, similar to the 2.1-fold increase seen with 500 mg/kg A1254 in a previous study. Congener #153, by contrast, did not promote, and when given along with #138, partially abrogated the latter's tumor promotive effect.

Since #138 is an Ah receptor agonist, and #153 is not, the results tend to implicate this receptor in promotion. The promotive effect of the A1254 mixture is probably not due entirely to #138, in view of the abrogating effect of #153. Later studies implicate #105 (see below). Also, the promoting effects of A1254 for mouse liver tumors (Anderson, *et al.*, 1994; Beebe, *et al.*, 1995) could be added to Table 2-2, which currently indicates that only foci have been quantified.

Table 1 Effects of HCB Congeners as Lung Tumor Promoters

Treatment[a]	No. of mice	No. with tumor	Av. no. tumors per mouse ± S.E.
NDMA	55	15 (27%)	0.42 ± 0.11[b]
2,2',4,4',5,5'-HCB	32	0	
2,2',3,4,4',5'-HCB	31	0	
2,2',4,4',5,5'- + 2,2',3,4,4',5'-HCBs	34	0	
NDMA + 2,2',4,4',5,5'-HCB	53	13 (24%)	0.30 ± 0.08[c]
NDMA + 2,2',3,4,4',5'-HCB	50	21 (42%)	1.0 ± 0.3[b,c,d]
NDMA + 2,2',4,4',5,5'- + 2,2',3,4,4',5'-HCB	46	14 (30%)	0.52 ± 0.13[d]
Saline/oil	26	0	

[a]Male Swiss mice [Cr:NIH(s)] were treated i.p. on postnatal day 4 (day of birth was day 1) with NDMA, 5 mg/kg (Sigma Chemical, St. Louis, Missouri) in saline or saline only. On postnatal day 8 they received i.g. the PCB congeners indicated (Ultra Scientific, North Kingstown, Rhode Island) in olive oil, each at a dose of 20 mg/kg. They were sacrificed at 16 weeks of age. Assay of randomly selected carcasses confirmed the presence of the two congeners in approximately equal quantities. Lung tumors were quantified in 1-mm hand sections of Bouin's fixed lungs with the aid of a dissecting microscope and confirmed in histological sections.

[b]p = 0.043, 2-tailed Student's t-test, significance of difference between the two values.

[c]p = 0.014, 2-tailed Student's t-test, significance of difference between these numbers.

[d]p = 0.10, 2-tailed Student's t-test, significance of difference between these numbers.

B. Correlations with congener #99 (*re*: Section 2.3./2.4.)

In a study of mouse lung tumor promotion, #99 (2,2',4,4',5'-PCB, not an Ah receptor agonist) was the only one, of the nine congeners measured, whose carcass concentration correlated with lung tumor incidence (see below; Anderson, *et al.*, 1994). Lung concentrations were not measured in this study. The second table below is from a study of human breast cancers, in which levels of #99 in breast tissue were significantly associated with risk, but only in estrogen receptor-positive cases (Dewailly, *et al.*, 1994).

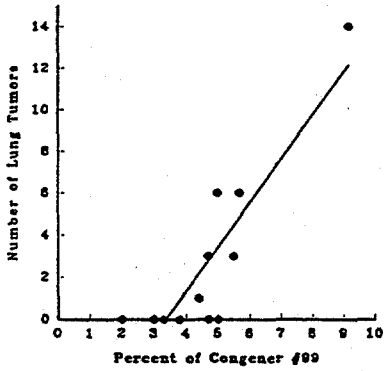


Fig. 1. Correlation of the percentage of total carcass PCBs as congener 99 with number of lung tumors in 12 mice killed at 28 weeks; $P < 0.01$ (see Table II).

Breast adipose tissue concentrations of organochlorines in women with benign breast disease (control subjects) or breast cancer with or without estrogen receptors (Quebec City, Canada, 1991–1992)

Organochlorine*	Control subjects (n = 77): concentration, μg/kg‡	Breast cancer case patients†: ER-negative (n = 9): Concentration, μg/kg‡	P§	ER-positive (n = 9): Concentration, μg/kg‡	P§
DDE	765.3 ± 526.9	608.9 ± 338.9	.63	2132.2 ± 2049.9	.01
HCB	33.4 ± 13.2	31.1 ± 11.5	.53	41.7 ± 15.5	.29
β-HCH	39.7 ± 23.4	34.7 ± 15.7	.92	39.7 ± 11.5	.77
Oxychlordane	31.1 ± 12.4	26.8 ± 7.4	.59	38.9 ± 13.8	.12
Transnonachlor	42.5 ± 17.8	34.8 ± 8.3	.37	50.3 ± 11.1	.07
Mirex	31.7 ± 28.2	13.2 ± 7.1	.04	18.2 ± 15.5	.20
PCB congener					
99	20.5 ± 11.7	14.6 ± 5.4	.18	30.7 ± 17.0	.05
105	6.0 ± 4.2	3.9 ± 1.8	.26	7.3 ± 5.1	.54
118	34.9 ± 20.1	19.1 ± 7.2	.02	37.7 ± 20.0	.91
138	70.1 ± 28.8	59.6 ± 15.0	.47	78.1 ± 26.8	.32
153	95.6 ± 36.3	82.2 ± 18.2	.50	100.0 ± 30.7	.59
156	17.9 ± 10.2	16.8 ± 5.3	.69	15.7 ± 3.9	.79
170	36.7 ± 18.3	28.2 ± 11.5	.22	30.6 ± 11.1	.61
180	86.2 ± 42.8	80.0 ± 25.3	.99	74.6 ± 21.7	.57
183	7.5 ± 2.0	6.8 ± 2.1	.50	10.1 ± 4.6	.12
187	20.4 ± 8.8	18.7 ± 7.3	.57	19.4 ± 6.5	.94
Total PCBs	397.0 ± 161.5	331.5 ± 74.7	.39	404.7 ± 130.7	.79

*HCB = hexachlorobenzene; β-HCH = β-hexachlorocyclohexane.
†ER status was missing for two case patients. ER-negative: ≤10 fmol/mg; ER-positive: >10 fmol/mg.
‡Concentrations are mean ± SD.
§*P* values are calculated with Wilcoxon rank sum test.

C. Specific congener retention and toxicokinetics (*re*: Section 2.4.)

Binding of methylsulfone derivatives of PCBs in lung has been studied extensively, and selective retention of hydroxylated PCB metabolites in blood was recently indicated for humans as well as rats and seals (Bergman, *et al.*, 1994). There have, however, been few systematic studies of the toxicokinetics of specific bioretained congeners in internal tissues. We carried out a detailed analysis of levels of the nine most prominent congeners in mouse liver, lung, and carcass after a single 250 mg/kg dose of A1254 to neonatal mice, at intervals up to 16 weeks (Anderson, *et al.*, 1993). The results showed specific retention of congeners in the two target tissues for tumor promotion, with the carcass behaving, as expected, as a passive, high-lipid storage compartment. Lung was a low-capacity but high-affinity binding compartment for all congeners except for #153: after one week there was no decrease except for that due to dilution. All of the congeners were retained in liver, relative to carcass, during the first 8 weeks, but then lost more rapidly during the last four weeks. Congener #105 (2,3,3',4,4'-PCB) was particularly likely to be retained in lung and liver. Several graphs are shown below.

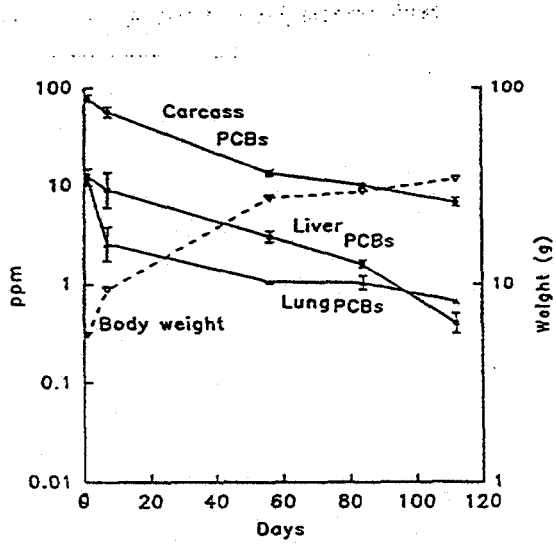
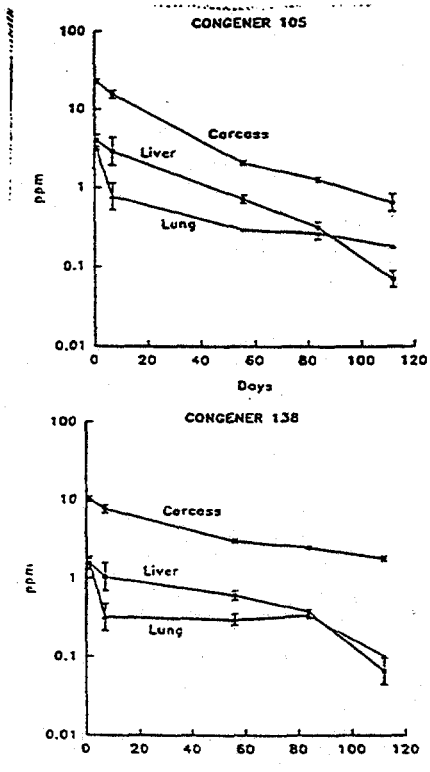


FIGURE 1. Geometric means of total PCBs in carcass, liver, and lung (solid lines) and of body weights (broken line) as a function of time after treatment with 500 mg/kg Aroclor 1254 on postnatal day 8.



Congener #105 was also significantly retained in mouse lung relative to carcass after 100 or 500 mg/kg doses of A1254 given to adult mice, whereas #118 was selectively lost (Beebe, *et al.*, 1992).

TABLE 2
PCBs in Carcass and Lung Over Time

	Carcass (C) or lung (L)	Total (mg/kg)	Percentage of total remaining PCBs								
			99	118	105	153	138	128	156	180	170
Aro 100											
48 hr	C	13.1 ± 3.6	10.01 ± 0.3	24.36 ± 0.8*	29.7 ± 0.3*	10.6 ± 0.3	14.5 ± 0.3	2.3 ± 0.2	6.2 ± 0.1	2.5 ± 0.06	1.2 ± 0.03
	L	1.52 ± 1.57	14.7 ± 2.0	15.1 ± 2.2	35.0 ± 3.6	11.7 ± 0.7	13.1 ± 0.4	3.2 ± 2.8	4.0 ± 1.0	2.3 ± 1.0	1.5 ± 1.3
96 hr	C	12.9 ± 3.8	16.1 ± 0.5	22.0 ± 0.7*	27.3 ± 0.5*	10.0 ± 0.3	13.6 ± 0.3	2.3 ± 0.0	4.9 ± 0.1	8.5 ± 0.1	1.4 ± 0.9
	L	1.05 ± 0.35	16.1 ± 1.4	15.5 ± 2.1	37.6 ± 3.8	10.0 ± 0.7	13.3 ± 0.6	3.3 ± 0.9	3.5 ± 0.4	1.6 ± 0.2	0.8 ± 0.1
1 wk	C	14.8 ± 3.2	10.6 ± 0.3	23.1 ± 0.7*	29.2 ± 0.7*	11.8 ± 0.4	14.6 ± 0.7	2.1 ± 0.1	4.7 ± 1.0	2.5 ± 0.1	1.2 ± 0.05
	L	0.814 ± 0.530	11.5 ± 3.4	13.2 ± 2.5	34.9 ± 2.7	11.2 ± 0.8	16.3 ± 1.4	2.8 ± 1.6	5.0 ± 3.0	3.2 ± 1.7	1.7 ± 1.2
4 wk	C	9.93 ± 2.7	10.4 ± 0.7	20.0 ± 1.2*	23.7 ± 1.6*	15.1 ± 1.4	17.1 ± 1.0	1.9 ± 0.2	6.5 ± 0.5	3.5 ± 0.4	1.35 ± 0.08
	L	0.961 ± 0.703	10.2 ± 4.2	12.0 ± 2.2	30.7 ± 2.6	13.8 ± 0.7	19.2 ± 2.5	2.2 ± 0.8	6.2 ± 2.1	4.1 ± 1.7	1.6 ± 0.6
12 wk	C	10.1 ± 3.89	11.9 ± 2.9	14.6 ± 3.3*	18.7 ± 5.3*	19.3 ± 5.0	20.2 ± 3.2	1.4 ± 0.3	7.7 ± 1.6	4.8 ± 1.6	1.4 ± 0.10
	L	0.506 ± 0.338	12.0 ± 3.0	8.2 ± 0.10	35.7 ± 9.6	13.1 ± 1.6	20.8 ± 3.7	3.4 ± 4.4	4.0 ± 2.1*	2.6 ± 2.1	4.7*
30 wk	C	2.8 ± 2.2	3.9 ± 3.8	4.3 ± 4.6	5.1 ± 5.3*	35.4 ± 8.2	21.4 ± 3.5	1.06 ± 0.7	15.9 ± 2.6	13.6 ± 5.8	1.4 ± 0.87
	L	0.27 ± 0.10	4.2 ± 2.2	2.8 ± 1.8	12.7 ± 9.0	40.8 ± 14.1	29.5 ± 15.2	1.45*	5.2 ± 1.5	5.7 ± 2.3	2.01*
Aro 500											
48 hr	C	57.7 ± 20.7	15.4 ± 1.9	21.9 ± 0.5*	29.7 ± 0.5*	10.8 ± 0.4	13.3 ± 0.4	0.8 ± 0.2	4.5 ± 0.2	2.1 ± 0.1	1.1 ± 0.1
	L	4.85 ± 3.99	15.8 ± 2.7	18.6 ± 0.7	33.7 ± 2.2	9.9 ± 1.0	11.4 ± 1.5	1.8 ± 0.4	4.0 ± 0.8	1.7 ± 0.4	0.9 ± 0.2
96 hr	C	79.2 ± 15.1	14.7 ± 0.2	21.2 ± 0.5	30.3 ± 0.3*	10.7 ± 0.4	13.8 ± 0.3	1.9 ± 0.2	4.3 ± 1.0	2.0 ± 0.03	1.0 ± 0.02
	L	7.15 ± 2.62	14.8 ± 2.5	19.9 ± 1.4	34.8 ± 1.2	10.1 ± 0.7	12.3 ± 1.0	1.6 ± 0.2	4.0 ± 0.6	1.8 ± 0.3	0.9 ± 0.1
1 wk	C	81.5 ± 19.3	14.0 ± 0.3	20.7 ± 0.6	28.6 ± 0.3*	11.5 ± 0.4	14.8 ± 0.2	2.0 ± 0.1	5.2 ± 0.2	2.3 ± 0.5	1.1 ± 0.03
	L	4.74 ± 3.74	13.3 ± 2.5	18.5 ± 2.1	35.0 ± 1.9	10.5 ± 0.8	13.3 ± 2.4	1.8 ± 0.3	4.8 ± 1.3	1.8 ± 0.6	1.0 ± 0.4
4 wk	C	52.0 ± 17.2	12.7 ± 1.4	19.7 ± 0.9	20.5 ± 2.8*	15.9 ± 1.8	19.0 ± 1.7	1.4 ± 0.2	6.4 ± 0.8	3.2 ± 0.5	1.2 ± 0.3
	L	3.47 ± 2.4	11.3 ± 2.8	15.7 ± 1.9	28.1 ± 4.6	14.5 ± 2.2	17.9 ± 2.6	1.4 ± 0.2	6.0 ± 1.1	3.8 ± 1.0	1.3 ± 0.3
12 wk	C	46.6 ± 18.3	8.7 ± 3.0	15.8 ± 3.3*	14.7 ± 6.9*	21.9 ± 5.13	23.4 ± 4.7	1.0 ± 0.3	8.9 ± 2.1	4.4 ± 1.4	1.3 ± 0.23
	L	1.27 ± 0.35	9.6 ± 0.9	11.5 ± 0.9	22.1 ± 2.4	18.6 ± 1.3	25.8 ± 2.6	1.2 ± 0.1	6.7 ± 0.3	3.9 ± 0.4	1.1 ± 0.1
30 wk	C	16.6 ± 7.6	2.8 ± 2.0	4.8 ± 3.1	3.3 ± 3.3	32.7 ± 4.7	25.5 ± 3.6	0.6 ± 0.3	16.4 ± 3.5	11.5 ± 3.9	1.5 ± 0.3
	L	0.6 ± 0.28	2.8 ± 1.3	3.4 ± 1.3	5.0 ± 2.0	28.4 ± 13.1	39.3 ± 11.3	1.2 ± 0.9	9.4 ± 2.3	9.9 ± 2.5	1.2 ± 0.3

* Significantly different from corresponding lung value, $p < 0.05$.

* Not detectable in all lungs within a group.

Congeners are identified by BZ designations. Chemical names for each congener are as follows: 99, 2,2',4,4',5-pentachlorobiphenyl; 118, 2,3,4,4',5-pentachlorobiphenyl; 105, 2,3,3',4,4'-pentachlorobiphenyl; 153, 2,2',4,4',5,5'-hexachlorobiphenyl; 138, 2,2',3,4,4',5'-hexachlorobiphenyl; 128, 2,2',3,3',4,4'-hexachlorobiphenyl; 156, 2,3,3',4,4',5-hexachlorobiphenyl; 180, 2,2',3,4,4',5,5'-heptachlorobiphenyl; 170, 2,2',3,3',4,4',5-heptachlorobiphenyl.

Total clearance rates of the congeners from carcass were identical for the 100 and 500 mg/kg doses, but differences were seen between the congeners, with the pentachlorobiphenyls cleared more rapidly at the higher dose, but the hexa- and heptachlorobiphenyls in general eliminated more rapidly after the lower dose.

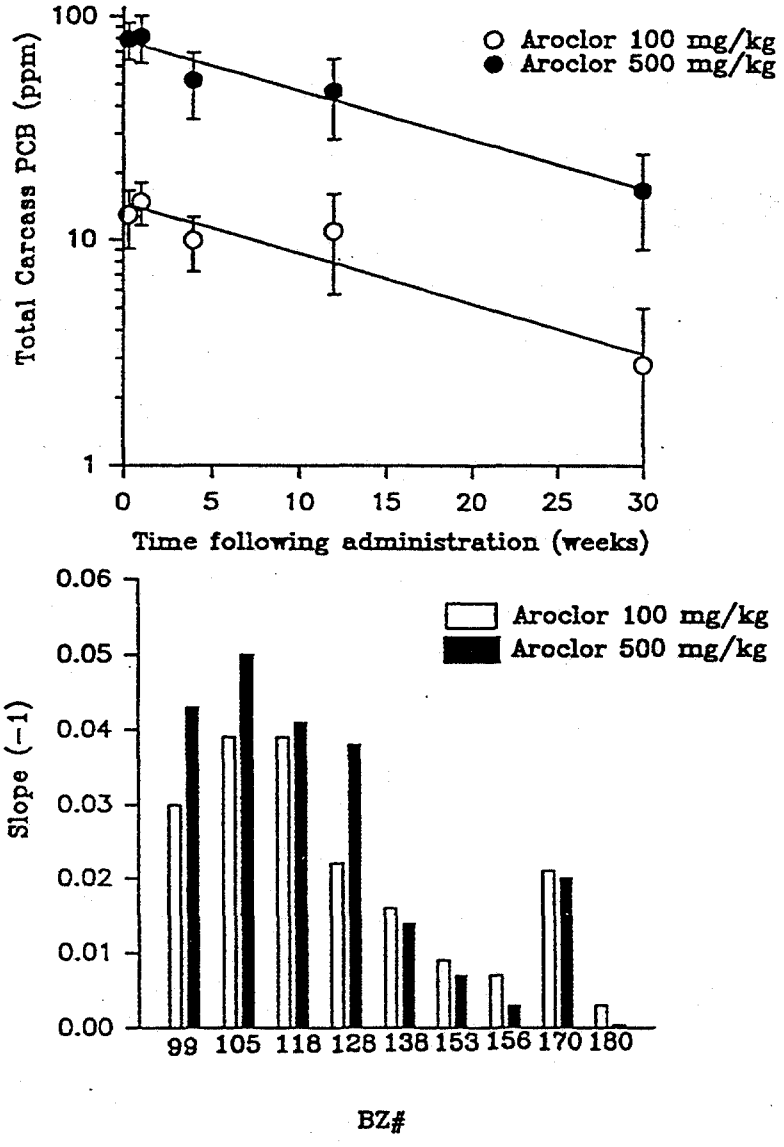


FIG. 5--Elimination of total PCBs from mouse carcass following Aroclor 1254 administration (TOP) and slopes of the elimination curves of several PCB congeners in carcass following each dose of Aroclor (BOTTOM) (Beebe *et al.*, 1990). Each value represents the mean ± standard deviation of 10-15 animals.

Data are also available for rats (Dragnev, *et al.*, 1994). In this study, A1254 was given in the diet, so results relate directly to the dietary tumorigenesis studies used for risk assessment. Both dose- and tissue-specific accumulation patterns were seen. Amounts in liver were dose-dependent, so the dose fed corresponded to relative internal dose to the liver. However, even at the lower doses, PCBs did not continue to accumulate over time. The liver appeared to maintain a more-or-less steady state of the chemicals. This is an important finding for risk assessment. If PCBs were to continue to accrue in a target organ over time, then net effective target-organ dose would be greater than the sum of dose over time, and more complex calculations would be required.

It is also of interest that the congeners with highest concentrations in the liver were #s 99, 105, and 118, all of which exhibited special behavior in mice (see above).

