Estrogenicity of Environmental PCBs

The paper by Bergeron, Crews, and McLachlan, "PCBs as Environmental Estrogens: Turtle Sex Determination as a Biomarker of Environmental Contamination" (EHF 102:780–781) presents data on the estrogenic activity of 11 chlorinated biphenyls or diphenyl ethers, or hydroxylated derivatives thereof, selected so as to represent a variety of structural types. Some of the PCB types examined (e.g., compounds A, C, D, E, and J) arise from PCB congeners actually detectable in the commercial Aroclors (1) and hence also in environmental samples, whereas other PCB types (e.g., those with heavily uneven chlorination of the two rings, as in compounds F,G, and L) arise from PCB congeners that are not detectable in either the Aroclors themselves (1) or even environmentally transformed PCB compositions (2). It is noteworthy that the only compounds found to have statistically significant estrogenic activity (compounds F and G) both represented 4-hydroxylation products of PCB congeners belonging to the nonenvironmental group, whereas the five compounds representative of PCB structures actually present in the environment were all negative. In short, the results present zero evidence that environmental PCBs present risk of estrogenic activity.

This is hardly what the authors claim, however. In their discussion they state (p. 781):

This report contributes laboratory evidence of the effect of PCBs on sex determination . . . and serve as a warning of conditions threatening wildlife populations. The PCB levels reported here as effective in disrupting normal gonadal differentiation in the turtles are comparable to average levels of PCBs found in human breast milk in industrialized nations.

This most misleading statement represents a false alarm that can only impede the search for the environmental contaminants that actually do present estrogenic risk.

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Response to Hamilton

There are several points we would like to address in our response. First of all, the compounds studied were chosen because these particular congeners were believed to be estrogen based on their conformational structure. The primary reason for conducting this type of study was to show the effectiveness of a temperature-dependent sex determined (TSD) species as a tool in assessing estrogenic activity in vivo. This point is furthered by an earlier report in EHP by Guillette et al. (1). While the PCB compounds we used may not be primary components of commercially available PCB mixtures, there are parallels between these compounds and other PCB congeners in the pattern of ortho-chlorine substitutions, as Korach et al. (2) indicate. It is important to study how these structures affect a developing organism in order to understand how PCBs can act as estrogens. Though these particular congeners may not be currently used in the readily available mixtures, McKinney et al. (3) make reference to goals of using PCBs that are easily detoxified via hydroxylation. If such considerations lead to composition of commercially available PCB mixtures away from congeners that exhibit a dioxinlike toxicity, these considerations should include assessment of the estrogenicity of these compounds. The TSD system can be used to test such mixtures in vivo. Furthermore, the appearance of hydroxylated PCB congeners in "nature" is an emerging issue: for example, Bergman et al. (4) report that the hydroxylated forms of heavily chlorinated (penta- or heptachlorinated) biphenyls were among the most retentive forms of PCBs found in blood from humans or seals. Clearly, this class of molecules may have environmental significance which is only now being appreciated.

The second point that we would like to emphasize regards the potential for second-generation exposure to PCBs as environmental estrogens. While the congeners that we found to clearly exhibit estrogenic activity may not be produced in the commercial PCB mixtures, or even found in soil and water samples, they may exist within animal tissue during metabolism of the environmental compounds. Maternal metabolic by-products may affect the reproductive system at a critical stage in development of the offspring, producing the detrimental estrogenic effect on the second generation. This is particularly a concern when enhanced estrogenic effects are produced by the synergy of different combinations of low-level PCBs, an issue brought to light by the study in question.

Finally, we share Dr. Hamilton's concern that false alarms may impede identification of contaminants that actually present risk of estrogenic activity. However, Dr. Hamilton's cited quotation of our discussion, and his interpretation of it, require clarification so as not to be misleading. Dr. Hamilton's abbreviated quote would lead readers to believe that it is our report which we say serves as a warning of threatening environmental conditions. In fact, the passage he omitted clearly identifies "the usefulness of a TSD species as a biomarker" to serve in the capacity which Dr. Hamilton apparently ascribes to our report.

Our findings support the call from the scientific community for the need to further study the mechanisms of estrogenic activity of environmental contaminants. These findings, together with the growing body of evidence that a number of environmental compounds mimic estrogens and do have an effect on developing reproductive systems, provide ample indication for further investigation of these mechanisms and their outcomes. Our report suggests a model by which to continue these efforts, and we appreciate the continued interest in our work and the opportunity to address questions regarding it.

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Human Toxicity of PBDDs and PBDFs

The brilliant literature review and health risk assessment by Mennear and Cheng-Chung, "Polybrominated Dibenzo-p-dioxins and Dibenzofurans: Literature Review and Health Assessment" [EPH 102(suppl 1):265-274], states that "reports of human toxicity associated with exposure to PBDDs and PBDFs were not found" (p. 272). In fact, in their review, no references are discussed or quoted regarding human studies.