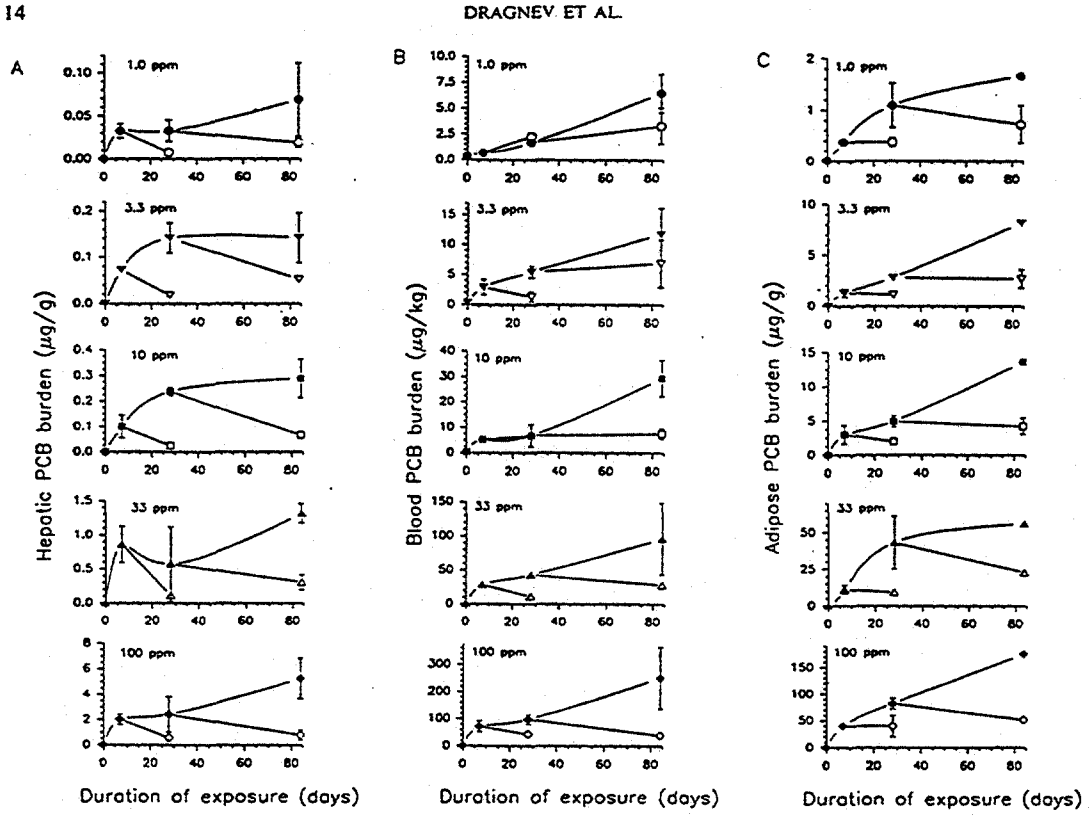


FIG. 1. Concentration and time-dependence of the hepatic (A), blood (B), or adipose (C) total PCB burden following administration of the indicated dietary concentrations of Aroclor 1254. Closed symbols represent PCB levels in rats following continuous administration of Aroclor 1254, while open symbols represent PCB levels in rats fed Aroclor for 7 or 28 days and thereafter fed control diet. Values given are means ± SD for three rats per treatment. Total PCBs are defined as the sum of the 10 major accumulated congeners. The specific congeners detected, and their approximate contribution to the total PCBs in liver are as follows: BZ 99 (25%), BZ 105 (15%), BZ 118 (15%), BZ 138 (11%), BZ 85 (10%), BZ 153 (9%), BZ 156 (9%), BZ 180 (2%), BZ 128 (2%), BZ 170 (2%). The substitution patterns for these congeners and the types of induction caused in rats (phenobarbital-type, TCDD-type, or mixed) are given under Materials and Methods.

D. Interactions between congeners (Section 2.3.)

Interaction between #138 and #153 in promotion of mouse lung tumors was noted above. Synergisms between congeners have been shown in rat liver, with foci numbers and/or volume as the endpoint. Sargent, *et al.* (1991) reported a 5.5-fold increase in the promotion index for 3,4,3',4'-TCB when given simultaneously with 2,5,2',5'-TCB, a much weaker promoter in itself. Similarly a more than additive effect on foci was recently described by Bager, *et al.*, for 3,4,5,3',4'-PCB (#126) and 2,4,5,2',4',5-PCB (#153), in female Sprague-Dawley rats initiated with partial hepatectomy/NDEA. On the other hand, such synergism was not seen for 3,3',4,4'- and 2,2',4,4',5,5'-PCBs in rats initiated with a high NDEA dose and fed a semisynthetic diet (Berberian, *et al.*, 1995).

II. Appropriateness of range, and use of exposure pathway for choice of end of range

Use of a range makes good sense, and exposure pathway is the parameter most likely to be available at present. Other data that could be included, in further refinements, might be: likely exposure to initiating agents; age at exposure to PCBs; congener profile in the tissues; congener interactions; and co-exposure to modifiers (for example, iron potentiates the hepatic carcinogenic effects of PCBs, whereas fruits and vegetables would be expected to reduce their effect). Much more animal data are needed on most of these points, before they could be factored in quantitatively.

III. Inclusion of internal exposure, and use of half-life

As noted above, internal exposure including target-specific retention of particular congeners would seem to be of critical importance to risk. Half-life values can be used, but should be specifically applied, with reference to the likely range of internal doses presenting a risk. Thus, if the body or target organ burden is high at the time of cessation of exposure, several half-lives may need to be added before the internal dose falls to a "safe" level.

IV. Food chain as source of highest risk, and need for an explicit food-chain adjustment factor

The food chain probably can be assumed to be the source of highest risk, except for specific occupational-exposure situations, for which different risk equations could be used.

V. Usefulness of the range/exposure approach for incorporating new information

This is probably as useful as any possible right now. As noted above, much more data are needed before more precise alternatives (e.g., a specific equation for each situation) are feasible.

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John Brown

Comments on Issues to be Considered During Peer Review Workshop in PCBs: Cancer Dose - Response Assessment and Application to Environmental Mixtures

Q.1. Are the studies fairly represented? Are any studies pertinent to dose/response assessment missing?

A.1. The literature on PCB environmental behavior and toxicology is enormous, and no 64–page document could hope to cover all of it. I shall comment later on a few significant studies relating to PCB accumulabilities, environmental composition, or composition-carcinogenicity relationships that were either overlooked or not available when the assessment document was written. As regards the specific area of dose/response assessment, however, I believe that all significant studies have been reviewed and are fairly represented in the document.

Q.2. Is range (instead of a single value) appropriate to represent the cancer potency of PCB mixtures? If so, is the exposure pathway a reasonable default indicator of which end of the range is appropriate?

A.2. To the first question, yes. Both the older and newer bioassay data show that tumorigenic potencies vary considerably from one PCB specimen to another, and it makes no more sense to assume a single potency for all types of PCBs than to assume one for all types of hydrocarbons. While this default may have applicability in many cases, there must be numerous situations that do not follow this default assumption. For example, PCB that have been anaerobically dechlorinated in sediments are considerably lower than expected in chlorination level, and particularly in dioxin equivalency. PCBs derived from incinerator emissions will vary sharply in the opposite direction (Brown et al., 1995. The Sources of The Coplanar PCBs. *Organohalogen Compounds 26*: 427-430.) This situation calls for a congener-specific analysis based on risk assessment, which could serve as an alternative to one based on a default assumption. Work on such an alternative is underway in our laboratory and is discussed below.

In parallel with the Battelle chronic bioassay of the four Aroclor specimens in male and female Sprague-Dawley rats, which produced the data on hepatotumorigenicity already communicated to EPA and cited in the draft assessment, we acquired congener-specific analytical data on adipose and liver PCB levels in the rats of all test groups and interim sacrifice points and have been using this data to identify the relationships between PCB composition and CSF. We hope shortly to have a full report on this work available as Appendix F of the Battelle Bioassay Study report. In the meantime, a summary containing the key data tables and figures that will appear in that report, along with a brief commentary, is attached to these comments as an appendix. Briefly, we found that the CSF of PCB mixtures appears to be contributed by both dioxin-like (only in females) and non-dioxin-like (in both sexes) activities just as indicated in the draft assessment document. These activities may be correlated with the total toxic equivalency (ΣTEQ) and total relative human accumulabilities (ΣRHA) of the PCB congeners present, using published values for the TEFs and RHAs. Thus, the CSF for any PCB composition for which congener-specific analytical data is available may be readily estimated. Accordingly, we would recommend that EPA subject our report to appropriate review and then consider allowing the use of the proposed relationship between CSF and PCB composition as an alternative to using exposure pathway as a default. We also note that use of such a relationship would not restrict the calculated CSF values to a limited range; for unusual PCB compositions, either lower or higher CSF values could be indicated.

Q.3. Is it important that exposure assessments include internal exposure that persists after external exposure stops? If so, is half-life in the body a reasonable way to do this, given the information currently available to risk assessors?

A.3. My answer to this question will come in two parts.

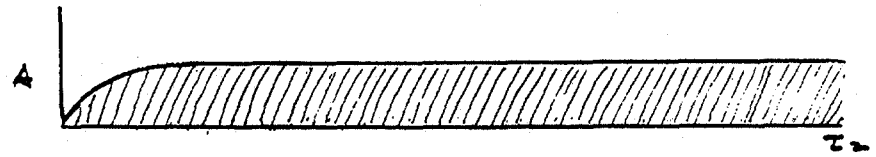
First, I do agree with the draft's position that PCBs are persistent in the human body. In addition, I believe the draft's documentation of that position could be strengthened by citing my 1994 paper on "Determination of PCB metabolic, excretion, and accumulation rates for use as indicators of biological response and relative risks," (*Environ. Sci. Technol. 28*, 2295-2305) which tabulates observed or estimated human clearance rates for the 140 more commonly measured PCB congeners in both normal individuals and chloracne

patients. The other literature sources cited as documentation of PCB persistence are generally also appropriate, except for the Luebeck et al. 1991 paper (cited on both p. 23 and 31) regarding the persistence of promotion of liver foci after cessation of dosing with 3, 4, 3', 4'-, but not with 2, 4, 2', 5'-tetrachlorobiphenyl. Both of these congeners are quite rapidly metabolized in rats (and in humans), and the 3, 4, 3', 4'-tetra (the "coplanar" PCB congener 77) is actually the more rapidly metabolized of the two. Thus, the persistence of the promotional effect must result from the persistence of some other chemical or biological change (a matter of current investigation in our laboratory) rather than persistence of that particular PCB congener. However, there are indeed some three dozen other PCB congeners that do persist, so that the question of continuing internal exposure after a short period of external exposure does merit examination.

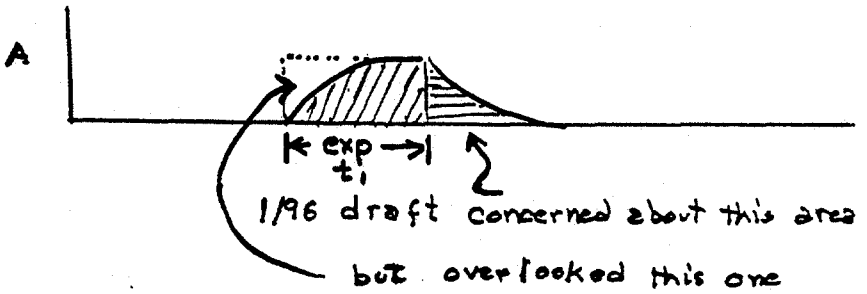
In the 1/96 draft document, this issue was addressed by estimating the impact of the residual internal PCB loading on the "area under the curve" (AUC) of an accumulation vs. time plot, a line of approach that I would endorse. Unfortunately, it would appear that the calculations of the AUC for the draft assessment omitted a significant term, and that after correction for this error the residual affect on relative AUC becomes too small and variable to be worth considered.

The basic modeling problem here is that when a slowly eliminated toxicant is administered for a finite time interval, not only will there be continued internal exposure after dosing has stopped, but there will also be a long time required to build the internal level of toxicant up to a steady state, so that there is a negative impact on AUC resulting from slow build-up that balances the positive impact of slow clearance. This is shown graphically on the attached sketch, which portrays idealized internal accumulation vs. timeplots for both lifetime and less-than-lifetime exposure, and gives an equation for the ratio of the areas. This shows that for periods of exposure that are long relative to $t_{1/2}$,

Internal Accumulation vs Time for Lifetime Exposure



Internal Accumulation vs Time for Partial Lifetime Exposure



Ratio of "Areas Under the Curve"
for partial (t_1) vs full (t_2) lifetime exposure

$$= \frac{\int_0^{t_1} [1 - \exp(-kt)]\,dt + \int_{t_1}^{\tau_2} [1 - \exp(-kt_1)] \exp[-k(t - t_1)]\,dt}{\int_0^{\tau_2} [1 - \exp(-k\tau)]\,d\tau}$$

Where k = clearance rate

$\Longrightarrow$ When $t_1 > t_{1/2}$, ratio $\sim \frac{t_1}{t_2}$

and occur early in life there should indeed be a correction for an exp (-kt) term that is proportional to half-life, but the correction should be applied to the denominator, as a subtraction from the presumed 70 year lifetime, rather than to the numerator as an addition to the time of exposure. This greatly diminishes its effect. Further, if the period of exposure occurs late in life the impact of the correction on the ratio of AUC's will be negative rather than positive. Thus, the correction can be either negative or positive, and small (typically less than 10%) in either case. If a mandate for a mathematically correct

calculation of this correction term were to be made part of an environmental risk assessment, it would require those doing such assessments, e.g. surveys of fish eating by fishermen, to determine not only consumption rates but also the ages of the fishermen, since this would be needed to establish whether the small addition to the AUC ratio for youngsters was outweighed by the diminished AUC ratio for retirees. In either case, the resulting correction term would be small relative to the uncertainty in the survey data. Accordingly, I would recommend that the Agency not bother trying to fix up its calculation of an adjustment factor, but instead drop the issue completely, so as to continue the present practice of presuming that for a less-than-lifetime period of exposure the relative risk is simply proportional to the ratio of the length of exposure to length of life, or t_1/t_2 in the diagram shown.

Q.4. Is the assessment correct in identifying food chain exposure as highest risk? Is it sufficient to say "risks may be higher than those estimated in this assessment" or should the risk estimates by multiplied by an explicit food-chain adjustment factor?

A.4. To the first part of this question, my answer is yes. Except in the few remaining situations where occupational exposure still occurs, all the reports of which I'm aware indicate the food chain, and specifically fish consumption, as the predominant source of human PCB exposure, and probably the only one that merits attention as a possible source of human cancer risk.

To the second part, my answer is probably not. As implied by my answer to Q.1., I've looked at a great deal of congener-specific analytical data on PCB composition in sediments, water, fish, and electrical workers, including that in the McFarland and Schwartz review papers cited in the 1/96 draft assessment document, and the papers that were reviewed during the preparation of the 1995 *Organohalogen Compounds* paper on coplanar PCB sources that I cited earlier. On the basis of this experience, the generalizations about fish PCB compositions that I'd currently regard as defensible are these: First, the overall tendencies of PCB homolog groups to bioaccumulate from the environment into fish (i.e., the bioaccumulation factors, whether BWAFs or BSAFs) appear to maximize at the Cl_5-Cl_6 levels of chlorination, with the accumulabilities being

slightly lower for the Cl_4BPs and Cl_7BPs, more so for the Cl_5 and Cl_8 homologs, and very low for the Cl_1, Cl_2, Cl_9, and Cl_{10} species. This means that the mean chlorination levels of the PCBs in fish from a water body contaminated with a more lightly chlorinated PCB composition, such as Aroclor 1242, will be increased (by about one Cl/BP), while the PCBs in fish from an area contaminated with Aroclor 1254 or 1260 will average about the same mean chlorination level as the contaminant. According to the relationship between CSF and PCB composition that we are evaluating, an increase in mean chlorination level, if it occurred, could either increase or decrease the CSF, depending on the magnitude of the TEQ. Second, anaerobic microbial dechlorination in sediments and P4501A-like metabolism in the fish both tend to attack the non-*ortho* and mono-*ortho* tetra- and pentachlorobiphenyl congeners selectively. The relative importance of these two processes in specific environments is often uncertain, but the levels of the mono-*ortho* penta's (i.e., congeners 105, 114, and 118, which are major contributors to the TEQ of Aroclor 1254) are generally considerably lower in fish than in Aroclor 1254, as may be noted from the McFarland review cited. Conversely, the levels of the non-orthos (especially congener 126, a major contributor of TEQ) may be higher in areas like the Great Lakes or Baltic Sea, where contributions of coplanar PCBs from combustion sources rather than Aroclors may be significant. Finally, the particular Aroclor 1254 sample that was used in the GE-Battelle rat bioassay appears to have had about four times the average PCB 126 level, and twice the TEQ, of ordinary Aroclor 1254 (which must at least partially explain the differences between in the Battelle and NCI findings for Aroclor 1254 tumorigenicity), so that any assumptions as to fish PCB CSFs based on its CSF will tend to be on the high side.

In short, I am not aware of any analytical data supporting the generalization that fish PCBs pose greater theoretical cancer risks than those of the Aroclors that have been bioassayed, and I am dubious about any unnecessary use of guesswork and generalizations as matters of public policy. As an alternative route to the estimation of CSFs for fish PCBs I would again call your attention to the possible use of congener specific analytical data and our observed empirical relationship between PCB composition and CSF. As I mentioned earlier, this would generate CSF values either above or below the range given in the draft assessment if the analytical data so indicated.

Minor point - On page 33, it is stated (without reference) that cooking PCB-contaminated food increases the PCDF contents. In actuality, the temperatures required to oxidize PCBs to PCDFs would char any food, and the oxidation doesn't occur in the presence of organic matter that is more easily oxidized than the PCBs. Deletion recommended.

Q.5. Is the assessment's approach of a range indexed by exposure pathway likely to be useful for incorporating new information? Should it be considered for non-cancer endpoints?

A.5. I'd like to emphasize that I think the Agency has, overall, done a good job of producing a draft risk assessment that recognizes the peculiarities of the PCB situation while still staying within the bounds defined by its basic policies and default assumptions. The alternatives that I've suggested in my answers to questions 2 and 4 are both based on information that was either not known at all or not well documented at the time the draft was written. The Agency clearly made an important step forward in recognizing that PCBs are mixtures of widely variable composition, and that these mixtures were most unlikely to all have the same CSF. The presumption that these CSF values would scale simply with chlorination levels was generally believed prior to the availability of the Battelle data. The presumption that exposure pathway would also be an indicator of chlorination level was one that was strongly suggested by equilibrium partitioning theory. In my view, the proposed linkage of CSF to exposure pathway represents a solution to a previously sidestepped problem that was creative, yet still completely in accord with the available data and contemporary scientific perceptions. It is a reasonable approach to be taken in the absence of congener-specific analytical data.

General Comment. Nevertheless, I saw in the 1/96 draft three significant shifts away from what I had believed to be time-honored Agency positions. The first, as just noted, was a shift away from the treatment of all PCB compositions as equally carcinogenic. The second was an evident willingness to consider tumorigenicity data from a new, very large, and carefully conducted study for which data has just become available. The third was

the effort to introduce an area-under-the curve analysis into the appraisal of the risks posed by a chronically accumulated toxicant, even though flawed. I want to commend the Agency for its willingness to take new positions on all these issues.

APPENDIX to J.F. Brown's Comments on Issues to be Discussed at May 21-22 Peer Review Workshop on PCBs: Cancer Dose-Response Assessment and Application to Environmental Mixtures.

This Appendix consists of excerpts from a draft report on Battelle Study No. SC920192, concerning the bioassay of Aroclors 1016, 1242, 1254, and 1260, and its Appendix F: "Characterization of PCB Composition, Tissue Accumulation, and Correlations with Tumorigenicity in Chronically Dosed Male and Female Sprague-Dawley Rats."

Contents

Conclusions from Battelle Study SC 920/92

DRAFT ***DRAFT*** ***DRAFT***

summary of Battelle Study

7.0 CONCLUSION

This study, which was designed to evaluate the chronic toxicity and oncogenicity of four PCB mixtures (Aroclor-1016, Aroclor-1242, Aroclor-1254 and Aroclor-1260), met all of the objectives set forth in the protocol. During the 2 years of dietary exposure to PCBs, daily observations for appearance and behavior did not reveal any clinical signs of toxicity attributable to treatment. Similarly, detailed evaluations during the first year of the study for neurotoxicity and neuropathology (which have been reported separately), lacked evidence of functional or morphological effects attributable to PCB exposure.

Survival for Aroclor-treated male rats was generally similar to control. Survival for females, on the other hand, was enhanced for all Aroclor mixtures, with the greatest increases evident for the Aroclor-1016, Aroclor-1242 and Aroclor-1260 Core treatment groups, and the Aroclor-1254 and Aroclor-1260 Stop Study groups.

Dose- and sex-dependent decreases in group mean body weight gains were measured in several (excluding Aroclor-1016) treatment groups (relative to control), with females exhibiting a greater response than males. Dose-related decreases in group mean body weights were clearly evident for both males and females for Aroclor-1254, with lesser effects also evident for females at the highest dietary concentrations of both Aroclor-1242 and Aroclor-1260. In all instances, when exposure to PCBs was discontinued (Stop Study animals), group mean body weights returned to control values, indicating reversibility in the effect. Although sporadic decreases in group mean feed consumption were measured for both males and females in the Aroclor-1254-treated groups, feed consumption was generally similar to control for all groups, with intake (mg/kg/day) of Aroclor test substances greater for females than males.

Measurements of hematologic indices in males showed group mean decreases, relative to control, in red blood cell parameters (Hb, Hct and MCH) for animals receiving Aroclor-1254. The decreases were most consistently measured at 105 weeks, the conclusion of the study.

Hematologic indices for females (RBC, Hb, Hct, MCV and MCH) showed both dose- and time-related group mean decreases, relative to control, for each Aroclor. The decreases were most notable for Aroclor-1254, followed by Aroclor-1260 ≈ Aroclor-1242 > Aroclor-1016.

Serum chemistries for males were generally unremarkable, with a suggestion for increased group mean aspartate aminotransferase (AST) and cholesterol in Aroclor-1254 groups (relative to control), which was not dose- or time-related.

Serum chemistries for females showed dose- and time-related group mean increases, relative to control, for GGT, AST and cholesterol for Aroclor-1242, Aroclor-1254 and Aroclor-1260, and for ALT for Aroclor-1254. The increases were most notable for Aroclor-1254, followed by Aroclor-1260 ≈ Aroclor-1242. These increases were attributed to alterations in hepatic function, which was supported by evidence of morphologic changes, as described below.

Sporadic treatment-related organ weight increases (normalized as organ-to-brain weight ratios) were identified for males for liver and thyroid gland. The group mean increases, relative to control, were not consistently dose- or time-related for either organ for any of the Aroclor test substances evaluated.

For females, group mean liver-to-brain weight ratios (relative to control) were generally increased in a dose- and time-related manner, with the magnitude and consistency of the effect following a pattern of Aroclor-1254 > Aroclor-1260 > Aroclor-1242 ≈ Aroclor-1016.

At necropsy, treatment-related macroscopic findings were most evident for liver from both males and females. The liver changes which were most notable were: enlargement, discoloration, foci, and nodules and masses. These changes generally correlated histomorphologically with hepatocellular hypertrophy, accumulation of pigment, areas of altered tinctorial properties, and neoplasms, respectively. The incidence and severity of macroscopic changes followed the same general pattern identified for histomorphologic lesions, as described below.

The key treatment-related histomorphologic diagnoses for the liver were hepatocellular hypertrophy, hepatic foci and neoplasms for both males and females. For males, the incidence of hepatocellular hypertrophy approached near maximum response by week 52, while the severity generally continued progressing in a dose- and time-related manner. For Aroclor-1260, the severity appeared to reach a maximum response by 52 weeks, while continued exposures showed slight increases for the remaining Aroclors, with time. The incidence of hepatic foci increased slightly with time for all Aroclor test substances and control, and with the exception of Aroclor-1016, were slightly greater than in control at 105 weeks. Hepatic neoplastic responses for Aroclor-treated males were not different from control, with the exception of a slight increase for Aroclor-1260 at 100 ppm. A dose-independent increased incidence for thyroid neoplasms (adenomas) was also noted in males for Aroclor-1242, Aroclor-1254 and Aroclor-1260, but not for Aroclor-1016.

For females, hepatocellular hypertrophy generally progressed in a dose- and time-related manner for each Aroclor test substance. The incidence of hepatocellular hypertrophy approached maximal response by 26 weeks, while the severity generally continued to increase thereafter, reaching maximal response by 78 weeks. Hepatic foci increased in incidence, for all Aroclors, with time, but lacked clear dose-dependence. Hepatic neoplastic responses (overwhelmingly diagnosed as adenomas) for Aroclor-treated females, showed clear dose- and Aroclor-related differences. Statistically significant

increased incidences were measured in females for all Aroclors at all exposure concentrations, except for Aroclor-1016 at 50 ppm. The neoplastic response differences between Aroclor mixtures followed a progression of Aroclor-1254 >> Aroclor-1260 > Aroclor-1242 >> Aroclor-1016. The incidence of mammary neoplasms was noted to be statistically significantly decreased for several Aroclor-treated groups. The decrease was particularly striking for Aroclor-1254 at 100 ppm, but was evident to a greater or lesser extent among groups from all Aroclors.

Discontinuing exposure to PCBs (Stop Study groups) resulted in a decrease (or lack of progression) in toxic, as well as protective, responses when compared with continued exposure groups. This is evidenced in females by the restoration of group mean body weights to control values, a decrease (or lack of progression) in the severity of hepatocellular hypertrophy, a decrease in the incidence of hepatic neoplasms (especially evident for Aroclor-1016 and Aroclor-1254 Stop Study/Core group comparisons), and a lack of inhibition (i.e. derepression) of mammary gland neoplasms (which occur spontaneously at a high incidence rate).

In conclusion, treatment-related toxicity was identified for all of the Aroclors evaluated. Chronic exposure to each Aroclor induced an increased incidence and severity of neoplastic and non-neoplastic lesions, which predominantly followed the pattern: Aroclor-1254 ≥ Aroclor-1260 > Aroclor-1242 > Aroclor-1016. It is important to also note that exposure to Aroclors increased survival and decreased the incidence of mammary gland neoplasia, and that withdrawal from exposure is associated with decreased toxic and protective responses.

Hepatocellular Tumor Incidence Data

		All Core Male Animals on Study											
Core Group Males	Group #	1	2	3	4	5	6	7	8	9	10	11	12
	Number	100	50	50	50	50	50	50	50	50	50	50	50
	Adenoma	4	1	1	2	0	3	2	2	6	2	5	6
	Carcinoma	3	1	1	2	1	1	2	2	0	1	1	3
	Hepatocholangiocarcinoma	0	0	0	0	0	0	0	0	0	0	0	0
	Hepatocholangioma	0	0	0	0	0	0	0	0	0	0	0	1
	% Liver Tumors	**7.0**	**4.0**	**4.0**	**8.0**	**2.0**	**8.0**	**8.0**	**8.0**	**12.0**	**6.0**	**12.0**	**20.0**

		All Core Female Animals on Study											
Core Group Females	Group #	1	2	3	4	5	6	7	8	9	10	11	12
	Number	100	50	50	50	50	50	50	50	50	50	50	50
	Adenoma	1	1	5	5	10	12	18	22	24	9	10	18
	Carcinoma	0	0	1	0	0	2	0	4	4	1	1	5
	Hepatocholangiocarcinoma	0	0	0	0	1	0	0	0	0	0	0	0
	Hepatocholangioma	0	0	0	0	0	1	1	2	0	0	0	1
	% Liver Tumors	**1.0**	**2.0**	**12.0**	**10.0**	**22.0**	**30.0**	**38.0**	**56.0**	**56.0**	**20.0**	**22.0**	**48.0**

APPENDIX F. Characterization of PCB Composition, Tissue Accumulation, and Correlations with Tumorigenicity in Chronically Dosed Male and Female Sprague-Dawley Rats.

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ABSTRACT

This Appendix presents data on: the pretreatment and chemical composition of the Aroclor specimens bioassayed in Battelle Study No. SC920192; the levels of Aroclor residues in the rats' tissues at successive time points; the PCB congener distributions in these retained PCBs; the relationships between dietary, adipose, and liver PCB levels; and the dependency of hepatotumorigenicity on PCB tissue levels and compositions.

The form of the latter dependency showed that there must be two processes whereby PCBs can contribute to rat liver tumor risk. One of these processes occurs only in females and correlates with the accumulation of dioxin toxic equivalency (TEQ) in the liver. The other process occurs in both sexes and correlates with the total PCB level in body lipids.

The observed dose vs. tissue accumulation and tissue accumulation vs. tumorigenicity relationships were used to derive an empirical relationship between PCB composition and cancer slope factor (CSF), using summed toxic equivalent factors (ΣTEF) and summed relative human accumulabilities (ΣRHA) as descriptors of PCB composition. Since TEF and RHA values for all environmentally significant PCB congeners have been published, use of the derived relationship should permit the calculation of a CSF value (shown to be conservative) for any environmental PCB specimen of known composition.

Commentary on Selected Tables and Figures

Table F-1. This table presents data on aggregate properties of the four Aroclors tested plus a laboratory reference specimen of Aroclor 1254 that appears to be more representative of run-of-the-mill production. The reported properties were calculated from individual congener levels as determined by either routine 118-peak DB-1 capillary GC analysis or special analyses for the PCDDs, PCDFs, and coplanar PCBs. RHA(70) denotes the relative human accumulability over a 70-year lifetime, as reported by Brown in *Environ. Sci. Technol. 28*, 2295-2305 (1994).

Tables F-2 through F-8 (not included) give details of the various tissue PCB accumulations at the various time points.

Figures F-1 through F-8 (not included) compare the PCB congener distributions in the rat tissues with those in the Aroclors, with a key given in Table F-9.

Table F-10. This summarizes the data from the omitted Tables F-1 through F-9, Figures F-1 through F-8 and also calculates the ratios of lipid-normalized liver to adipose PCB and TEQ as calculated from WHO TEF values. Note that the tendency of TEQ to accumulate in the liver falls rapidly with the level of Aroclor chlorination, except in the case of Aroclor 1016, where all the measurements of TEQ-contributing PCB peaks are in the noise range.

Figure F-9. This shows plots of tumor incidence versus lipid PCB concentrations for the various Aroclors in both male and female rats. Note that in males there appears to be a simple proportionality between lipid PCB and % rats with tumors, whereas in females there are large positive deviations from any such relationship for Aroclors 1242 and 1254.

Figure F-10. This figure shows that all of the data of Figure 9 for both sexes can be quite well described ($r^2 = 0.897$) by a simple linear equation that relates tumor risk (R) to total lipid PCB accumulation (A), liver TEQ (T), and a sex factor (F), having a value 1.00 in

females and 0.00 in males. Thus, PCB-derived TEQ, like dioxin itself, contributes to rat liver tumor risk only in females.

Table F-11. The relationship of Figure F-10 cannot be directly translated into a relationship between CSF and the accumulability and TEQ of a bioassayed Aroclor because while total lipid PCB accumulation is proportional to dose and total PCB accumulability (ΣRHA), liver TEQ is not similarly relatable to dose and administered ΣTEF. Instead, there is a roughly inverse dependence of liver TEQ on PCB accumulability (Table F-10), which probably has several causes. Accordingly, it appeared that the CSF would be best modelled in terms of the type of relation shown as eq. 2, which could be fitted to the data using the parameters indicated. The parameter a, which actually indicates the portion of the Aroclor 1260 CSF (in female rats) that is contributed by non-dioxin-like PCB activities, turned out to be about twice the similarly calculated Aroclor 1260 CSF in males (i.e., 0.43 per mg/kg-da vs. 0.22), which was in accord with the relative accumulabilities of the higher Aroclors in the two sexes. (Table F-10). The parameter b, which indicates the CSF for the dioxin equivalents in the PCBs, had a value of 7.0×10^4 per mg/kg-da.

The use of eq. 2, with parameters defined by the Aroclor test results, should allow the calculation of a CSF for any environmental PCB composition for which congener-specific analytical data is available.

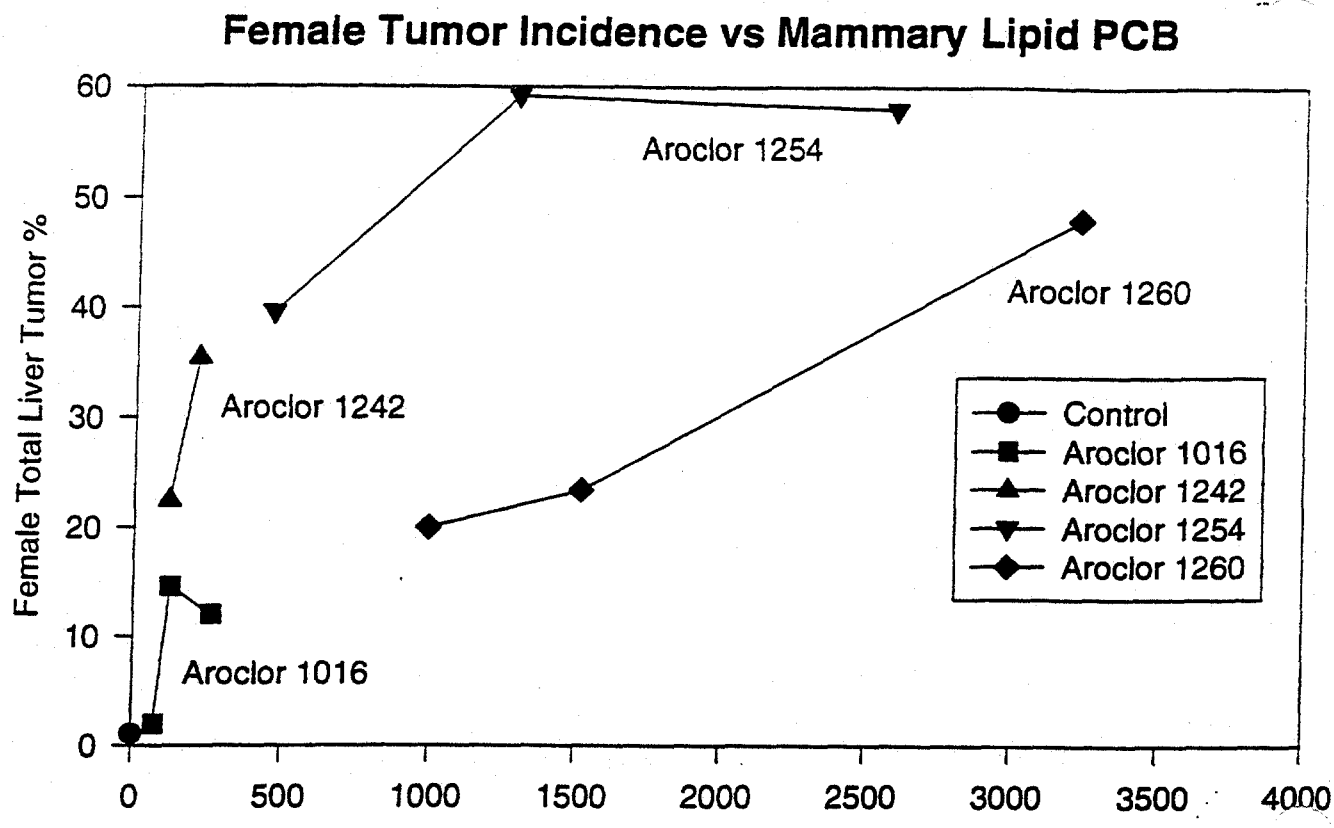
Table F-1. Summary of Aroclor Usage and Bioassayed Sample Composition

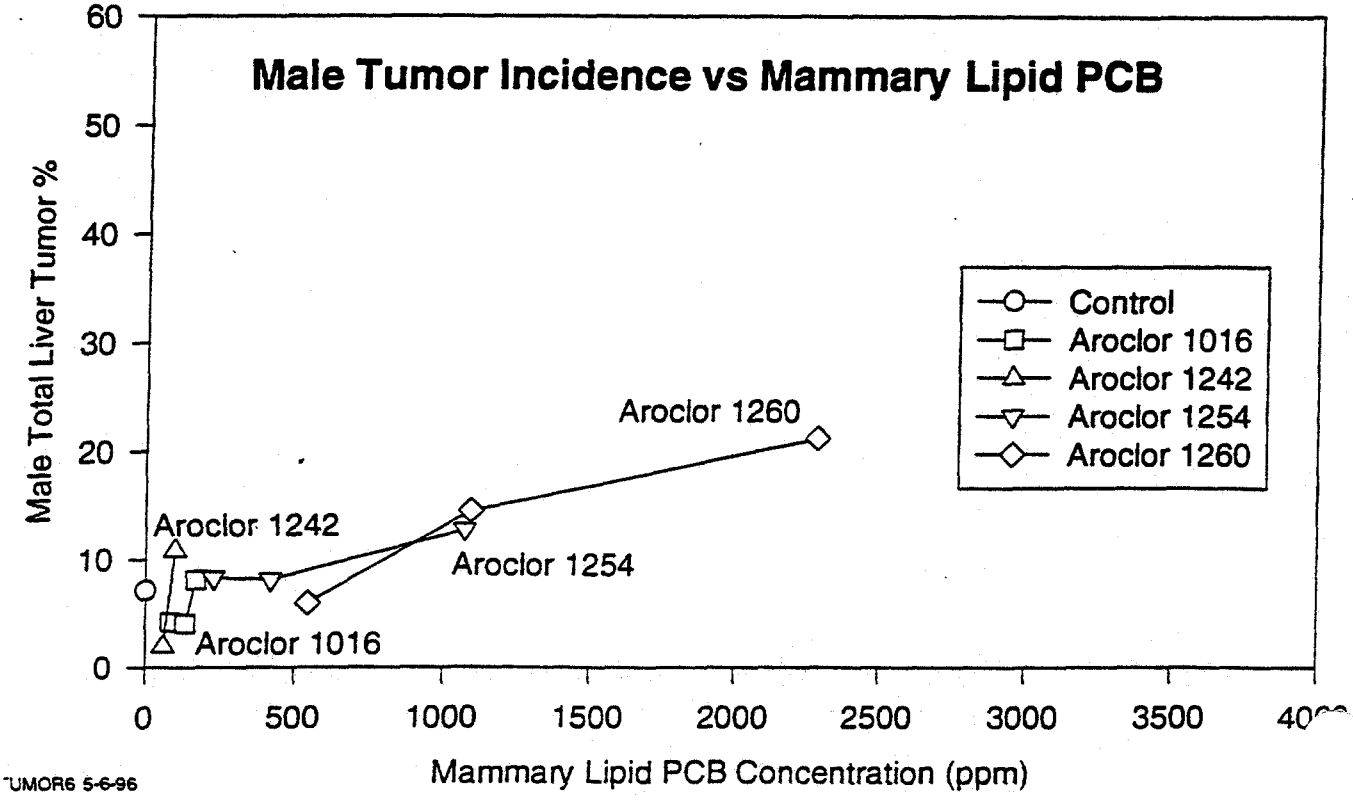
Aroclor No.	1016	1242	1254	1254	1260
% 1958-77 US production	12.88	51.76	15.73	-	10.61
Lot No.	129	01141	(laboratory	122-078	021-020
Role in study SC92012	bioassayed	bioassayed	reference)	bioassayed	bioassayed
Monochloro-BPs (wt%)	0.83	0.08	0.00	0.00	0.00
Dichloro-BPs (wt%)	17.64	14.48	0.35	0.12	0.15
Trichloro-BPs (wt%)	54.98	42.83	2.25	0.66	0.48
Tetrachloro-BPs (wt%)	25.84	33.49	20.62	19.67	2.41
Pentachloro-BPs (wt%)	0.69	6.64	43.68	45.33	11.96
Hexachloro-BPs (wt%)	0.01	1.70	29.22	31.38	39.28
Heptachloro-BPs (wt%)	0.00	0.10	3.78	2.76	36.38
Octachloro-BPs (wt%)	0.00	0.01	0.11	0.07	7.67
Nonachloro-BPs (wt%)	0.00	0.00	0.00	0.02	1.59
Decachloro-BPs (wt%)	0.00	0.00	0.00	0.00	0.07
Ortho Cl / BP	1.47	1.46	1.92	1.80	2.31
Meta + Para Cl / BP	1.55	1.81	3.11	3.29	3.98
TOTAL Cl / BP	3.02	3.27	5.03	5.10	6.29
% PCBs >CL_4/BP	0.70	8.45	76.79	79.56	96.95
RHA (70)- normal human	0.026	0.049	0.31	0.33	1.00
RHA (70) chloracne patient	0.021	0.054	0.44	0.46	1.00
ppm PCDD (total)	0.0	0.0	0.0	0.0	0.0
ppm PCDF (total)	0.05	2.2	1.0	0.13	5.5
ppm PCB 77	66.0	3340.0	380.0	918.0	31.0
ppm PCB 126	0.95	44.0	38.0	134.3	0.0
ppm PCB 169	0.0	0.0	0.6	1.52	0.0
ppm TEQ (PCDD/F)	0.002	0.1	0.07	0.01	0.08
ppm TEQ (PCB)	0.14	8.1	22.6	46.4	7.1
ppm TEQ (Total)	0.14	8.2	22.7	46.4	7.2
ppm TEQ (Total - PCB 77)	0.11	6.5	22.5	45.9	7.2

Table F-10. Relationships between Dietary, Adipose, and Liver Total PCBs and TEQs in Aroclor-dosed Sprague-Dawley Rats

				6, 12, and 18 Month Group Mean Lipid-Normalized Levels					
Group	Aroclor No.	Sex	Dietary Level (ppm)	Total Adipose PCB (ppm)	Total Liver PCB (ppm)	Ratio Liv/Adip PCB	Total Adipose TEQ (ppm)	Total Liver TEQ (ppm)	Ratio Liv/Adip TEQ
2	1016	M	50	83	69	0.83	0.011	0.005	0.423
3	1016	M	100	134	107	0.81	0.008	0.006	0.758
4	1016	M	200	167	157	0.94	0.038	0.011	0.300
						0.86 mean			0.494 mean
5	1242	M	50	60	56	0.93	0.447	5.245	11.737
6	1242	M	100	97	87	0.90	0.792	9.559	12.070
						0.92 mean			11.903 mean
7	1254	M	25	227	193	0.85	2.352	8.450	3.593
8	1254	M	50	418	370	0.88	5.134	20.677	4.028
9	1254	M	100	1076	821	0.76	11.680	38.840	3.325
						0.83 mean			3.649 mean
10	1260	M	25	545	415	0.76	1.470	1.820	1.238
11	1260	M	50	1095	773	0.71	3.133	2.122	0.677
12	1260	M	100	2284	1965	0.86	6.225	4.425	0.711
						0.78 mean			0.875 mean
2	1016	F	50	72	60	0.83	0.011	0.006	0.591
3	1016	F	100	123	95	0.78	0.022	0.057	2.583
4	1016	F	200	259	200	0.77	0.047	0.068	1.461
						0.79 mean			1.545 mean
5	1242	F	50	117	103	0.88	0.817	11.977	14.667
6	1242	F	100	207	168	0.81	1.517	20.576	13.560
						0.85 mean			14.113 mean
7	1254	F	25	454	322	0.71	5.476	21.320	3.893
8	1254	F	50	1277	834	0.65	13.811	46.852	3.392
9	1254	F	100	2589	1861	0.72	27.051	98.204	3.630
						0.69 mean			3.639 mean
10	1260	F	25	994	650	0.65	3.415	3.669	1.074
11	1260	F	50	1516	977	0.64	4.637	4.623	0.997
12	1260	F	100	3227	1836	0.57	11.360	7.594	0.669
						0.62 mean			0.913 mean

Figure F-9

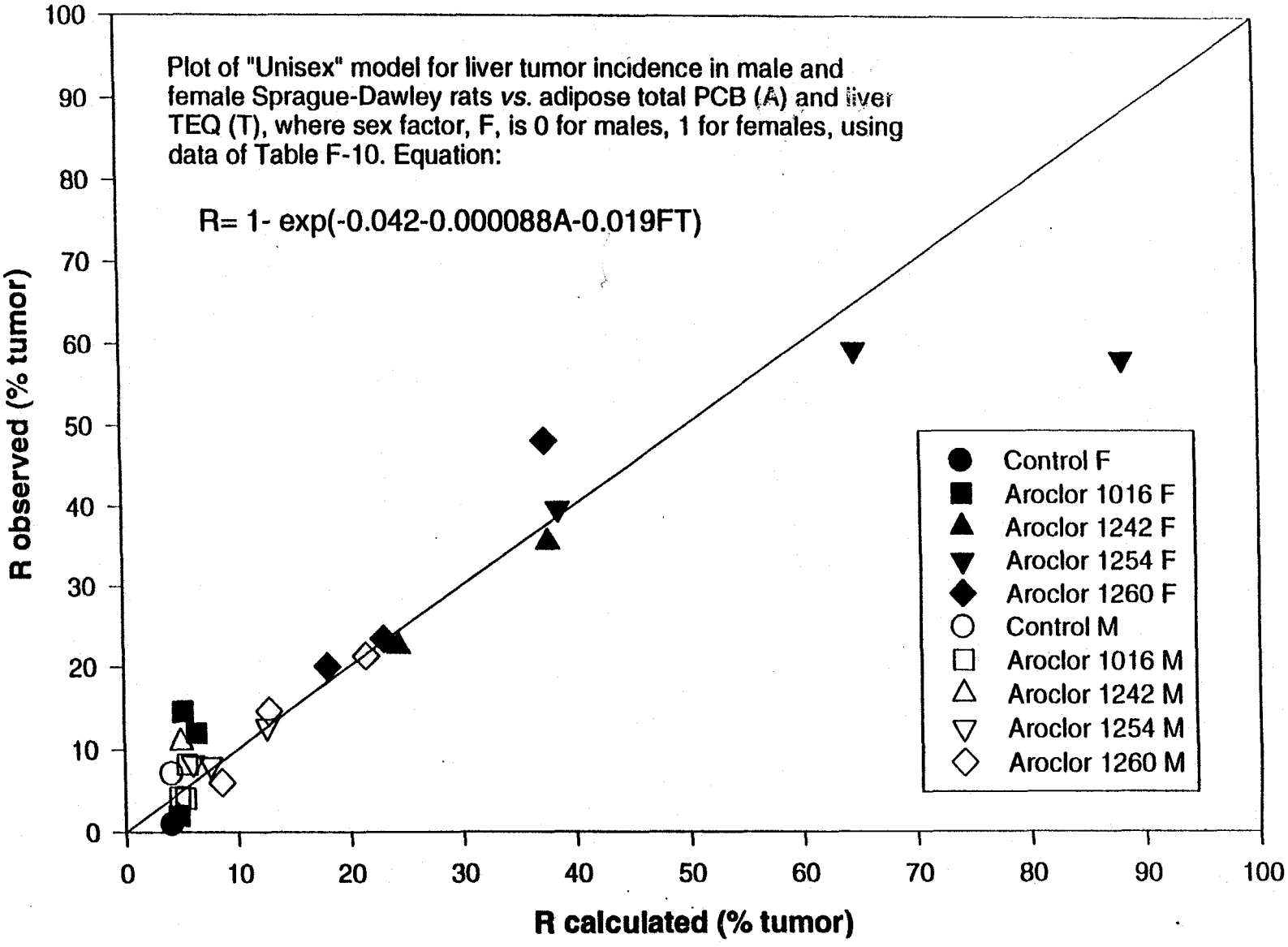




TUMOR6 5-6-96

Figure F-10

Unisex Model



AMODEL2 SPW 5-6-96

TABLE F-11. Use of Model Relating Cancer Slope Factor (CSF) to Relative Human Accumulability (RHA) and Toxic Equivalency (TEQ) to Estimate their Relative Contributions to Cancer Risk and Predict CSFs for Untested Compositions.