Two papers have been published on the human toxicity of these compounds. The first (1) is a recent report, previously presented at the Dioxin '90 Congress (2), about a chemist who was exposed to 2,3,7,8-tetrabromodibenzo(dioxin (TBDD) and to 2,3,7,8-tetrachlorodibenzo(dioxin (TCDD) in March and September 1996, respectively, when synthesizing these chemicals. The chemist was defined as "in good health" in 1990, when determinations of chlorinated and brominated dioxins and dibenzo furans were performed on whole blood. High concentrations of several congeners were detected, and the results were used to discuss the half-life of the chemicals in humans. The subject presented a mild chloracne after an unspecified time from his exposure to bromodioxins in March, suggesting that TBDD could produce skin effects as chlorodioxins. Other more relevant symptoms occurred after the exposure to TCDD in September, and the patient was hospitalized for a short period.

The second was a study of subjects exposed to PBDDs and PBDFs as a result of working at a BASF factory in etrusion blending of polybutylene terephthalate with decabromobiphenyl ether, used as a flame retardant. The intensity of exposure was determined in 1990 through air monitoring (3). The paper presents blood levels of 2,3,7,8-TBDD and TBDD and of total congener profiles for some exposed workers and the results of a comparison of several immunological tests in a population of exposed versus a population of unexposed deriving from the same working cohort. Workers had detectable blood levels of TBDD and TBDF; half-life estimates of these chemicals are presented. The results of immunological tests were described as "not adversely impacted at these burdens of PBDDs and PBDFs," even though the results of several tests showed a correlation with exposure, and in the subject having the highest blood levels of PBDDs and PBDFs, immunological changes were quite relevant. The authors stated that clinical examination did not reveal "skin lesions consistent with an acenegetic response."

It should be stressed that the results of the two quoted articles do not change the conclusions of Mennear and Cheng-Chung on the health risks of PBDDs and PBDFs. However, slightly different suggestions for future research can be derived.

Human populations have been or are exposed to these chemicals because of their use in several work processes involving flame retardants, environmental exposures (mainly due to municipal incinerators), or because of accidents due to thermal decomposition of flame retardants. These exposed human populations can be suitable, at least in theory, for toxicological and epidemiological observations.

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Arsenic Risk Assessment

A commentary highly critical of two of our published studies was recently published in Environmental Health Perspectives (Carlson-Lynch et al., 102:354-356). One of our papers examines the epidemiological evidence for a methylation threshold for inorganic arsenic and concludes that there is no consistent evidence to support the hypothesis of such a threshold in humans (1). The second paper criticized by Carlson-Lynch et al. estimates the human cancer risks of arsenic at several sites (lung, liver, kidney, and bladder) using linear extrapolation from Taiwanese data (2). Carlson-Lynch et al. contend that the analyses conducted in our studies are flawed and that the conclusions reached in our publications are erroneous, rendering them unsuitable for use by the EPA in risk assessment.

We either not our studies are used by the EPA for risk assessment is of little concern to us, but we are certainly concerned about statements that they are flawed. Careful examination will show that all of the major points raised in the commentary are either incorrect or have no valid basis. We would like to respond to the criticisms made, point by point in the order presented, beginning with the methylation paper (1).

Critique: The average arsenic exposures in almost all the studies analyzed were too low to observe methylation saturation.

Response: The commentators base this statement on three issues. First, the authors state that evidence from an experimental study (of only four human volunteers each receiving only one dose level) suggests that methylation would be completely saturated at exposures greater than 500 μg/day (3). However, at the highest oral dose in this study, 1000 μg/day, the amount of urinary arsenic in the inorganic form was only 25%, hardly demonstrating methylation saturation even at this level. Bucher et al. (3) state that they made the statement that "speciation of the arsenic metabolites in urine indicated that the arsenic methylation capacity of the human body was not yet saturated, even with an oral daily dose of 1000 μg As."

The evidence of any metabolic saturation from this study is not conclusive. Each of four arsenic dosing levels was assigned to a different individual subject, making it impossible to differentiate interindividual differences in methylation efficiency from dose-dependent effects that might apply to a general population.

Second, the authors state that we analyzed only two groups with average urinary arsenic levels at or above 190 μg/l, which they hypothesize corresponds to the concentration above which methylation saturation occurs. This statement obscures the fact that 1) the two groups combined had a total of 35 people, 2) our analysis of available individual data (see Figure 2 of our paper) included 14 persons with urinary arsenic levels >190 μg/l. No trend of higher relative proportions of unmodified arsenic is suggested for those 14 individuals.

Third, the authors state: "... a regression analysis on the individual data within the Yamauchi et al. (4) population was borderline significant at p = 0.10. . ." (p. 354). However, this was just one of nine regression analyses we presented. The slopes were positive in four (including the Yamauchi study) but negative in five (J: Table 9).

As a matter of interest, in our more recent studies of chronically exposed popu-