Model: $$CSF = a\,(\Sigma RHA) + \frac{b\,(\Sigma TEQ)}{1 + c\,(\Sigma RHA)}$$

Provisional Parameters: $a = 0.43$, $b = 7.0 \times 10^4$, $c = 2.84$

PCB Specimen	PCB accum. (ΣRHA)	ΣTEQ (ppm)	CSF Contrib. by PCB accum.	CSF Contrib. by TEQ accum.	Calcd. Total CSF	Prelim. Report. CSF(a)
Aro 1016, lot 129	0.021	0.11	0.01	0.01	0.02 (b)	0.09
Aro 1242, lot 01141	0.054	6.5	0.02	0.39	0.42	0.42 (c)
Aro 1254, lot 122-078	0.46	45.9	0.20	1.39	1.59	1.6 (c)
Aro 1254, reference	0.44	22.5	0.19	0.70	0.89	—
Aro 1260, lot 021-020	1.00	7.2	0.43	0.13	0.56	0.56 (c)

(a) EPA 1996 estimates of upper-bound limits on CSF in female Sprague-Dawley rats bioassayed in Battelle Study No. SC92012, adjusted for difference between actual and presumed food intake rates.

(b) The original data on Aroclor 1016 Tumorigenicity in female S-D rats falls in the noise range (see Figure F-10) and is incoherent. The CSF calculated from the model is consistent with the tumor yields observed in the 50 ppm female dose group, (and with those of the males in all dose groups) but not with those of the females in the higher dose groups.

(c) Values used in calculating provisional parameters a, b, and c.

Peter deFur

Review of EPA's proposed IRIS listing -- "PCB's : Cancer dose- response assessment and application to environmental mixtures." May 8, 1996, Peter L. deFur, Ph.D.

General Comments:

The report and the pages that will replace pages 5-6 of the existing IRIS listing are a good summary of PCB toxicity (briefly) and the dose-response characteristics of PCB mixtures. The report adequately explains the state of knowledge, current understandings and information about PCB toxicity, especially focusing on cancer. The report and IRIS text are coherent and understandable.

Several points regarding related issues (e.g. non-cancer health effects, non-human effects) are raised in the report. EPA correctly notes that there is insufficient space in the report to deal with these in any detail, but more specific reference to further information should be provided.

EPA refers to the fact that PCB's are associated with health effects in addition to cancer. Some of these other effects may turn out to be causally related to carcinogenicity; immunosuppression is one such an effect. EPA should more explicitly identify these relationships and indicate how the effects on another system may affect carcinogenicity.

While EPA notes in several places that the exposure of nursing infants, developing fetuses, and other sensitive individuals and groups is of particular concern. One major concern in the exposure scenario is the timing of the exposure. Are there windows of sensitivity for children to carcinogens, as with developmental toxicants? Should this point be raised here, as part of a background on explaining human risk from PCB mixtures?

Responses to Specific Questions:

1) Are the studies fairly represented?

Generally, yes. There are additional data on fish contamination and sediment to water to fish transfer of PCB's, mostly in the Great Lakes. These studies deal mostly with contaminated sediments and offer good quantitative estimates of rates of transfer through trophic systems into the human exposure path through food ingestion. EPA should refer to these.

2) Is a range of cancer potencies appropriate? This range seems to be consistent with some of EPA's related efforts to quantify and characterize risks from similar carcinogens, e.g. dioxin-like compounds. EPA has noted that the general population risk is based on central trends, but a local population may have higher risks, considering exposure factors. EPA is likely correct in this conclusion, and the application will prove problematic unless additional details are provided. EPA should probably more explicitly identify those factors that cause the higher risk factors to apply. In this case, the food chain carries PCB congeners with higher potency, and the factors need to be spelled out clearly.

3) Is internal exposure critical?

Yes, internal exposure is absolutely critical, as is "background" or existing exposure. The half-life in the body is not sufficient to characterize the risk for all individuals. As noted in the report, early exposure can, and likely does cause greater accumulation than subsequent exposure. The age of the individual, the developmental status of the individual and the temporal exposure profile are all determinants. EPA needs to account for all of these. Further, the prorating of exposure (e.g. section 4.3) as a simple ratio of exposure/ 70 year life span is not consistent with a non-linear uptake and accumulation /

effect relationship that is described (as per Kimbrough).

4) Is food chain exposure the highest risk?

All the information supports this conclusion and this reviewer concurs. It is unclear why EPA does not include quantification of the food-chain multiplier. EPA can at least provide a range of multipliers based on bioaccumulation factors. The data exist in the Great Lakes database, in analyses from bioaccumulation research (Conolly, Thomann, etc.).

5) Is the indexing to exposure pathway a useful approach that is applicable to non-cancer assessments?

The indexing approach works here because the congeners separate according to pathway, the increases in pathway utilization are likely (or certainly) independent of one another, and multiple pathways are of low probability. It is not clear that other cases, other mixtures and compounds satisfy conditions appropriate for using a similar indexing. EPA should clearly delineate the criteria for using a pathway-based indexing approach. Those criteria are appropriate here as well, and EPA should be prepared to provide further explanation of the criteria.

Revisions related to new cancer guidelines:

The report and listing for PCB mixtures reflects some of the newer approach taken in the Cancer Risk Assessment Guidelines proposed in April 1996. These new guidelines depart from the previous ones, and from some past practices in incorporating cellular, molecular and genetic information into the screening and identification steps. These steps will add an entirely new level of decision-making. The new steps are based less on a quantitative dose-response relationship from classical toxicology, and more on an understanding of mechanistic processes. This departure will initiate substantial discussion over the applicability.

Dale Hattis

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May 7, 1996

Eastern Research Group, Inc. (ERG)
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Lexington, MA 02173-3198
Attn: Susan J. Brager, 617-674-7347

Dear Colleagues:

Overall I think the EPA staff should be commended on an innovative and concise analysis of the difficult subject of PCB carcinogenic risks. The proposal to recommend adjustments to the PCB cancer potency factors in the light of the differential transfer of different PCB congeners via different modes of exposure is I think a creative suggestion with considerable merit. And the conciseness of the analysis is a pleasant surprise in comparison with the substantial documents produced in the dioxin analysis. (Of course, however, some my own suggestions will be to revise and expand the analysis in ways that would make it markedly less concise.) Below are my responses to the major questions posed to us in EPA's request for pre-meeting comments:

Are the studies fairly represented? Are any studies pertinent to dose/response assessment missing?

In my search of the recent literature I did run across a couple of references to apparently interesting papers that were not cited--one co-authored by the principal author of the EPA document:

Vater ST., Velazquez SF., Cogliano VJ
TI - A case study of cancer data set combinations for PCBs.
AB - Results of several animal bioassays have demonstrated the carcinogenic potential of polychlorinated biphenyl (PCB) mixtures. Although PCBs are no longer manufactured, cancer risk assessment for PCBs remains an important issue because of continued potential human exposure from many sources. The existing cancer risk estimate for PCBs used by the U.S. EPA is based on liver tumors observed in female Sprague-Dawley rats in a

lifetime bioassay. Liver cancer has been observed in other long-term bioassays as well. In this case study, experimental designs and biological characteristics of the data from these studies were evaluated to determine whether a combination of the data sets is scientifically reasonable. A statistical analysis of the data sets based on likelihood ratio theory was used to assess the compatibility of individual data sets to a common multistage dose-response model. The results from these biological and statistical assessments suggest that at least two data sets could be combined to derive a quantitative risk estimate for PCBs. Increased confidence in the quantitative estimate would result from such combination because more data are being used to assess the dose-response relationship.
RF - REVIEW ARTICLE: 26 REFS.
SO - Regul Toxicol Pharmacol 1995 Aug;22(1):2-10

At least two studies suggest important interactive effects of different PCB congeners:
Bager Y., Hemming H., Flodstrom S., Ahlborg UG., Warngard L
TI - Interaction of 3,4,5,3',4'-pentachlorobiphenyl and 2,4,5,2',4',5'-hexachlorobiphenyl in promotion of altered hepatic foci in rats.
AB - This study was undertaken to investigate tumour promoting interactions of 2,4,5,2',4',5'-hexachlorobiphenyl (PCB 153) and 3,4,5,3',4'-pentachlorobiphenyl (PCB 126) in female Sprague-Dawley rats. Five weeks before the promotion treatment, the rats were partially hepatectomized and initiated with nitrosodiethylamine. The test substances were administered by weekly, subcutaneous injections for 20 weeks. **The results from this study suggest that treatment with a combination of these two congeners causes a more than additive effect on the formation of gamma-glutamyltranspeptidase-positive hepatic foci.** Co-exposure to PCB 126 and PCB 153 caused a dose-dependent reduction of the PCB 153-induced CYP2B1/B2-activity in these livers.
SO - Pharmacol Toxicol 1995 Aug;77(2):149-54

Harper N., Connor K., Steinberg M., Safe S
TI - Immunosuppressive activity of polychlorinated biphenyl mixtures and congeners: nonadditive (antagonistic) interactions.
AB - The dose-response inhibition of the splenic plaque-forming cell (PFC) response and serum IgM units to the antigen, trinitrophenyl-lipopolysaccharide, was determined for several polychlorinated biphenyl (PCB) mixtures and congeners in female B3C3F1 mice. The **ED50 values for Aroclor 1260-, 1254-, 1248-, and 1242-induced**

immunotoxicity varied by less than twofold from 355 to 699 mg/kg. The range of ED50 values for 2,3,7,8-tetrachlorodibenzo-p-dioxin (TCDD), 3,3',4,4'-tetrachlorobiphenyl, 3,3',4,4',5-pentaCB, 3,3',4,4',5,5'-hexaCB, 2,3,3',4,4'-pentaCB, 2,3',4,4',5-pentaCB, 2,3,3',4,4',5-hexaCB, 2,3,3',4,4',5,5'-heptaCB, 2,2',3,3',4,4',5-heptaCB, and 2,2',3,4,4',5,5'-heptaCB were 4.6 to 4.9, 134 to 245, 4.7 to 7.0, 6.9 to 11.1, 88,000 to 121,000, 122,000 to 132,000, 99,000 to 157,000, 89,000 to 129,000, 117,000 to 240,000, and 132,000 to 238,000 micrograms/kg, respectively. The immunotoxicity-derived toxic equivalency factors (TEFs) for these congeners could be calculated from the ED50 (TCDD)/ED50 (congener) ratios and the TEF values were within the range of those previously determined for other aryl hydrocarbon receptor-mediated responses. **Based on the known concentrations of these congeners in the PCB mixtures, TCDD or toxic equivalents (TEQs) in the mixture were calculated [i.e., TEQ = sigma (PCBcongener x TEF)] using the immunotoxicity-derived TEFs (plaque-forming cells/10(6) viable cells). TEQ values for Aroclors 1260, 1254, 1248, and 1242 were 16.0, 54.4, 260.4, and 197 ppm, respectively.** Based on the ED50 value for the immunosuppressive activity of TCDD (4.8 micrograms/kg), the calculated ED50 values for immune suppression by Aroclors 1260, 1254, 1248, and 1242 were 300, 88, 18, and 24 mg/kg, respectively. **The ED50 (observed)/ED50 (calculated) ratios were 1.2, 5.9, 21, and 22.0 for Aroclors 1260, 1254, 1248 and 1242, respectively.(ABSTRACT TRUNCATED AT 250 WORDS)**
SO - Fundam Appl Toxicol 1995 Aug;27(1):131-9

The second abstract above provides data that seems to call into serious question the straightforward assumption that the potencies of different PCB mixtures can be attributed to the dioxin-like AH-receptor mediated activity associated with immune suppression, at least in these short term experiments traditionally used to define TEFs. It can be seen that calculations based on individual congener concentrations predict markedly lower potency for higher-chlorinated mixtures (such as 1260) relative to the mixtures of predominantly lower-chlorinated congeners--contrary to the cancer potency findings. The question may still be open, however, because the TEF potencies should really be adjusted for the relative persistence of different congeners in the body if one hopes to predict cancer potency (see additional discussion below).

Is a range (instead of a single value) appropriate to represent the cancer potency of PCB mixtures? If so, is the exposure pathway a reasonable default indicator of which end of the range is appropriate?

I think a range is appropriate, although I think we should try to be a little more helpful to the risk analysts in the field by assembling more quantitative information on the cancer potencies estimated for different kinds of mixtures, the uncertainties in those potencies, and the specific differences between the tested commercial mixtures and mixtures likely to be delivered via different exposure routes (inhalation, drinking, dietary consumption of fish, consumption of breast milk). Helpful quantitative information is given in the document on the composition of the original commercial PCB mixtures subjected to chronic bioassay testing. How does that compare exactly with what is observed in air, water, fish, human breast milk, human serum and human fat?*

In order to explore the feasibility of more quantitative guidance for inferring the likely cancer potency of the field samples, I reviewed the data and model fits provided in Appendix A. My goal for purposes of summarizing the potency data was to extract a central estimate of the linear term in dose response modeling for use in comparisons among the various bioassays of different congener mixtures.

Unfortunately, I found that in several cases, the multistage modeling was done in unusual ways that I believe should be changed before the document is published--resulting in inconsistencies in the estimation of the linear term I wished to use in analysis. Particularly, in several cases where the study consisted of only two data points (e.g., observations at a single feeding level vs. a control) the program was allowed to "fit" an "overdetermined" multistage model including three parameters (a parameter, usually called q_0, representing the "background" tumor incidence in the absence of exposure; a linear-dose coefficient, conventionally called q_1; and a coefficient for a dose^2 term, conventionally called q_2). The problem with such a procedure is that only two parameters are generally necessary to fit two data points perfectly. With three parameters, an infinite number of perfect fits is possible with different combinations of values for q_1 and q_2. In practice, the fitting program seems to have allocated the excess tumors over background equally

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* In addition to the potential use of this information for gauging appropriate cancer potencies for field samples, comparative information on specific congener concentrations in these four places could provide the basis for a rough pharmacokinetic analysis of persistence and relevant internal exposure in humans (fish and breast milk are principal media of exposure, human fat is the principal repository for storage, and human serum may be the most proximate index of the biologically effective internal exposure).

between the linear and dose^2 terms, yielding estimates of the linear term that are just half as large as they would be if there were no q_2 term.

Looking further, I found that I was almost as unhappy with the multistage model fits done in several other cases. For example Appendix Table A-3 reflects a model choice to fit both linear and dose^6 terms to a data set that is marginal by any standard, but certainly provides no reasonable basis for an inference of an extraordinarily steep upward-turning nonlinearity. Dose^6 terms are used without any linear term to fit the similarly marginal data in Tables A-15 and A-16, and a dose^6 term is included in combination with a linear term in Table A-17.

For reasons that I think are well articulated in the document, some finite linear term is likely to be actually present in the real dose response relationships. In fitting these same data myself, I found that there was no case in which more than a linear term is required to adequately describe the dose response information from a statistical standpoint. Table 1 compares the linear terms and ED10's for simple spreadsheet optimized fits* to a one-stage model for the liver tumor data to the similar values given in the document. (For consistency in modeling, I have excluded the few data sets for non-liver tumors. Also to avoid introducing a statistical bias into the data I have done fits to obtain "potency estimates" even for some data sets where the tumor response would not be judged to statistically significant by ordinary criteria if it were standing by itself without support from other information.)

Figure 1 shows a straightforward exploratory plot of the potencies calculated in this way vs the % chlorine in the various PCB mixtures, calculated from the composition data given in the document. There is considerable scatter in the results of the different bioassays, but overall it seems reasonable to infer that both % chlorine and gender affect the apparent cancer potency findings. Figure 2 shows the results of a simple multiple regression analysis (with gender treated as a "dummy" variable--males assigned 0 and females assigned 1). Both parameters are statistically significant at $P < .05$.

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* For this purpose I used an adaptation of the version of the multistage model published by Haas for Excel spreadsheets-- Haas, C. N. "Dose Response Analysis Using Spreadsheets" Risk Analysis 14:1097-1100 (1994). I would be happy to provide a disk version of this on request.

Table 1

Comparison of the EPA Draft Potency Estimates With Simple One-Stage Fits for Different Data Sets

		EPA Draft Fits		My One-Stage Fits		
Study	Appendix Table	ED10 (mg/kg-day)	Slope (1/mg/kg-day)	ED10 (mg/kg-day)	Slope (q1) (1/mg/kg-day)	P for fit
Kimbrough 1260 females	A-1	0.18	0.5	0.10	1.0	not meaningful
NCI 1254 males	A-3	not given for liver only	0.089	1.05	0.10	0.87
NCI 1254 females	A-6	1.25	0.084	1.15	0.092	0.50
Schaeffer A 30, males	A-7	1.8	0.03	2.1	0.050	not meaningful
Schaeffer A 60, males	A-8	0.11	0.9	0.058	1.8	not meaningful
Norback 1260 males	A-9	1.1	0.06	1.03	0.10	not meaningful
Norback 1260 females	A-10	0.11	0.8	0.06	1.7	not meaningful
GE 1016 females	A-11	1.8	0.06	1.78	0.059	0.15
GE 1242 females	A-12	0.3	0.4	0.30	0.35	0.89
GE 1254 females	A-13	0.07	1.4	0.09	1.2	0.12
GE 1260 females	A-14	0.22	0.5	0.18	0.57	0.39
GE 1016 males	A-15	3.4	0.004	45	0.002	0.55
GE 1242 males	A-16	1.5	0.01	7.2	0.015	0.07
GE 1254 males	A-17	1.5	0.02	2.5	0.042	0.93
GE 1260 males	A-18	0.97	0.05	0.89	0.12	0.60

Figure 1

Simple Regression Plots of Apparent Liver Tumor Potency vs % Chlorine for PCB Rat Bioassays By Gender

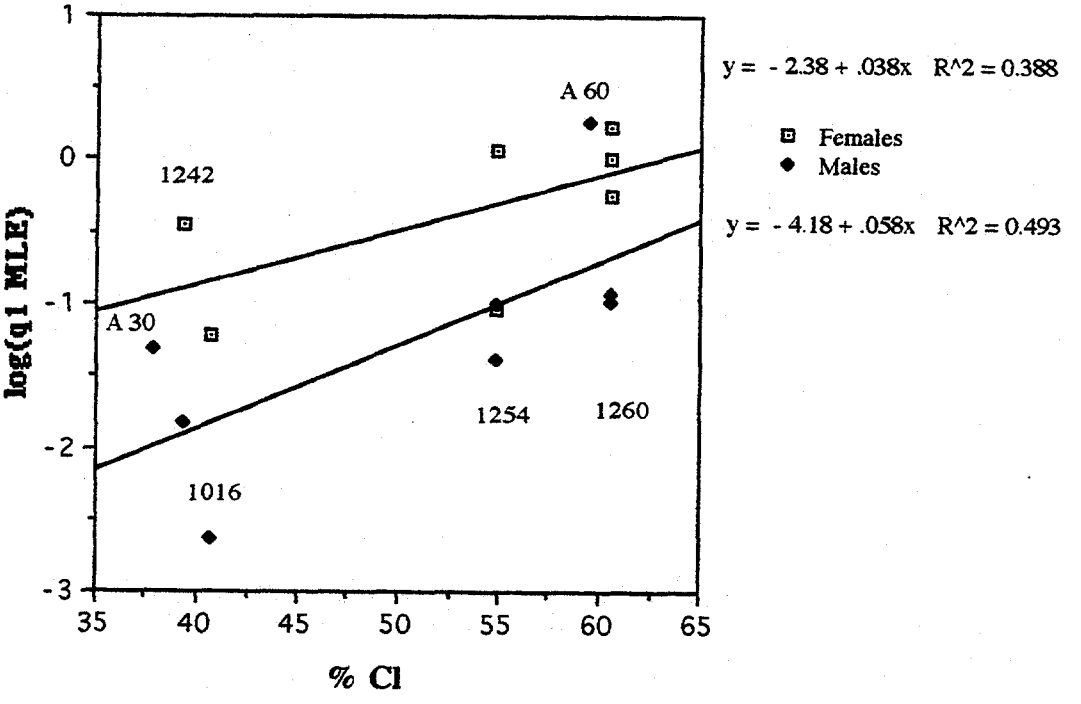

Table 2

Simple Unweighted Multiple Regression Analysis of the Effects of Gender and % Chlorine in PCB Mixtures on Log(q1) for Rat Liver Tumors

Dependent Variable Log(q1 MLE)
Count 15
Adjusted R^2 .538
F 9.14
P (regression) 0.0039

Independent Variable	Coefficient	Std. Error	Std. Coeff.	t-Value	P
Intercept	-3.748				
%Cl	.049	.016	.567	3.10	.0091
Gender	.745	.289	.471	2.58	.0243

It must be stressed that this is a crude analysis. Minimally, a better analysis should weight the various points inversely with the log variance in the estimation of the q1's. However, I think that an analysis of this sort could provide less arbitrary guidance in adjusting estimated cancer potencies for environmental mixtures in ways that depend on the bioassay data, but do not make the objectionable assumption that environmental mixtures are the same in composition or potency to the original manufactured mixtures.

Is it important that exposure assessments include internal exposure that persists after external exposure stops? If so, is half-life in the body a reasonable way to do this given the information currently available to risk assessors?

I would prefer to do the entire analysis of potencies not in terms of external exposure but in terms of lifetime-averaged internal concentrations. (It is likely that the greater persistence of the higher chlorinated congeners is responsible for much of the increased apparent potency of the PCB mixtures with greater amounts of chlorine.) Failing that, I would like to use the half life data that exists in ways that are consistent with that basic notion. The current proposal is a step in that direction, but I am not sure it is as good as we can do. This needs further thought. If potencies are expressed in terms of internal

concentration, then the body weight$^{3/4}$ interspecies translation of "dose" can probably be dispensed with.

Is the assessment correct in identifying food chain exposures as highest risk? Is it sufficient to say "risks may be higher than those estimated in this assessment" or should the risk estimates be multiplied by an explicit food-chain adjustment factor?

I think food chain exposures are likely to be of highest risk, for the reasons stated in the document. However, as indicated in my response to the second question, I would try to suggest some explicit adjustment factor(s) for the overtaxed risk assessors "in the trenches," depending on the composition of the PCBs being transferred by each exposure route, and some analysis such as the one I have given based on the based mechanistic understanding of PCB actions that we can muster.

Is this assessment's approach of a range indexed by exposure pathway likely to be useful for incorporating new information? Should it be considered for non-cancer assessments?

Yes, this approach (made explicit in numerical form as suggested above) should be adapted for use for different non-cancer effects, depending on available information on the relative importance of different mechanisms of action (and therefore different congener classes) for each effect.

Best Regards,

Dale Hattis

Dale Hattis, Ph. D.
Research Associate Professor

Kim Hooper

Comments on EPA Draft Document
"PCBs: Cancer Dose-Response Assessment and Application to Environmental Mixtures"

Kim Hooper
Hazardous Materials Laboratory
Science, Pollution Prevention, and Technology Program
California Environmental Protection Agency

May 6, 1996

I Summary

This draft document proposes some significant advances over earlier risk assessments for exposure to environmental PCB mixtures. Earlier assessments either assumed a single cancer potency for all PCB congeners, or environmental residues were assumed to be similar to the commercial mixtures from which they were derived. Here it is acknowledged that different congeners have different mechanisms of action and potencies, and that source mixtures and environmental or tissue residues differ in composition due to differential partitioning of congeners, and/or sensitivity to biological or chemical degradation. Although more data are required to fully address these problems, a reasonable and workable interim approach is proposed.

II Discussion

Overall, an excellent document. Concise, judicious, thought-provoking. A pleasure to read. The two major points are both welcome and useful innovations: 1) human exposures are to environmentally transformed, and not commercial, PCB mixtures; 2) persistent PCB congeners remain in tissues long after external exposures have ceased, where they provide extended internal exposures and sustained biological activities. These two innovations make our assessments more closely mirror what is going on in the real world, and they should be used generically in future risk assessments of other chemicals, where appropriate.

We could enhance the value of the document to risk assessors at the state and local levels by giving several more examples of practical risk assessment calculations. These practical examples would illustrate the proper application of the concepts advanced here.

The major concepts are good ones, and well expressed:

1) that general population exposures to PCBs are not to commercial mixtures, but to environmentally transformed mixtures (1/9, 2/20-26, 6/3-5, 22/32, 35/1-2);

2) that environmental transformations tend to selectively remove the less toxic PCB congeners, leading to mixtures enriched with the more toxic congeners, and making environmental mixtures more toxic than the parent commercial mixtures (3/10-12, 26-28, 28/18);

3) that environmental mixtures may undergo further transformation in humans and become further enriched with bioaccumulating congeners (15/20-28-16/1-10), some of which are tumor promotors whose activities persist after dosing ceases (e.g. 3,4,3',4'-tetra) (5/5, 13 Table 2-2, 15/20-25, 23/5-20);

4) that internal human exposures to these persistent congeners may extend years after external exposures to environmental mixtures have ceased (15/25-16/10, 31/3-10, 32/2, 32/11-12);

5) that PCB mixtures are likely to employ a variety of mechanisms in cancer causation: some congeners have dioxin-like activity, some are promotors, and some are metabolized to what may be mutagenic metabolites (18, 23-25, 33/15);

6) that these mechanisms are shared with other compounds in the exposure "background", making the cancer risk of PCB mixtures at low doses additive to background risk and, therefore, linear with dose (19, 34-36); and

7) that the major exposure pathway is via food ingestion, where selected groups are at high risk (game-fish consumers and breast-fed infants) (3/26-4/1, 37/3-10).

III Questions

1) The range (100-fold) of differences in congener compositions created by environmental transformations is much larger than the range of cancer potency estimates (a factor of 10) for different PCB commercial mixtures (tested in different sexes/strain of rats). We need potency factors to cover the range of environmental mixtures. Is bias introduced when we represent the potencies of environmental mixtures by the narrow range of potencies derived from cancer tests on commercial mixtures?

2) The new potency estimates in this document are lower than earlier estimates published by USEPA for certain studies. For example, the previous estimate of potency derived from liver tumor data in female rats (Morback and Weltman, 1985) is 7.7/(mg/kg-d). The revised potency based upon the same experiment is 2.3/(mg/kg-d). The reduction in potency has three components (% reduction: see footnote A):

a) revised liver tumor incidence data (Moore et al., 1994) (34% reduction);

b) revised effective lifetime dose, calculated using t1/2 for PCBs in rodents (31%);

c) revised interspecies scaling factor (for extrapolating from rodents to humans) of three-fourths power of body weight rather than the two-thirds power used earlier (36%).
Not having reviewed the PWG report, I can't comment on "a)". "b)" seems reasonable. What's the basis for "c)"?

3) The new GE data (interim results from PCB bioassay by the General Electric Co.) covers Aroclor mixtures containing a wider range of chlorine content than previous studies.

a) With regard to the significance of the new PCB bioassay results, is the statement, "The significant results for Aroclor 1260 males indicate a nondioxin-like mode of action is also operating" (43/12-13) justified by the GE data presented?

b) Table 5-1 indicates gender difference (females more responsive than males) in liver cancer effects from all PCB commercial mixtures. Why? A hormonal effect? Differences in liver metabolism? In sulfotransferase activities? Shouldn't we note this pronounced gender difference in the risk assessment?

4) With regard to the statement, "...the ED01 method...is mostly independent of choice of model, and is statistically stable..." (21/6-8), don't the EDs have some flutter? Very different values for ED01 can be obtained from data sets (with many dose groups) that show an increased effect only at the highest dose, depending upon whether the multistage or one-hit model is used. Furthermore, a change in the diagnosis of one individual can have a significant effect on the linear term in the multistage model and, thereby, on ED01. Relevant conclusions from Crump and Allen (benchmark dose methodology) could be cited here. (The example given in the document avoids the problem, because the ED01/linear and the linearized multi-stage (LMS) approaches give similar low-dose slope factors.)

5) In describing high-risk subgroups, the document includes breast-fed infants and game-fish consumers. It makes no mention of potential susceptible populations, e.g. infirm, persons on immunosuppressive drugs, persons with compromised immune systems. Some dioxin-like PCB congeners are known to affect the immune system. Although present data are inadequate to incorporate/factor these effects into a cancer risk assessment, shouldn't such sub-populations and immune system effects be noted?

6) Is much known about the effect of timing of exposures to PCBs? If so, insert into the discussion of risks to infants breast-fed on PCB-contaminated milk.

IV Policy Issues

The recommendations below seem useful, and have implications beyond the risk assessment of PCB exposures.

1) The proposed matches of cancer potency values with policy purposes (36/12-14) seem reasonable:

a) public health (sensitive groups) = upper bound;

b) aggregate risk (population) = central estimate; and

c) risk ranking = central estimate.

2) Two factors are identified which require changes from earlier EPA default approaches:

a) Persistence of some PCB congeners in human tissues years after exposure has ceased. To reflect the continued internal exposure, it is proposed to extend the effective duration of exposure commensurate with the anticipated half-lives of congeners in the mixture.

b) The distribution of congeners in PCB environmental mixtures (involved in environmental or food chain exposures) differs from that in PCB commercial mixtures, and may be enriched with congeners of greater persistence and higher toxicity. To reflect this, it is proposed to use higher potency values with some environmental exposures (e.g. food sources, dust inhalation, and sediment ingestion) and lower potency values in others (air, water, and dermal exposures).

V Suggestions

1) The principles described in this document (see Discussion, 1-7 above) should serve as a basis for a generic approach to risk assessment of environmentally important mixtures as well as other environmentally important compounds of a similar nature (e.g. chlorinated pesticides, napthalenes, dioxins, dibenzofurans and paraffins).

2) To be most useful, the document needs to go farther. Whereas the updated Cancer Policy document outlines scientific policy "principles" to be used in risk assessment, this draft PCB risk assessment should implement these principles. To be useful at the state and local level, where the "rubber meets the road," more examples of risk assessment calculations are needed which apply these principles to specific cases. For example:

Calculate aggregate risk to 16-year old girl whose family lives in Upstate New York in a village on the Hudson River near a GE facility and has regularly consumed PCB-contaminated game-fish: exposures to the girl include PCB-contaminated breast milk, game-fish, playing in river sediment and riverbank soil, and swimming in the river.

Use the congener-specific PCB data for residue levels in breast milk, game-fish, riverbank sediments and river water. Describe how congener-specific data is applied to the risk assessment (e.g. values for persistent congeners, dioxin-like congeners, and tumor-promoting congeners (44/27-21)). Describe factors in selecting high or low potency values for PCB contaminants/congeners in water, dust, sediment, fish, and breast milk to calculate risks from oral, dermal and inhalation exposures. Discuss selection of t1/2 values for different congeners. Discuss effects of timing of exposures.

VI Research Needs

For risk assessment purposes, we need congener-specific data on PCB residues in fish, shellfish, sediments, and breast milk in the US. In particular, few data are available on congener-specific PCB-residue levels in breast milk samples taken from US populations (see footnote B). Such data would fill three important gaps: describe bioaccumulating congeners in different US populations; suggest congeners that may be persistent in US populations; and describe congeners that infants receive from breast milk (high exposures). (Additionally, such data may identify congeners that play a role in breast cancer.) The exposure of infants to PCBs via breast milk illustrates the importance of correctly defining the risk estimates for a sub-population that has already been identified as highly exposed and potentially highly sensitive.

VII Details

1) Put small diagram of PCB structure in footnote 1 that indicates structure of dioxin-like congeners.

2) 22/26: Typo (ED10, not ED01).

3) Table 3-1: Didn't all these studies use rats? If so, indicate. In any case, give species.

Footnotes

A. Potency Estimates

The new potency estimates for PCBs in this document are lower than estimates previously published by U.S. EPA. For example, the previous estimate of potency derived from data on liver tumors in female rats reported by Norback and Weltman is 7.7/(mg/kg-d). The revised potency based on the same experiment is 2.3/(mg/kg-d). The reduction in potency has three components:

1. Use of revised tumor incidence data from re-evaluation of histopathology (Moore *et al.*). The lower incidence of liver tumors in the reevaluation reduces the potency estimate by 34%. I have not reviewed the pathology working group (PWG) report that reexamined histological specimens from PCB.

2. Use of the dose rate for the first 16 months as the dose rate for the experiment. Originally, EPA calculated a time-weighted average (TWA) of the doses used in the study (5 mg/kg-d for the

first 16 months; 2.5 mg/kg-d for the next 8 months; no exposure until the experiment ends at 29 months). This TWA dose was 3.45 mg/kg-d. This change in the dose rate calculation reduced the potency estimate by 31%.

Because the half life of PCBs in rodent tissues is at least several months, the new method of estimating the effective lifetime dose is clearly an improvement, and should be recommended to risk managers as the method of choice for assessing the effective duration of exposures of human tissues to persistent PCB mixtures.

3. Use of inter-species scaling factor (for extrapolation from rodents to humans) based on the three-fourths power of body weight rather on the two-thirds power results in a 36% decrease in the potency estimate (for a 0.35 kg rat). This approach was recommended in the USEPA's draft revision of the Cancer Risk Assessment Guidelines. Scientific data seem presently insufficient to decide this issue. Is this a policy decision to achieve consistency with the US FDA?

B. Research Needs: Congener-specific PCB data

The document correctly states that PCBs in environmental and biological samples differ from industrial mixtures (Aroclors, Clophen, Kanechlor) because of selective weathering and metabolism of certain low-chlorine congeners and selective bioaccumulation of certain high-chlorine congeners. It is this altered profile of PCB congeners that humans are exposed to, mainly via the diet, and this is what should be measured to assess exposure. Unfortunately, most assessments of human and ecological health still rely on measurements of Aroclors, even though reliable analytical techniques have been developed for congener-specific analysis. In addition to reporting "Aroclors", there is a tendency to report "total PCBs". The latter can be quite deceptive since the "total" is just the sum of congeners the chemist chose, or was able, to measure, and it varies from report to report depending on the methodology, instrumentation, etc. For example, the EPA proposed PCB "Mega-Rule" did not include congener-specific analytical methods. In brief, there is an urgent need to generate congener-specific PCB data to fill the data gap, and to conduct reliable exposure and risk assessments. Two examples where information on specific PCB congeners is important are exposures of nursing infants and recent findings in breast cancer studies.

1. Nursing infants

Infants may receive extremely high doses of PCBs. Yet, few data exist on PCB congeners in human milk (Newsome, 1995; Borlakoglu, 1989; Tuinstra, 1994; Larsen, 1994; Noren, 1991; Georgii S, 1995; Liem, 1995; Schecter, Becher, 1995; She, 1996). Most of these reports had few, not necessarily representative samples. No data exist for the USA. Systematic studies are needed to characterize PCB profiles in human populations and assess risks.

2. Breast cancer

Recently, the anti-estrogenic potential of a number of PCDD/PCDF and PCB congeners has been shown. In general, their order of potency parallels their binding affinities for the Ah receptor (Krishnan, 1993). On the other hand, commercial PCBs (Aroclors) did not exhibit analogous anti-estrogenic behavior (Krishnan, 1993). Therefore, unless these specific congeners are measured and controlled for in the analysis, exposures may be misclassified and associations missed.

Higher levels of Aroclors were found in serum (Dewailly, 1994) or adipose (Falk, 1992; Dewailly, 1994) of breast cancer cases than in controls. However, no such association was found in other studies (Krieger, 1994; Wolf, 1993; Mussalo-Rauhama, 1990). One reason for the disparate results reported in recent studies (Falck, Krieger, Wolff) may be explained by the fact that these

studies focused on Aroclors or "total" PCBs rather than on the individual congeners, a shortcoming acknowledged by Wolff (Wolff, 1994), an investigator on all three studies. One team of State of California and Stanford researchers are currently examining body burdens of PCDD/PCDF and PCB congeners in women with and without breast cancer (Petreas, 1994). Clearly, more studies are needed in this area.

References to Footnote B

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Wolff MS, Toniolo PG, Lee EW, Rivera M, Dubin N. Blood levels of organochlorine residues and risk of breast cancer. JNCI 85:648-652, 1993.

Wolff MS, P Toniolo. Response (to letter). *JNCI* 86: 1095-96, 1994

Marty Kanarek

Premeeting Comments: May 7, 1996

Peer Review Workshop on PCBs: Cancer Dose-Response Assessment and Application to Environmental Mixtures

- *Are the studies fairly represented? Are any studies pertinent to dose/response assessment missing?*

It appears that the existing studies are fairly represented. A review article should be added: Kimbrough R.D. (1995) Polychlorinated biphenyls (PCBs) and human health: an update. Crit. Rev.Toxicol. 25(2):133-163. This is a recent review article by a well known expert on PCBs. Dr. Kimbrough emphasizes the high incidence of 60% chlorination in rat tumor causation as compared to rats fed mixtures with 54 or 42% chlorination. In reviewing the human data on workers and the general population, she finds "...no clear and convincing evidence that PCB exposures were casually associated with adverse health effects ...this included cancer for a wide range of body burdens and exposures for serum PCB concentrations > 1000ppb.. and adipose PCB levels >400 ppm..." p.133 (abstract).

- *Is a range (instead of a single value) appropriate to represent the cancer potency of PCB mixtures? If so, is the exposure pathway a reasonable default indicator of which end of the range is appropriate?*

A range is more appropriate than a single value. The exposure pathway is a reasonable default indicator for which end of the range is appropriate.

- *Is it important that exposure assessments include internal exposure that persists after external exposure stops? If so, is half-life in the body a reasonable way to do this given the information currently available to risk assessors?*

If possible, it would be desirable to use internal dose to the critical organ. Given information currently available, half-life in the body is at least some information on internal dosing.

- *Is the assessment correct in identifying food chain exposure as highest risk? Is it sufficient to say "risks may be higher than those estimated in this assessment" or should the risk estimates be multiplied by an explicit food-chain adjustment factor?*

Food chain exposure is by far the highest for PCB mixtures. Since the difference between food chain and the other exposures is orders of magnitude in difference,, a food-adjustment factor may be wise.

• *Is this assessment's approach of a range indexed by exposure pathway likely to be useful for incorporating new information? Should it be considered for non-cancer assessments?*

This assessment's approach of a range indexed by exposure pathway seems more realistic than approaches of the past. Certainly it should be considered for non-cancer assessments.

• *Compared with past assessment, this assessment reflects some changes in guidelines and approach. Any reaction to the new features in this assessment?*

The IRIS summary has more information on the human carcinogenicity data, which, even though it is not directly used in a cancer dose-response assessment, gives some further weight of evidence that the dose response derived from animal studies may be reasonable. Specifically, the Brown (1987) and Bertazzi (1987) studies show some small few excess numbers of human liver cancers, which are too few to be of much statistical significance but given the animal results are significant biologically. The new assessment approach should not forget human data as an important component of the total evidence to be weighed in a risk assessment. If the epidemiological data is too sparse to contribute to a quantitative dose-response because of inadequate cohort size or follow-up period, or some other reason, it still can contribute qualitatively, as it does in the PCB mixtures case.

Specific Line Comments

p. vi, line 11: "Although.... PCBs also *may* have significant..." Add [may]

p. vi., line 14: "Toxic effects have been observed *in animal studies* from acute..." Add [in animal studies]

p. 1, line 30: "Coplanar molecules have dioxin-like properties..." Does "properties" mean activity? Reference to "dioxin-like" here decreases the usefulness of this document by itself without reference to a dioxin document.

.p. 16, lines 11-13 and 23-27. The point here about the half-life of a mixture is interesting, but if the chemistry is not sufficiently advanced to measure each congener, a mixture value is certainly better than none, and if the mixture is realistic as to common human exposure than its half-life is very informative.

p. 18, lines 17-19, 22-23: Reference to "dioxin-like" without explanation decreases the usefulness of this PCB mixtures document by itself. Probably the solution

is to give an explanation of "dioxin-like" which the document does latter on.

p. 22, line 32: ....Environmental processess *can* have profound effects that can... Add [can]

p. 27, line 28: Chlorine content appears to be *generally* associated with cancer ... Add [generally]

p. 51, line 12 add Kimbrough R.D. (1995) Polychlorinated biphenyls (PCBs) and human health: an update. Crit. Rev.Toxicol. 25(2):133-163.

Nancy Kim

Are the studies fairly represented? Are any studies pertinent to dose/response assessment missing?

The studies are fairly represented given the stated purpose and focus of the limited risk assessment. I am unaware of any studies pertinent to the dose/response assessment that are missing. The new studies being carried out by General Electric are extremely important and will probably substantially lessen the importance and reliance on the earlier studies.

Is a range (instead of a single value) appropriate to represent the cancer potency of PCB mixtures? If so, is the exposure pathway a reasonable default indicator of which end of the range is appropriate?

Whether or not a range or single value is used and what range should be used depends on the question being asked. For example, if the question is "What is the range of cancer potency values for PCB mixtures?," then the range of all potency values should be given. However, in making a decision as to whether or not a range accurately reflects the potency for PCB mixtures, then the question should be answered by determining whether or not the potency values for the different mixtures are significantly different from each other. For example, if one looks at the new data coming out of the General Electric studies, one can make the decision that the potencies for Aroclor 1242, 1254 and 1260 are basically the same and, given our limited knowledge of the degree of experimental variations in cancer bioassays, that the same range of potencies should be given for all three mixtures. Statistical tests or other evaluations could be carried out to answer the question as to whether or not the range of potencies for these three different mixtures substantially overlap. However, the Aroclor 1016 reslts from the new studies indicate that the mixture appears to be less potent than the other mixtures and the potency for Aroclor 1016 should be given separately. In any case, only one potency factor should be selected for any particular PCB mixture. If the data show no discernable difference in potency between different mixtures, then a single number should be picked for all those mixtures.

I am not convinced that the exposure pathway is a reasonable default indicator of which end of the range is appropriate. That decision should be made based on the analytical data for the particular medium being evaluated. The potency of the PCB should be used

that most closely represents the PCB mixture in the environmental media. One of the sample calculations illustrates a problem with this approach. For drinking water exposure, the calculation used the potency factor for Aroclor 1254 because it was at the low end of the range; however, Aroclor 1254 is one of the more highly chlorinated mixtures and, therefore, less water soluble. I agree, in many cases, that the PCB congeners in the exposure pathway are likely to reflect chemical/physical properties and, therefore, may tend to be at one end of the range for PCBs.

Is it important that exposure assessments include internal exposure that persists after external exposure stops? If so, is half-life in the body a reasonable way to do this given the information currently available to risk assessors?

The suggestion put forth by the U.S. Environmental Protection Agency (EPA) to use half-life in the body as a way to account for internal exposure after external exposure stops is an interesting one. However, before that procedure is used, I recommend that EPA consider the feasibility of validating that approach with data that are already in existence. For example, the Kimbrough/Linder 1974 study exposed mice to Aroclor 1254 for 11 months or for 6 months followed by 5 months without exposure. These data might provide an opportunity to determine whether or not the results of that study agree with the procedure suggested. I am reluctant to recommend that this procedure be adopted without additional justification (1) using data on the underlying biological mechanisms and from long- and short-term studies that would qualitatively support the process and (2) as to why the suggested quantitative approach may be more accurate than the existing procedures. Moreover, the procedure should probably not be used in some situations (e.g. exposure time is short compared to halflife).

Is the assessment correct in identifying food chain exposure as highest risk? Is it sufficient to say "risks may be higher than those estimated in this assessment" or should the risk estimates be multiplied by an explicit food-chain adjustment factor?

Based on my knowledge of exposure to PCBs, food chain exposure is the highest exposure route and, therefore, provides the greatest risks from PCBs. However, I don't believe the data in the paper provide sufficient information to justify the statement that food chain exposure is the highest risk. In many of the recent EPA risk assessments, statements are made that the risks may be actually lower or higher than those estimated.

I favor qualifying estimates of cancer risk in both directions. Given the data for this risk assessment, without further justification, I would not multiply the risk estimate by an explicit food chain adjustment factor.

Is this assessment's approach of a range indexed by exposure pathway likely to be useful for incorporating new information? Should it be considered for non-cancer assessments?

My response to this follows my response to the second issue. A range and a single cancer potency should be given for individual Aroclors (assuming that individual Aroclors have sufficiently different cancer potencies). The preferred value for a given risk assessment should be dictated by the mixture in the environmental sample. In terms of using the same procedure for non-cancer assessments, the same recommendation would hold that the toxicity be evaluated using data on the Aroclor that the environmental sample most closely mimics. However, I would not recommend this procedure for non-cancer assessments at this point.

Individual Comments

The approach that EPA has taken in this new document is interesting and the agency is to be commended for attempting to derive general procedures to use in many different situations. This approach may well be scientifically sound, however, the document needs to provide sufficient information to justify some of the general statements that are made in it. If additional information were provided to justify those statements, the document would be of more value.

Page 3: The statement, *"In general bioaccumulative PCBs appeared to be more toxic than commercial PCBs on a weight-to-weight basis..."* EPA may want to reevaluate the statement and add some qualifications to it. It is a fairly broad-reaching statement and may not be justified by the studies that have been done.

Page 5: The summary statement is made that, *"Overall, the animal studies have been considered to provide sufficient evidence of carcinogenicity..."* EPA may want to consider qualifying that statement. Without using the new General Electric data, the data

may not be sufficient to provide evidence of carcinogenicity for Aroclor 1016.

Page 22: EPA makes that statement that *"The two data sets showing lowest potency may reflect the low sensitivity of male rats; consequently, this assessment focuses on the remaining five data sets."* Additional justification is needed to throw out the two studies with the lowest potency. There are other studies that also used male rats. Given this approach, EPA should throw out all the male rat data.

Page 29: Given my responses to the second and fifth issue to consider, I believe that the statement, *"Consequently, the low end of these ranges is appropriate for drinking water ingestion or vapor inhalation, where environmental processes are likely to decrease risk, while the high end is appropriate for food chain exposure or ingestion of contaminated sediment or soil, where environmental processes are likely to increase risk"* should be revised to reflect that the potency should be chosen based on the PCB mixture in the environmental sample.

Page 29: The bottom of the page a statement is made, *"Overall, use of the low end of the potency ranges...for dermal exposure appears appropriate in light of the substantial but incomplete absorption through the skin."* The reasoning should be reevaluated for this statement. A better approach may be to reduce the amount that is absorbed through the skin which seems more scientifically precise than just deciding to use the low end of the potency range.

Page 30: Some of the other statements on the top of page 30 should be reconsidered. It isn't clear to me how *"Rapid absorption, however, suggests potency by inhalation is comparable to potency by ingestion"*. In fact, I would suggest that EPA relook at that whole paragraph and consider whether or not the reasoning can be justified further.

Page 31: The statement *"This would allay concern for short-term exposure but increase concern as exposure duration increases"*. I would feel more comfortable if the statement were qualified by adding something like, *"assuming the same exposure levels."*

Page 33: The paragraph mentions *"cooking food contaminated with PCBs can cause*

formation of chlorinated dibenzofurans." Then it goes on to give the concentrations of the dibenzofurans in Aroclors. This may be useful information but it does not provide information on how much dibenzofurans may be formed by cooking.

Page 35: The statement is made *"In contrast, the environmental processes of partitioning, transformation, and bioaccumulation have been extensively studied, and exposure pathway is a reliable indicator of whether toxicity has been decreased or increased by environmental processes."* Again, I don't believe that this document has provided information to prove this statement, although it may be true some cases.

Page 36: There is a paragraph beginning on line 10 that starts, *"Depending on specific application.....comparing a central estimate with its upper bound indicates whether the central estimate is precise enough to support credible risk estimates."* I recommend that EPA review this paragraph with the idea of making some changes. For example, the statement *"There is no scientific basis for expecting less sensitive groups or an average of exposed groups to be representative or protective of a heterogeneous human population."* While I understand the point that is trying to be made, one could interpret the sentence to say that there is no scientific basis for using studies on workers exposed to a chemical to set guidelines or standards for humans. The last sentence of this paragraph should also be reviewed.

Page 43: The statement is made that *"the significant results for Aroclor 1260 males indicate a nondioxin-like mode of action is also operating."* It isn't clear to me that the document has provided sufficient information to support that statement.

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Loren Koller

PEER REVIEW WORKSHOP

PCB'S CANCER DOSE-RESPONSE ASSESSMENT

AND APPLICATION TO ENVIRONMENTAL MIXTURES

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- The research studies included in the document are adequately represented and referenced. Although heptacellular carcinomas are the primary neoplasms associated with PCB exposure, other types of neoplasia also occur; e.g., leukemia, lymphoma, gastric cancer. Although the "GE 1995" study in progress reports the incidence of liver tumors, do other types also occur in these animals?
- Using a range to represent cancer potency appears to be appropriate for PCB mixtures. The congeners vary from no to low to high potency in inducing neoplasia. Exposure pathways appear to be important in this process with exposure occurring via ingestion, inhalation and/or dermally. Although multiple routes of exposure can add to the total body burden, the actual route of exposure has little bearing on the type of cancer produced.
- PCB's partition to fat in the body. Although the half-life of stored PCB's can be up to several years, the internal dose (exposure) is of questionable biological relevance. PCB's can become mobilized from lipid storage but the total body burden is self-limiting; e.g., the longer the half-life, the smaller the internal dose released from fat and mobilized via blood to other cells, tissues and organs. Starving could provoke mobilization of large amounts of PCB's which would deplete the overall body burden much more rapidly thus markedly reducing the biological half-life. The extremely low dose of mobilized PCB (internal exposure) over several years would appear to be an insignificant contribution to adverse health effects.
- It is important to recognize that PCB mixtures do partition, transform, and bioaccumulate in the environment. PCB's that are highly lipophilic do bioaccumulate in the food chain. This route of exposure would constitute a higher risk of exposure than other routes of exposure.

The assessment approach of a range indexed by exposure pathways would appear to have merit for PCB mixtures. The "secondary exposures" frequently add very little risk compared to the principle exposure pathway. Separating risk by exposure pathways allow comparisons between pathways. A comparison between this method of risk assessment and combined exposures in relation to cancer lifetime average daily dose would be of interest in estimating risk.

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May 13, 1996

Ms Susan Brager
ERG
110 Hartwell Avenue
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Dear Ms. Brager:

As you know, it will not be possible for me to participate in the peer review workshop on PCBs dealing with cancer dose response due to a previous international commitment. As encouraged by you and Bill Farland I would like to offer several brief comments for the Workshops consideration.

1. What is the appropriate data base for use in PCB risk assessment?

While I strongly support the position that all data should be used in a weight of evidence process it would appear to me that there is a new data base that dominates. I refer to the recently completed Battelle studies in rats where four specific Aroclors were studied in parallel adhering to a similar protocol. It provides the only meaningful data on the long term effects of Aroclor 1016 and 1242. Further, it does not suffer the limitations of group size that compromised the interpretation of the NCI study with Aroclor 1254. The studies with Aroclor 1260 provide the first opportunity to assess response to more than one dose and carry the additional benefit of temporal relevance to the other Aroclor studies. The Battelle studies have the additional benefit of being conducted in a manner that was fully compliant with EPA Good Laboratory Practices. Finally, there is the additional, powerful advantage of congener specific analytical chemistry data which permits determination of dose to target site. In my opinion, the previous studies should now be relegated to a secondary utility, including the "re-read data" that I published a little over a year ago. There is one gold standard - the Battelle study - from which to derive any type of quantitative estimates of potency; the other data sets are not remotely of the same quality.

2. How to derive quantitative estimates of potency?

Were I able to participate at the meeting I would have argued strongly for consideration of a BMD approach with an appropriate MOS for each Aroclor. There is much evidence to suggest that the non coplanar congeners promote rather than induce carcinogenesis. The data from the Battelle study indicate that Aroclor 1254 is not leading to the same type of

response seen with other Aroclors. Current belief would tend to account for this by invoking a duality of response due to the relative concentrations of coplanar congeners in this mixture in addition to the non coplanar promotion.

Should one not opt for BMD plus MOS it is assumed one will favor the estimation of an ED10 with a line then drawn to 0. Given the unsettled nature of the discussion regarding use of the lower 95% confidence bound for the line drawing it would be my hope that the central estimate and the lower bound data be presented.

The total value of the Battelle study has not been realized given that there is a plethora of analytical data that has yet to be published. Such data may provide the opportunity to propose different model considerations for potency estimation of the Aroclor 1254 data in particular. It would be my hope that the Workshop formally recognize this and encourage the consideration of such data should it become available

What potency values should be used for field data?

I am sure this topic will provoke lively discussion. Rather than offer my own "potency recipe" let me make one recommendation. In those instances where there is credible field data encourage its use in a risk assessment in lieu of a default assumption. Encourage the comparison of their analytical results with Aroclor congener profiles to determine the most relevant Aroclor from which to estimate cancer potency. In other words, foster the use of data and the application of common sense. Where appropriate analytical data are not available at a site the default approach would be recommended.

Sincerely,

John A. Moore

Christopher Portier

Comments by Dr. C. Portier, NCEA-W-059; Risk Assessment of PCB's.

In general, I find this document to contain a practical and useful approach for the assessment of risks from exposure to PCB's. In general, the following areas are methodology deviances by the EPA that I feel are very good ideas:

1. Focus on the ED01 as a basis for extrapolation. This dose, being slightly outside the range of doses with a statistically detectable response, is able to reflect some of the nonlinearity of the response (this is obvious in Table 3-1) which would be lost by using a higher response dose.

2. Correction for environmental decay and modification of the congeners and isomers. This approach appropriately corrects for modifications of the original mixtures and will lead to more scientifically defendable decisions.

3. Use of variable potencies depending upon current make-up of the mixture. The use of hard estimates TEF's has met with considerable scientific debate which may, in the long run, delay the implementation of reasonable procedures for controlling exposures. The use of ranges related to knowledge of potency allows for greater scientific certainty in the estimated risks without locking these estimates into overly specific potency values.

4. Correction for persistence. The addition of expected duration of persistence to the estimated time of exposure is useful, practical and corrects for an oft ignored aspect of chemical exposures.

There are several areas that are mentioned as being of importance in the document which, I feel, are still inadequately addressed in the current approach and which may be part of our focus in discussions of this document. Included in this category are:

1. The impact of age on the risk of cancer from a given exposure. It would be nice if the methods developed for evaluating risks from a given exposure could encompass differences in age-related potencies of the compounds. Perhaps three broad categories of young, adult and mature would be beneficial. There appears to be sufficient data to consider this option.

2. The importance of current exposure as well as accumulated exposures in the potency calculations. The method prescribed is mostly driven by the current exposure and does not directly account for accumulation.

All told, Dr. Cogliano should be commended for provided the US EPA with a very thoughtful and comprehensive analysis method for risks from exposure to chemicals in a congeneric class.

C. Portier

Paul Price

Peer Review Workshop on PCBs: Cancer Dose-Response Assessment and Application to Environmental Mixtures

Issues Considered During Review

Question 1

Are the studies fairly represented? Are any studies pertinent to dose response assessment missing?

The review and presentation of historical PCB bioassay data appears to fairly represent the findings of the studies. However, the basis for the derivation of the range of values is unclear and raises a number of questions.

- Should all studies be given equal weight? Certain studies used larger numbers of animals and may be viewed as being more important. If studies are given different weights how can this be reflected in the guidance?

- Why are the results in males excluded from the distribution for certain studies (Norback and Weltman, 1985) but not for others (Schaeffer et al., 1984)? EPA indicated that the two data sets showing the lowest potency may reflect the low sensitivity of male rats and therefore did not consider them in the proposed range of slope factors. Two other data sets for male rats; however, were included in the slope factor range. This removal of low potency data may not be appropriate when developing a range of potencies. In the case of the data from the Schaeffer et al. (1984), male rats were considered an appropriate model of carcinogenesis for Clophen A60, but not for Clophen A30. If male rats are considered inappropriate for characterizing PCB carcinogenesis, then all male rat data sets should be removed from consideration. Alternatively, all bioassays should be included in development of a slope factor range.

- Slope factors presented for the NCI study (1978) represent the sum of slopes calculated for liver and gastric tumors and male leukemia and lymphoma. Justification should be provided for the combination of distinct tumor types in dose response assessment. Data indicating that tumors are of the same histomorphogenic origin are needed in order to support data combination (see: McConnell, E.E., Solleveld, H.A., Swenberg, J.A., and

Boorman, G.A. (1986). Guidelines for combining neoplasms for evaluation of rodent carcinogenesis studies. J. Natl. Cancer Inst. 76: 283-289). Also, it is important to note that the incidence of leukemia/lymphomas in males were not considered by the study authors to be clearly related to Aroclor 1254, since Fisher Exact tests of the data were not significant. Therefore, use of these data in combination with liver and gastric tumors does not appear to be justified.

It should be noted that many of these problems are reduced if the range is based on the Battelle study.

Question 2

Is a range appropriate to represent cancer potency of PCB mixtures? If so, is the exposure pathway a reasonable default indicator of which end of the range is appropriate?

As discussed by EPA and many experts in the field quantity estimated of carcinogenic potency is subject to numerous types of uncertainty. Acknowledging this uncertainty in the risk management process by discussing carcinogenic potencies as a range rather than a single point estimate is a significant improvement in the dose response characterization portion of the risk assessment policy and should be strongly supported. However, it is not clear what types of uncertainty the agency intended to incorporate into its proposed range of potencies. There are numerous sources of uncertainty in developing quantitative estimates of carcinogenic potency (see for example Evans recent paper on formaldehyde in Risk Analysis). It appears that EPA is only attempting to describe the uncertainty that arises from separate measurements of carcinogenicity at different times in different species and genders and with three different types of PCB mixtures. EPA is not including the uncertainty that arises from the use of a non-threshold dose response model or the uncertainty in the application of rodent bioassay results to humans. In addition, the agency is not including the uncertainty that results from the limited number of animals available in the bioassays. The proposed regulations would benefit from a transparent discussion as to which sources of uncertainty are supposedly addressed by the proposed range and what sources of uncertainty have been excluded. (This is not to say that the proposed range is inappropriate for guidance; merely to indicate that the agency needs to be explicit in their intentions.)

A significant question in establishment of a range of potencies is the role that the new PCB bioassay will play. The current document suggests that there is no need to adjust the range since the results of the PCB bioassay general fall in the same range as the historical studies. While it is understandable that EPA should initially purpose a range based upon historical studies (as the results of the Battelle bioassay were not available as final numbers at the time the EPA draft was written), it is inappropriate for the agency to continue to rely upon the historical studies once the Battelle results have been finalized. The Battelle study offers a number of important advantages over the historical studies including:

- The availability of results for multiple dose rates

- Because the studies were performed at the one time on the same species and strain of rat using the same protocol the results of the Battelle study provide an unique opportunity to identify how carcinogenic potency varies across PCB mixtures.

- The new bioassay data are superior to a number of the historical studies in the number of animals used in the controls and dose groups and in the absence of problems that occurred in certain historical studies (for example variable dose rates in the Norback and Weltman studies).

Because of these benefits EPA should base the range of PCB carcinogenic potency factors solely upon the results of this bioassay and use the historical studies to provide qualtitative information on how the carcinogenic effects of PCBs may vary by gender and species.

The use of an exposure pathway as the basis for selecting a value from the range of potencies is a reasonable default approach when little information on PCB mixture composition is available. It should be acknowledged, however, that in many instances other factors such as the composition of PCBs initially released, and biotransformation may play an important role in determining the composition of the PCB mixture and therefore should be considered in selecting a single potency value on a site-specific basis.

EPA should also acknowledge potential problems with the theoretical framework which relates persistence and bioaccumulation of PCB congeners with postulated mechanisms of carcinogenicity. For example chlorine content may be directly related to the potential to bioaccumulate, however not all highly chlorinated congeners are considered potent promoters

of carcinogenicity. Structural determinants may be equally as important as chlorine content in determining carcinogenicity.

Several inconsistencies are present in the report regarding chlorine content, congener composition, and carcinogenesis, for example:

- It was concluded that congener toxicity cannot be characterized by chlorine content alone (p. 24).

- Chlorine content appears to be associated with cancer risk (p. 27).

- The similar slopes for Aroclor 1242, 1254, 1260 females in the Battelle study, together with the overlapping composition of these mixtures, casts doubt on chlorine content of the original mixture being a useful indicator of cancer potency in this range of chlorine content.

- The document states that bioaccumulation increases with increasing chlorine content and that bioaccumulated PCBs appear to be more toxic (on a weight basis) than commercial PCBs (pg. 3). However, EPA supports this statement by citing two studies, both of which were conducted with mink, a species whose relevance to the evaluation of human health effects is questionable.

EPA should revise the document to reflect a consistent position. As part of this revision EPA should consider developing a new section that clearly presents the available data and the agency's position on this issue.

Question 3

Is it important that exposure assessments include internal exposure that persists after external exposure stops? If so, is half-life in the body a reasonable way to do this given the information currently available to risk assessors?

My understanding of EPA's concern on this issue is as follows. EPA's goal is to determine the risk associated with human exposure to PCBs. Since the late 1980s EPA has been using a less than lifetime exposure duration (30 years). Human exposure to PCBs may persist

beyond the 30 year exposure duration due to the accumulated body burden of PCBs. Under the earlier policy of lifetime exposure this issue did not arise since EPA assumed that individuals were exposed for their entire life span.

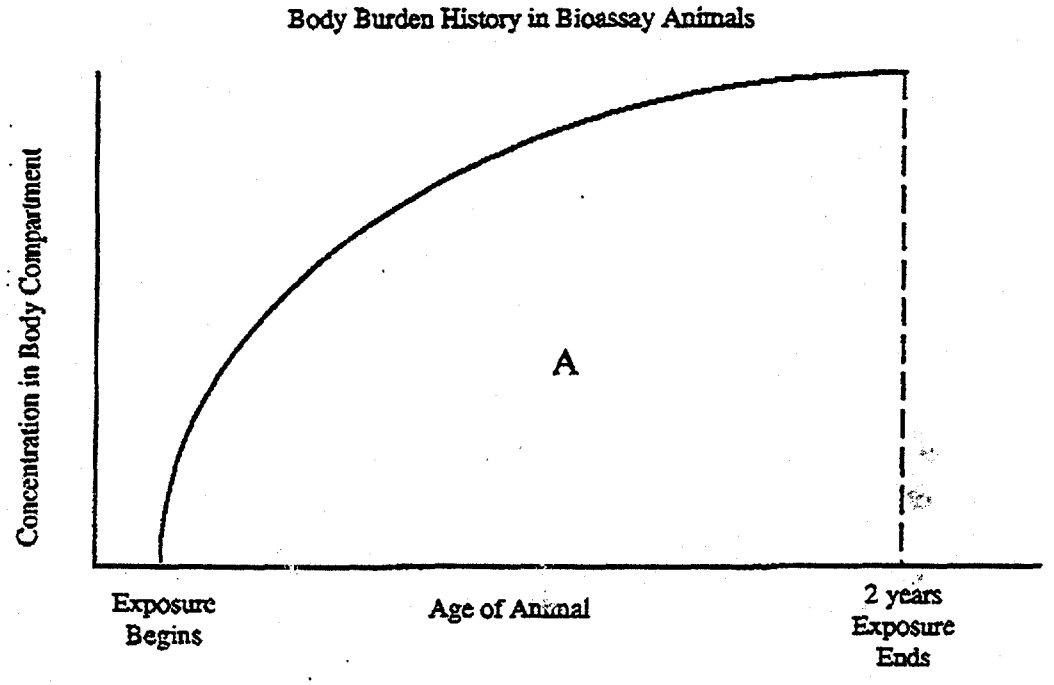
In general, animal bioassays reflect responses that are the result of continuous lifetime exposure. The tumorigenic response therefore does not correspond to the human exposure scenario. The net effect is a potential underestimation of risk associated with exposure to PCBs in humans.

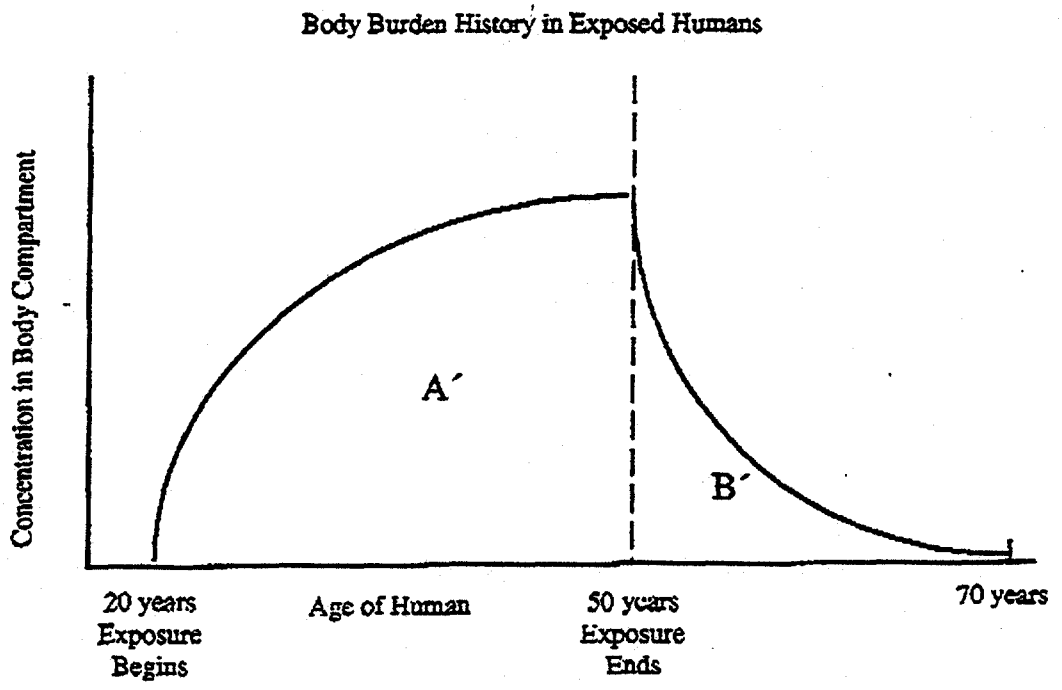
This issue is presented graphically in Figure 1. The response in the animals is viewed as a function of Area A; in humans the risk is a function of Areas A' and B'.

The goal for EPA is to find a way to account for the risk from Areas A' and B' based on the bioassay data which only reflects Area A. EPA's proposed approach (see Figure 2) is to assume an internal exposure duration of 9 years for an exposure duration of 30 years. The 9 year internal exposure duration adjustment is based on a model of the half-life of PCB body burdens in workers.

EPA should be commended for investigating issues that go beyond the use of administered doses, as the basis for evaluation of risk. However, because the mechanism by which PCB causes carcinogenicity in rats is unclear, it is premature to conclude that an adjustment for internal dose is necessary or appropriate. While a number of researchers have suggested that area under the curve (AUC) measurements are useful for predicting adverse affects associated with persistent chemicals, such a hypothesis requires the assumption that the components of PCB mixtures that drive the risk are those that persist in the body. Unlike TCDD, PCBs are mixtures of compounds which greatly vary in their rates of metabolism and excretion. Because it is possible that the carcinogenic effects in the rodents may be driven by readily metabolized congeners rather than the congeners that persist in rodents, it is not clear that the area under the body burden curve is a better metric for response than administered dose. Because of this uncertainty, it is inappropriate at this time to quantitatively include an adjustment for internal dose. If the Agency is aware of any data that indicates that risk is driven by body burden, then an adjustment for internal dose may be appropriate.

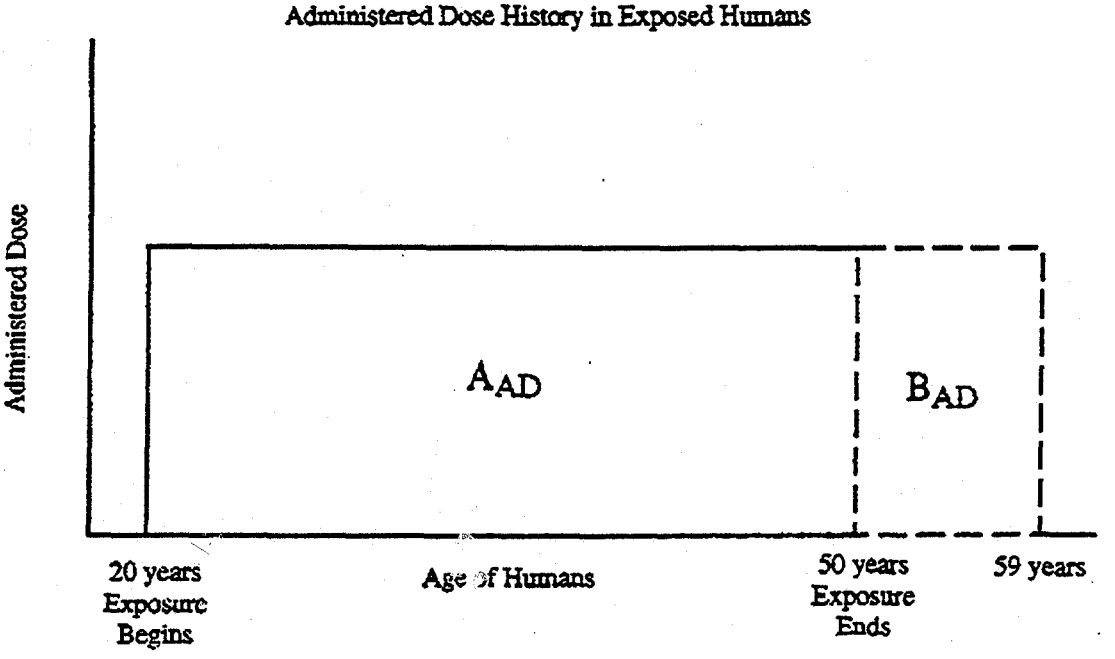
Figure 1. Body Burdens Over Time

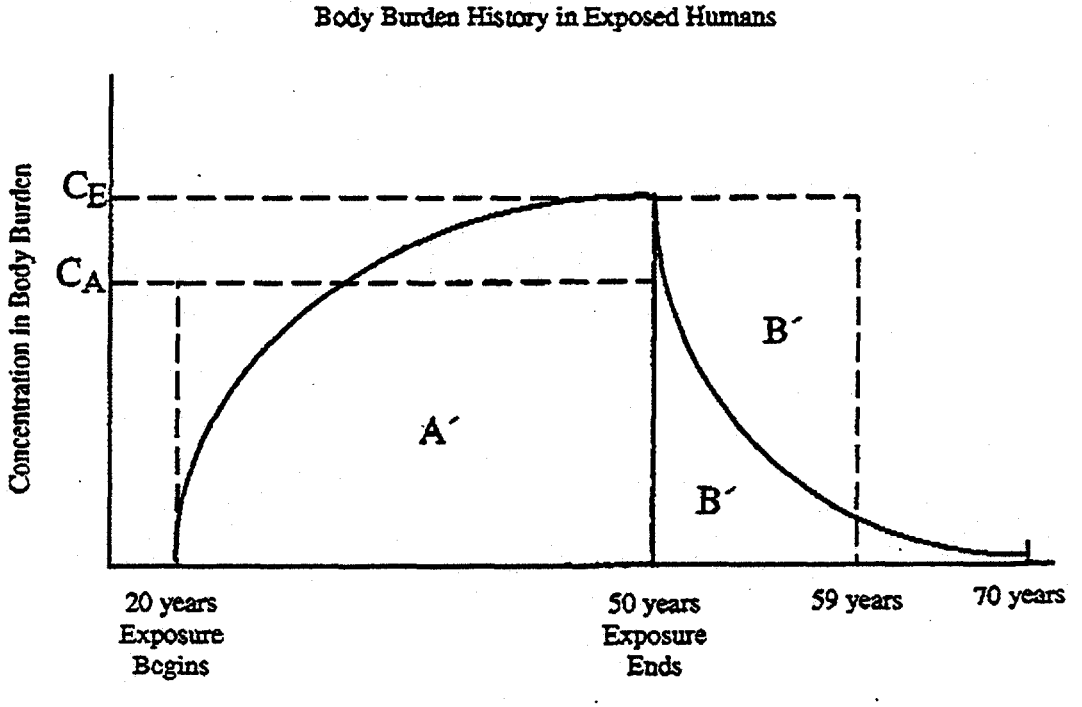




Comments/Figure 1

Figure 2. Proposed Modification of Exposure





A second issue with the proposed regulation is the proposed mechanism for dealing with internal dose. Assuming the agency concluded that an adjustment for internal dose is necessary, it is not clear that the recommended approach is appropriate. The following is a series of problems with EPA's proposed approach for dealing with internal dose.

1. The approach assumes that the extended exposure duration will be predictive of the human body burden vs. time curve. This can be graphically illustrated in Figure 2, where increasing the area A_{AD} to $A_{AD} + B_{AD}$ (or 30 to 30 + 9) is assumed to produce the equivalent increase from A' to A' + B'. In order for this association to be predictive the following equation must be true:

$$(A_{AD} + B_{AD})/A_{AD} = (A' + B')/A'$$

EPA has suggested that the Area of B' can be estimated as:

$$B' = 9 \text{ years} \times C_E$$

C_E is the body burden (concentration) in the human at the end of the period of exposure. Substituting in this value of B' and the values of 30 and 9 for A_{AD} and B_{AD}, the value of Area A' can be calculated to be 30 years x C_E. Since C_E is the concentration that the individual reaches only at the end of the exposure, the Area of A' cannot be equal to 30 years x C_E. The actual area will be given by:

$$A' = C_A \times 30 \text{ years},$$

where C_A is the average body burden over the duration of exposure (see Figure 2). By inspection C_A will always be less than C_E, therefore, the assumption that the extended exposure duration is a prediction of body burden vs. time curve is not correct.

2. The EPA approach leads to illogical conclusions when applied to other exposure durations.

EPA's analysis on the area under the body burden curve following exposure (B') assumes that a 30 year exposure B' is approximately equal to 9 years x C_E. However,

this is also true for any duration of exposure, even durations as short as 1 year or 1 day. Thus the methodology would seem to imply that the same additional 9 years should be added to any exposure of any length. This is implausible since the internal dose should be affected by the body burden which is related to total administered dose.

This problem occurs because C_E is strongly dependent on duration of exposure. However, EPA has not provided any guidance on how to adjust the value of B_{AD} to reflect the changes in C_E that occur as a function of exposure duration. EPA needs to provide an approach that corrects for internal dose that is a function of exposure duration.

3. The approach of extending exposure duration to account for body burden, assumes that humans are exposed at a fairly constant dose rate over a 30 year period. However, in many instances long-term temporal trends exist for human exposure to PCBs. These trends have a major influence on the body burden at the end of the exposure duration (internal dose). For example, if a high level of exposure occurs early in the exposure period (0-10 years), followed by minimal exposure later (20-30 years), the body burden at the end of exposure will be small and the exposure period probably does not require an extension. However, if exposure occurs predominantly late in the period of exposure duration, an extension of 9 years may not be adequate to account for internal dose. Thus, a simplistic addition of a single number cannot account for internal doses even when the duration of exposure is specified to be 30 years.

4. The proposed approach implies that the ratio of average body burden (during the period of exposure) to average administered dose rate is the same in humans exposed for 30 years, and rats exposed over their lifetimes. This may not be the case and additional supporting evidence for this position should be provided.

5. The Area B' is determined by the number of years that an individual lives following the cessation of exposure. The basis for the equation for B' assumes that the humans remain alive for an infinite period of time following exposure cessation. However, the equation is reasonably correct if the period of time is 4 or more times greater than the half-life of PCBs (6.5 years). If the 30 year exposure occurs later in an individual's life and ends at 50 or 60 the individual may not live long enough for the equation to be valid. In these cases the value of 9 years may be too large.

6. The adjustment suggested by EPA in the draft document makes the implicit assumption that exposures to PCBs received late in life are equally potent at producing tumors to those exposures received early in life. The agency has not provided evidence that this is necessarily the case.

7. The issue of internal dose is in reality a problem of modeling human exposure patterns in the dose rates administered in the bioassay. The bioassay dosing regime is a model of a constant lifetime dose in humans. In reality, human exposures are highly variable on both a short and moderate timescale and may be subject to long-term trends. In addition, exposures may be episodic and occur over various portions of individual's lives. Exposures may also be in the form of bolus doses rather than low-level continuous exposures. The internal dose issue highlights the fact that the average body burden that occurs from a thirty-year human exposure within a 70 year life span may have a different relationship to the administered dose than the body burden history that occurs in the bioassay.

   Historically, the agency has not quantitatively adjusted risk estimates for dose related factors such as bolus dosing, episodic dosing, dosing during different periods of the individuals' life span. It is not clear why the agency is considering an adjustment for this effect at this point in time when substantial uncertainty surrounds any type of adjustment short of developing a pharmacokinetic and pharmacodynamic model of PCB carcinogenicity in humans.

Summary

Because the mechanism by which mixtures of PCBs exert carcinogenicity in rodents is unclear, it appears to be premature to consider a modification of the risk assessment process based upon the internal dose issue. However, if the agency does decide to issue such guidance the proposed approach is not sufficient to describe the complex nature of this issue.

Question 4

Is the assessment correct in identifying food chain exposure as highest risk? Is it sufficient to say that "risks may be higher than those estimated in this assessment" or should the risks estimates be multiplied by an explicit food-chain adjustment factor?

As discussed earlier in my comments, consideration of bioaccumulation that may occur in exposure pathways is a plausible basis for determining a value from the range of carcinogenic potencies. The reason for this is that, in general, carcinogenicity across Aroclor mixtures is greater in higher chlorinated Aroclors as compared to lower Aroclors. (The recent Battelle study shows a higher cancer slope factor for Aroclor 1254 than Aroclor 1260.) Since bioaccumulation will result in the preferential enrichment of higher chlorinated congeners, the assumption that foodchain exposures present the highest risk of all the exposure pathways appears to be plausible.

However, it is not clear why the Agency wishes to claim "risks may be higher than those estimated in this assessment". If the Agency has evidence to suggest that PCB carcinogenicity continued to increase with higher chlorination beyond Aroclors 1254 or 1260 and if the Agency had information to support the theory that bioaccumulation will result in increased enrichment of even higher homologue groups, such a claim may be justified. However, the current data does not support either contention. First the results of the Battelle study suggest that carcinogenic potency for Aroclor 1260 is lower than the carcinogenic potency of 1254. Therefore, it is not clear that exposure to PCB mixtures which as a result of preferential bioaccumulation resemble 1260 rather than 1254, would result in increased risk. As second reason why carcinogenicity may not increase with increased chlorination is that the total amount of coplanar PCBs decreases with chlorination for mixtures with chlorination greater than Aroclor 1254. Thus bioaccumulation may result in a reduction of risks that due to dioxin like effects.

It is not clear that bioaccumulation through the foodchain would necessarily result in PCB mixtures resembling Aroclor 1260. Bioaccumulation is a complex process, and is a function of both the lipophilic nature of the compound and its bioavailability. As compounds increase in chlorination their lipophilicity increases and their solubility decreases this increase their preferential accumulation in the foodchain, but decreases their bioavailability. As a result, it is not clear that mixture of PCBs with moderate levels of chlorination such as Aroclor 1242

would necessarily be modified into compositions that resemble Aroclor 1260 or higher chlorinated PCB mixtures. Based upon these two factors, it is more appropriate to conclude that the range of carcinogenicity reported in the historical and new studies has a high likelihood of characterizing the upper bound carcinogenic potency of all PCB mixtures.

The use of a explicit foodchain adjustment factor for carcinogenic potency does not appear to be warranted at this time for the reasons given above.

Question 5

Is the assessments approach of a range indexed by exposure pathway likely to be useful for incorporating new information? Should it be considered for non-cancer assessments?

This question is in fact two separate questions. In response to the first question, indexing a range based on an exposure pathway does not in-and-of-itself assist in the incorporation of new information into the process of selecting a carcinogenic potency for a PCB mixture. It is not clear what EPA intended by this question

In answer to the second question, it should be noted that the non-carcinogenic effects of PCBs may be a function of multiple mechanisms and may not be readily predicted based upon the gross composition of PCB mixtures. As a result it may be premature to consider extrapolating the approach used in the carcinogenic assessment to non-carcinogenic effects.

Additional Comments

EPA should be commended for the many advances in risk assessment presented in this draft.

As the authors of the document point out (pg. 6), the draft assessment incorporates information not typically included in dose-response assessments prepared by the Agency, such as: using a range of potency estimates, calculation of central tendency and upper-bound slope estimates and guidance on use of both, use of internal as well as external dose, discussion of biologically-based models, application of the draft revised cancer guidelines, including the use of the compromise scaling factor. We agree that this document represents

an improvement in PCB cancer risk assessment, and EPA is to be commended for its efforts in preparing this draft assessment.

Characterization of Uncertainty in PCB Potency Estimates

The agency should provide more information on the uncertainty in potency that results from limited numbers of animals in key studies. The way to consider such uncertainties is to establish a distribution of potency values that reflect the uncertainty and to incorporate these uncertainties into the estimates of the total uncertainty in a risk assessment. This could be done by:

1. Reporting the entire uncertainty distribution for the potency estimates instead of merely reporting the MLE and the upper 95 percent confidence limit. This approach was used in a recent National Academy of Science project performed by Dr. Edmund Crouch.

2. Consider weighing the studies based on the number of animals in the study.

3. Selection of a potency value from the range whould also include the uncertainty distribution for the corresponding bioassay.

Premature conclusions were made regarding the role of dioxin-like mechanisms of carcinogenicity.

EPA is inconsistent in its discussion of the role of dioxin-like toxicity in the observed carcinogenic activity of PCB mixtures. Although EPA is careful to state that PCB carcinogenesis likely arises by both dioxin-like and non-dioxin-like modes of action (p. 33: 1-3), the majority of the document presents the theme that the dioxin-like activity of PCBs is of concern to public health, especially when one considers exposures to other sources of dioxin-like equivalents. EPA has also considered sex-related differences in potency in the Battelle bioassay as evidence of dioxin-like toxicity. The document should be revised to present a single consistent position on this issue.

Larry Robertson

1. Are the studies fairly represented? Yes. Are any studies pertinent to dose/response assessment missing? There are at least two large-scale epidemiology studies in the pipeline looking at cancer mortality in PCBs-exposed individuals. One study looked at cancer mortality in over 130,000 men employed in the electric power companies in the US. In the second study, or group of studies, researchers are conducting nested-case control studies using pre-diagnostic serum collections. The association of organochlorine compounds, including PCBs, and the incidences of several different tumors including breast, prostate and hematopoietic malignancies are being tabulated. As soon as these studies are accepted in peer-reviewed journals, summaries should be incorporated in the Assessment and the conclusions considered.

2. Is a range (instead of a single value) appropriate to represent the cancer potency of PCB mixtures? It is clear from published reports and from the preliminary data from the GE study that all commercial PCB mixtures were not equally toxic. Furthermore environmental PCB mixtures differ from commercial PCB mixtures due to processes of partitioning, transformation and bioaccumulation, as outlined in the Assessment. This variability in composition implies a variability in toxicity, which is confirmed in several studies. Unfortunately knowledge of the toxicity of each PCB congener, and how that toxicity is influenced by the presence of all other congeners, as well as knowledge of the absolute composition of any PCB mixture, would be required in order to calculate the toxicity associated with a particular mixture. Since we lack this knowledge, using a range (acknowledging differences in toxicities) seems a reasonable alternative.

If so, is the exposure pathway a reasonable default indicator of which end of the range is appropriate? This seems to be a reasonable compromise at this point. Should we not, however, strive toward the goal of understanding the toxicities of PCB mixtures based on their compositional makeup and their multiple interactions?

3. Is it important that exposure assessments include internal exposure that persists after external exposure stops? It is certainly an important concept that PCBs may be retained for considerable periods of time and that they continue to exert their toxic actions long after application of the PCBs has been terminated. The persistence and recirculation of PCBs within the organism dictates that the PCBs are potentially available to interact with critical targets. It should also be noted that toxic effects may persist after a considerable portion of the PCBs have been eliminated. This later concept is demonstrated by the example of the Yusho victims whose serum PCB levels have dropped to near background, but who are continuing to suffer the severe ill effects of their exposure.

As a toxicologist, I have problems with the terms "internal exposure" and "external exposure". I find that they are potentially misleading. We are, after all, only interested in those PCBs which enter the body. Isn't exposure = (absorbed) dose X time? Is there not a better choice of terms?

Is half-life in the body a reasonable way to do this given the information currently available to risk assessors? No, see the above comment. Distinguish between persistence of the PCB congener and persistence of the toxic effect.

4. Is the assessment correct in identifying food chain exposure as highest risk? Is it sufficient to say "risks may be higher than those estimated in this assessment" or should the risk estimates be multiplied by an explicit food-chain factor? Do we not have sufficient data to attempt a quantitation?

5. Is this assessment's approach of a range indexed by exposure pathway likely to be useful for incorporating new information? Should it be considered for non-cancer assessments? For non-cancer endpoints the assumptions will be very different of course and PCB environmental mixtures of highest risk for cancer will likely not be those of highest risk for causing other changes.

Stephen Safe

Document: PCBs: Cancer Dose-Response Assessment and Application to Environmental Mixtures

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Page 4 (Section 1.2): Some of the occupationally-exposed groups were probably exposed to other chemicals used in preparation of various PCB products. This would include chlorinated benzenes and these compounds alone or in combination with commercial mixtures may contribute to the variability in the carcinogenic outcomes.

Page 4 (line 25): What are "nonsignificant increases"? Why don't you mention all of the nonsignificant decreases? The authors should be consistent and either list only significant increases (or decreases) or summarize all significant and nonsignificant effects.

Page 7-12: In discussion of various cancer studies where more than one dose was used, the document should indicate at which doses the effects were observed (this is done in the Partial Lifetime Studies in Animals section). For example, in the NCI studies (Page 7), it is stated that "Hepatocellular adenomas and carcinomas appeared increased". What does this statement mean? The results summarized on page 8 indicate that no increases were observed in males at the 25 and 50 ppm dose and, in females, no increases were observed in any of the dose groups. Therefore, the statement in the text refers to only the high dose (100 ppm) males. For this group, the descriptor" appeared" should be clarified.

Page 9 (line 22): Incidences were lower (not smaller). In the Norback male rats, the significance of the cancer incidence in the 50 and 100 ppm should be indicated separately.

Page 13 (Table 2-2): The nomenclature for the PCB congeners in the Table and text should be corrected (e.g. 2,2',4,4'-, not 2,4,2',4'-). Since PCBs probably act as tumor promoters and not initiators, you may want to point out the failure to detect DNA adducts using the ^{32}P-postlabeling assay in rodents treated with Aroclor 1254 (*Toxicology* 68, 275, 1991).

Page 14: The statement "as all ortho-substituted congeners in Table 2-2 are abundant in commercial mixtures (Schulz et al., 1989) and have been found in environmental samples" is misleading since some of the tetraCB congeners are not particularly persistent in the environment.

Page 15 and 16 (lines 25-28 and 1-13): It must have been a "real stretch" to determine Aroclor 1242 half-lives when this mixture does not persist as an entity in human or the environment. It would be more useful to quote half-lives on total PCB body burdens than on Aroclors (particular lower chlorinated mixtures). How were levels calculated for the Great Lakes fisheaters (Hovinga et al., 1992)? You address this problem at the end of the section whereas it should appear at the beginning. You may want to point out that half-lives after exposure to lower chlorinated PCB mixtures are initially low and then increase due to the preferential retention of persistent congeners.

Page 24 (line 10-12): Hydroxy-PCBs bind to transthyretin and may modulate thyroid hormone-mediated pathways exhibit estrogenic or antiestrogenic activities and are found in human samples and in wildlife tissues. However, I do not understand the evidence for their "genotoxic or carcinogenic potential".

Page 24 (lines 25-26): There are at least 2 recent papers which painstakingly show that the calculated TEQs and experimentally-derived TEQs for Aroclor 1260 are low (*Fund. Appl. Toxicol.* 20, 456, 1993; 27, 131, 1995). This is an important point which should be supported by recent primary data.

Page 30 (lines 8-11): Although PCBs induce lung tumors in mice, one wonders how important this response is in humans since there is no evidence for this effect in occupationally-exposed workers. Based on conversations with workers and personal visits to several facilities which used PCBs in the early 1970s, there may have been high exposure via inhalation.

Page 32 (lines 1-7): I question the value and meaning of these half-lives.

Page 32 or 33 (lines 10-12): Cooking food can also result in formation of potent carcinogens such as PAHs and heterocyclic amines. Which contributes more to cancer potency: cooking-induced amines/PAHs or cooking-induced PCBs/PCDFs?

Pages 34-38: This section is a useful discussion of the dose-response characterization of PCB mixtures based on animal studies. However, I think it is also important to point out that the human cancer data is derived from highly exposed workers. The human data for this end-point should play a role in risk assessment and if this is not the case, then there should be an explanation.

Page 37 (line 7): The statement "Bioaccumulated . . . commercial PCBs" is not true for all responses. The TEQs for Aroclor 1248 are probably higher than for many bioaccumulated PCB mixtures.

Susan Velazquez

- *Are the studies fairly represented? Are any studies pertinent to dose/response assessment missing?*

Yes, the key studies are well represented. I am not aware of any additional studies pertinent to dose-response assessment. As indicated in more detail in my review notes following the answers to these five main questions, some of the lesser important studies (with regard to the establishment of a dose-response relationship) should perhaps be reported in greater detail.

- *Is a range (instead of a single value) appropriate to represent the cancer potency of PCB mixtures? If so, is the exposure pathway a reasonable default indicator of which end of the range is appropriate?*

Yes, I think a range is highly appropriate to represent the cancer potency of PCB mixtures. Caution is warranted, however, in how one describes the range of potencies that can be calculated from the various bioassay data. Specifically, it is likely that the range of potencies expressed in Table 3-1 is a reflection of different sensitivities of strains/sexes of rats as well as a reflection of different potencies of mixtures of PCBs. This is better illustrated in Table 5-3, where data for male rats exposed to Aroclor 1260 suggest that Wistar rats are ~15-fold more sensitive than Sprague-Dawley rats. It is interesting that, according to the slope estimates in Table 5-3, Sprague-Dawley female rats appear to be about an order of magnitude more sensitive than F344 rats to Aroclor 1254, but that male Sprague-Dawley rats are about an order of magnitude less sensitive than F344 rats (this is discussed further below).

So, given that the range of potencies expressed in Table 3-1 is likely related to strain- and sex-specific responses as well as differences in response to different mixtures, this should be clearly stated. The newer data from the GE study provide (with the exception of female Sprague-Dawleys exposed to Aroclor 1254) a very nice illustration of increasing cancer potency with an increasing degree of chlorination of the PCB mixtures

without strain and sex differences confounding the results. These newer data strongly support the use of a range of potencies for various exposure scenarios that, due to environmental processes (e.g., partitioning, transformation, and bioaccumulation) result in exposures to different congeners by different routes.

- *Is it important that exposure assessments include internal exposure that persists after external exposure stops? If so, is half-life in the body a reasonable way to do this given the information currently available to risk assessors?*

This is an interesting problem. It seems appropriate to consider internal exposures that persist after external exposure has ceased, and this is consistent with the decision made to not use a time-weighted-average daily dose for the rat bioassays done by Kimbrough et al. (1975) and Norback and Weltman (1985).

It also seems appropriate to use the biological half-life to determine a reasonable factor for estimating persisting internal exposures, but I'm wondering whether, since PCBs bioaccumulate, the length of initial external exposure should not influence this analysis. For example, would it be recommended that an exposure to less biologically-persistent congeners (e.g., ingestion of water-soluble congeners) be increased by a factor of 4 years regardless of whether the initial exposure was for 1 year or 10 years? Clearly, with a longer exposure, body burdens will be higher than those following shorter exposures to the same environmental concentrations. Accordingly, the internal exposure that results following cessation of external exposure, seems like perhaps it should be a function of the length of the initial exposure.

Has consideration been given to use a similar approach to analyzing the less-than-lifetime assays in laboratory animals?

- *Is the assessment correct in identifying food chain exposure as highest risk? Is it sufficient to say "risks may be higher that those estimated in this assessment" or should the risk estimates be multiplied by an explicit food-chain adjustment factor?*

Yes, it appears to be correct to identify food chain exposure as the highest risk. It is not really satisfying to state that for food chain exposures, risks may be higher than

those estimated in this assessment. Of course, risk managers faced with making decisions will want to know how much higher they might be.

Unfortunately, it does not appear that there are many data that can be used to provide scientific rationale for any particular adjustment factor. Reference is made on p. 28 to a study in Mink fed contaminated fish, with resulting toxicity being comparable to that seen in Mink fed Aroclor 1254 at levels 3-times higher. Is this the only study of this nature? Are there toxicity studies with individual congeners that may help us with this evaluation?

If there are no data to support the use of a particular adjustment factor, then the rationale for recommending the use of the 95% UCL on the high end of the range of potency factors can be more easily justified.

- *Is this assessment's approach of a range indexed by exposure pathway likely to be useful for incorporating new information? Should it be considered for non-cancer assessments?*

Yes, I think the approach of using a range, the use of which is dictated by specific exposure information, is useful for incorporating new information and should also be evaluated for noncancer assessments. In particular, the preliminary GE bioassay data can be easily incorporated into this approach, particularly because all studies were performed in the same strain of rat. To reiterate a point I made earlier, we should be careful not to imply that the range of potency factors calculated from the various bioassays is due solely to exposure differences; strain and sex differences are important as well.

- *Reaction to "new features" in this assessment (reflecting changes in new cancer risk assessment guidelines)*

Several innovative approaches to the assessment of cancer risk from PCBs are presented in this document and are reflective of changes found in the 1996 proposed cancer guidelines. The PCB assessment reflects in a very positive way the movement toward increased flexibility and emphasis on making risk assessments more transparent. One decision made in this analysis that perhaps should be reconsidered, however, is the decision to "drop" the two lowest cancer potencies (see p.22) because they may be

reflective of the lower sensitivity of males. While we certainly want to produce a risk assessment that is protective of both males and females (and which would therefore focus on data from the more sensitive gender), to "drop" this information early in the assessment may not be appropriate if one then goes on to indicate that the resulting range is for the population as a whole. Rather, consideration should be given to including all estimates, and perhaps even calling out separate ranges of potency factors for males and females.

One other area for consideration, in keeping with the 1996 proposed cancer guidelines, is to give greater consideration to how the data from less-than-lifetime exposures may be more explicitly factored in to the dose-response assessment for PCBs. Indeed, since most humans are exposed for only fractions of a lifetime, a more in-depth analysis of these data may be valuable.

Specific Comments on the Draft PCB Document:

In addition to the page-specific comments that follow, I suggest the addition of some text to help define some of the terms used in describing neoplastic changes in the liver. For example, are "adenomatous liver nodules" the same as "adenomas"? Terms that are used include carcinomas, adenomas, hepatomas, neoplastic nodules, adenomatous liver nodules, adenofibromas (not entirely clear to me what these are).

Page	Comment
9	It would be helpful to include more information about the differences seen between male and female rats -- what role do hormonal influences play?
10	I suggest moving the discussion of Moore et al. (lines 3-9) up to precede Table 2-1 -- perhaps before the discussion of the key studies starting on page 7.
10	More information should be provided on the results of the Kimbrough et al. (1972) study. Specifically: what were the incidences of adenofibrosis in rats fed lower doses (<1000 ppm) of Aroclor 1260? What were incidences at 1000 ppm for Aroclor 1254 (one assumes 100%)?. Also, it is stated that a difference between males and females was not seen for Aroclor 1254, but at 100 ppm, the incidence was 1/10 in males and 7/10 in females. This certainly seems indicative of a sex-related difference in dose-response.
12	Paragraph starting on line 19: what initiator was used in these studies?. Perhaps a brief statement on the significance of alterations of ATPase and GGT should be given here.
14	Line 19: Are there any data providing a quantitative analysis of absorption of PCBs from ingestion or inhalation?
15	Lines 3-5: If exposure to rhesus monkeys was occluded, this should be stated. Lines 18-19: What are typical half-lives for some of the lower chlorinated congeners? Lines 20-22: what was the time course in the experiment by Anderson et al., 1991? For how long was enzyme activity significantly higher?
19	Lines 5-6: Supralinearity in the experimental range does not preclude sublinearity in the lower-dose region, yes? Line 9: Suggest replacing "sublinear" with "nonlinear"
20	Line 2: This statement can be supported by referring to observed latency periods.

Line 23: It is true that in general the ED01 is just below the experimental range. For many of the PCB datasets, however, the incidence goes from 0 in controls to very high (80-90%) in the only exposed dose group.

22 Lines 24-25: While it appears that female rats are more sensitive than males to PCB-induced liver cancer, the statement here is somewhat limiting. It may well be that the males in the study by Schaeffer et al. (1984) exposed to Clophen A 30 had a lower tumor incidence because of the lower chlorination of the mixture (the males exposed to Clophen A 60 had a much higher response).

22 Lines 30-31: The ranges in Table 3-1 do reflect experimental uncertainty and variability of commercial mixtures, but they ALSO reflect strain and sex differences, which may be more similar to the variability one would expect to see in a heterogeneous human population.

23 Lines 12-15: More information (perhaps actual data) should be supplied here.

33 Line 4: More important that the **number** of PCB congeners that are dioxin-like is the fraction of total congeners in "typical" environmental or biological samples (i.e., one congener could account for 50% of a sample).

36 Lines 20-21: So - what do the data for PCBs suggest (when one compares the central estimate with the UCL)?

38 Lines 16-18: It is indicated here that "all" potency estimates from lifetime cancer studies have been used to develop a range. However, the two lowest were dropped....

40 Table 5-1: It appears (based on tumor incidences) that the same control animals were used for each of the 4 groups; however, the way the data are presented in the table, it appears that there were 340 control females and 392 control males. In footnote "a" to Table 5-1: When was the first tumor observed?

43 First bullet: It is true that the slopes for Aroclor 1242 and 1260 in females are quite similar; however, they are 30-fold different for male Wistar rats. So – it may be more appropriate to say that the difference in chlorine content has mixed results in terms of cancer potency.

Minor Editorial Comments:

Page	Comment
iii	Section 3.4: Insert "-SPECIFIC" after "CONGENER" Section 4.4: Should "TOXIC" read "TOXICITY"? (Note that throughout text, "toxic equivalency factor" is used -- I am more accustomed to seeing "toxicity" used here)
1	It would be nice to replace part of footnote #1 with a figure showing how PCBs are numbered. Also, include here that the last 2 numbers in the Aroclor numbering system represent the % chlorine in the mixture.
15	Line 3: Suggest replacing "through" with "following"
16	I suggest incorporating footnote #3 into the text (following line 13)
17	Line 23: Cite 1996 proposed cancer guidelines here?
19	Line 25: Insert "linearized" before "multistage". Also, first citation here is for EPA's Water Quality Criteria Documents -- seems that Howe et al. is more appropriate.
21	Line 12: Insert "for" before "which trends..."
22	Line 26: Note typo: Should be ED10s on line 26, not ED01s.
23	Lines 16-20: Is it common terminology to refer to foci "becoming extinct", as opposed to "regressing"?
26	Table 3-3 should indicate (in the table itself) that the TEFs are relative to a value of "1" for 2,3,7,8-TCDD.
37	Line 14: Insert "that" after "recommends" Line 15: Suggest re-writing as: "Although the half-life for a mixture can underestimate a specific congener's long-term persistence..."
38	Lines 14-15: Update now that proposed guidelines are out.
45	Line 18: Suggest rewriting: "4 years duration to exposures to less persistent congeners..."

Gary Williams

Are the studies fairly represented? Are any studies pertinent to dose/response assessment missing?

Page vi paragraph 2. A very sweeping statement is made about the health effects other than cancer. This statement should either be documented or eliminated. This statement does not appear to be based upon studies of PCBs in humans but on studies in animals or on effects found in the episodes in Japan and Taiwan, which were primarily due to polychlorinated dibenzofurans. Additionally, no mention is made of chloracne, which is the well-established human health effect of PCBs. For a discussion of human health effects see Delzell et al. (1994).

Page 13. Reference is made to weak initialing activity of PCBs. None of the findings noted provide evidence of weak initiating activity. Rather, these findings are evidence of tumorigenicity. Initiation refers to a specific initiation-promotion study in which PCBs would be given first, followed by the use of a promoting agent. The study by Hayes et al., (1985) was reported to be negative for initiating effects. The changes in altered foci in the liver noted in this paragraph can also be evidence of the action of a tumor promoter. Most importantly, PCBs are not considered DNA reactive. Whysner et al. (1995) have communicated results demonstrating a lack of DNA reactivity of PCBs by the sensitive ^{32}P-postlabeling method.

Page 14. This section might better be entitled toxicokinetics. Perhaps it should be noted that there is evidence for interactions between PCBs e.g., de Jongh et al. (1993).

Page 18, line 21. A statement is made about the production of mutagenic metabolites. The source in the literature for this finding is not clear. The original study should be cited. If these are the findings that have been reviewed by Safe (1989), the original experiments were reported to be non-reproducible.

Page 19. The shape of the dose-response curve is discussed. There is evidence of dose-response from tumor promotion studies. (Greim et al., 1984). In this study on Clopen A50, no effect was found at up to 0.5 mg/kg give three times per week, whereas the effects above 1 mg/kg were evident. Since PCBs are acting as tumor promoters, this study provides some evidence of a sublinear dose-response.

The section 3.4 discusses TEFs. However, the binding of dioxins to the Ah receptor and associated enzyme induction has been considered necessary but not sufficient for tumorigenic effects. Tumor formation appears to be based upon toxicity of dioxins. How can dose-response information be based upon a response that is necessary but not sufficient? There is evidence that a higher dose of dioxin is necessary to produce tumors compared to the dose required for Ah receptor binding. Consequently, the relative doses required for Ah receptor binding would not necessarily predict relative tumorigenic potency.

On page 33 various articles *in press* are referred to for supporting statements. Are published original research reports available?

Page 33. What is the evidence that bioaccumulated PCBs are more toxic than commercial PCBs?

Is a range (instead of a single value) appropriate to represent the cancer potency of PCB mixtures?

The EPA cancer risk assessment guidelines stated that the risks represent an upper limit and that the risks may be as low as zero. Based upon the mechanism of PCBs as tumor promoters, a threshold may be involved. (See also reference Greim et al., 1984). The mechanism of tumorigenesis supports the concept that the lower end of the range may very well be zero.

Page 44. Nothing in the document supports a cancer risk for humans.

References

1. de Jongh, J., Wondergem, F., Seinen, W. and Van den Berg, M. (1993) Toxicokinetic interactions between chlorinated aromatic hydrocarbons in the liver of the C57BL/6J mouse: I. Polychlorinated biphenyls (PCBs). Arch. Toxicol. 67:453-460.

2. Delzell, E., Giesy, J., Munro, I., Doull, J., Mackay, D. and Williams, G. (Expert Panel Members) (1994) Polychlorinated Biphenyls. Reg. Toxicol. Pharm. 20:S187-S307.

3. Greim, H., Deml, E. and Oesterle, D. (1984) Drugs and environmental chemicals as promoters. *In: Models, Mechanisms and Etiology of Tumour Promotion.* IARC Scientific Publication No. 56, pp. 487-494. M. Borzsonyi, K. Lapis, N.E. Day and H. Yamasaki (Eds.). IARC, Lyon.

4. Hayes, M.A., Safe, S.H., Armstong, D. and Cameron, R.G. (1985) Influence of cell proliferation on initiating activity of pure polychlorinated biphenyls and complex mixtures in resistant hepatocyte in vivo assays. JNCI 74:1037-1041.

5. Safe, S. (1989) Polychlorinated biphenyls (PCBs): mutagenicity and carcinogenicity. Mut. Res. 220:31-47.

6. Whysner, J., Montandon, F., Verna, L.K., McClain, R.M. and Williams, G.M. (1995) ^{32}P-Postlabeling assay for detection of adduct formation by phenobarbital, chlordane and polychlorinated biphenyls in mouse liver DNA. The Toxicologist 15:83.

APPENDIX D

AGENDA

United States Environmental Protection Agency
National Center for Environmental Assessment

Peer Review Workshop on *PCBs: Cancer-Dose Response Assessment and Application to Environmental Mixtures*

Holiday Inn Bethesda
Bethesda, MD
May 21-22, 1996

Agenda

Tuesday, May 21, 1996

8:30AM **Opening**

Director's Welcome and Remarks ..... *Michael Callahan, National Center for Environmental Assessment (NCEA), U.S. Environmental Protection Agency (EPA), Washington, DC*

Logistical Information ..... *Susan Brager, Eastern Research Group, Inc. Lexington, MA*

Chair's Remarks and Charge to Panel ..... *Loren Koller, Oregon State University, Corvallis, OR*

Disclosure of Conflicts of Interest ..... *Panel*

9:30AM **EPA Remarks**

Overview of PCB Assessment ..... *James Cogliano, NCEA, U.S. EPA, Washington, DC*

Research Perspective ..... *Linda Birnbaum, National Health & Environmental Effects Research Laboratory, U.S. EPA, Research Triangle Park, NC*

Tuesday, May 21, 1996 (continued)

9:30AM (continued)	**Regulatory Program Perspective**	*Edward Ohanian, Office of Water, U.S. EPA, Washington, DC*
	Regional Office Perspective	*Marian Olsen, Region 2, U.S. EPA, New York, NY*
10:00AM	**Panel Questions to EPA**	
10:30AM	BREAK	
10:45AM	**Presentation on Battelle/General Electric (GE) Rat Studies**	*Brian Mayes, General Electric Company, Schenectady, NY*
11:15AM	**Panel Questions of GE**	
11:30AM	LUNCH	
12:30PM	**Public Comment**	
2:30PM	BREAK	
2:45PM	**Panel Remarks and Discussion**	
4:30PM	ADJOURN	

Wednesday, May 22, 1996 *

8:30AM	**Panel Discussion and Drafting of Comments**
12:00PM	LUNCH
1:00PM	**Finish Drafting Comments**
4:30PM	ADJOURN

* Please note that the second day of the workshop is scheduled as a working session for the panel to draft the workshop summary report. Observers will not be permitted to comment during this time.

APPENDIX E

FINAL PRESENTER LIST AND SCHEDULE FOR PUBLIC COMMENT

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National Center for Environmental Assessment

Peer Review Workshop on *PCBs: Cancer-Dose Response Assessment and Application to Environmental Mixtures*

Holiday Inn Bethesda
Bethesda, MD
May 21-22, 1996

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Peer Review Workshop on *PCBs: Cancer Dose-Response Assessment and Application to Environmental Mixtures*

May 21-22, 1996
Bethesda, MD

SCHEDULE FOR PUBLIC COMMENT

May 21, 1996—12:30-2:30PM

12:30PM	John Schell, TERRA, Inc.
12:45PM	Brent Finley, McLaren Hart/Chemrisk
1:00PM	Thomas Starr, ENVIRON Corporation
1:15PM	
1:30PM	
1:45PM	
2:00PM	
2:15PM	

Additional requests:

APPENDIX F

FINAL OBSERVER LIST

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National Center for Environmental Assessment

Peer Review Workshop on *PCBs: Cancer-Dose Response Assessment and Application to Environmental Mixtures*

Holiday Inn Bethesda
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May 21-22, 1996

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