CONSIDERATION OF A LINK BETWEEN DIOXINS AND DIABETES

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Introduction

Humans are exposed to halogenated aromatic compounds (HAHs) occupationally and environmentally, but particularly from fatty foods such as meat, fish, and dairy products, at the rate of some 3-6 pg of TEQ/kg bw/day.[1] Occupations that formerly also led to exposure included the manufacture and use of 2,4,5-trichlorophenol and associated products such as herbicides, and bleaching of wood pulp with chlorine.[2, 3]

Like diabetes,[4, 5] the lethal toxicity of TCDD is a metabolic disorder involving abnormal energy metabolism. For the TCDD-induced wasting syndrome of rats, Rozman demonstrated nearly identical dose-response behavior by TCDD for suppression of food intake and inhibition of the enzyme phosphoenolpyruvate carboxykinase (PEPCK) involved in gluconeogenesis.[6]

The epidemiological studies noted below have shown a consistent weakly positive association between accidental or occupational exposure to TCDD and glucose metabolism disorders, including insulin resistance, hyperinsulinemia and adult onset (type II) diabetes.

- German employees of factories where phenoxy herbicides were manufactured[7]
- US employees of factories in Newark, NJ and Verona MO where 2,4,5-trichlorophenol was produced [8, 9]
- Residents of Seveso, Italy where a 1976 explosion at a 2,4,5-trichlorophenol factory [10]
- US Air Force veterans involved in spraying defoliants during the Vietnam War [11]

In all these cases, exposure occurred many years previous to the epidemiological studies. Dioxin levels at the time of exposure must be back-calculated from current concentrations in serum lipids, using the whole body half-life of TCDD. This is quite variable in humans, 5.8-9.6 years,[12] probably reflecting differences in body fat, which sequesters HAHs. The study subjects were all exposed specifically to TCDD (a ubiquitous contaminant in 2,4,5-trichlorophenol and 2,4,5-T), but also to much larger amounts of phenols and phenoxy herbicides; both they and the referents were also exposed to other HAHs.

We have argued that the statistical association might involve opposing biochemical actions of the activated aryl hydrocarbon receptor (AhR) and peroxisome proliferator activated receptors (PPARs).[13] Activated PPARg promotes the differentiation of adipocytes, which mediate glucose and lipid homeostasis, and expression of the glucose transporter protein GLUT4. Consistent with this mechanism, thiazolidinedione drugs, which bind to and activate PPARg in adipocytes, enhance insulin-mediated removal of glucose from the circulatory system via increased expression of GLUT 1 and GLUT 4. By contrast, dioxin-like compounds inhibit pre-adipocyte differentiation and reduce glucose transport activity and copy number of both GLUT4 and its mRNA in adipocytes from TCDD-treated guinea pigs or mice by Ah receptor-mediated mechanisms. Type II diabetes has also been linked to tobacco smoking, which also involves Ah receptor agonists, with odds ratios for diabetes, corrected for confounding by obesity, remarkably similar to those for individuals accidentally or occupationally exposed to dioxins.[14]

Sweeney et al.[8] compared 281 living male US workers, employed more than 15 years earlier in the production of chemicals contaminated with TCDD, with an unexposed group of 260 volunteers. The

workers had a mean serum lipid-adjusted level of 220 pg TCDD/g lipid (= ppt); that of the referents was 7 ppt. Exceptionally, these investigators measured serum lipid concentrations of other HAHs, which were similar in both workers and controls. The TEQ values for workers and referents were 256 and 35 ppt.[13] There was a slight increase in the risk of diabetes (OR = 1.12, P < 0.003) and high fasting serum glucose (P < 0.001) with increasing serum concentrations of TCDD, but age, weight and family history remained the principal risk factors in the development of diabetes.[8] In a follow-up study of the same cohort, but excluding individuals under treatment for diabetes, the odds ratio for diabetes as a function of serum lipid TCDD concentration was 1.49 (95 % CL 0.77, 2.91) for all subjects, irrespective of TCDD level, but no dose-response relationship was seen between (present) TCDD levels of the exposed workers and either diabetes incidence or mean glucose levels. [10]

We were interested in whether the available data could also yield an estimate of the risk of diabetes in the nominally unexposed population of the 1997 study by Sweeney et al., assuming their reference population to be typical of "unexposed" North Americans. On the basis of a first order elimination of TCDD, the integrated dose of dioxin-like compounds is the sum of two terms: Eq. [1], in which $[TCDD]_t$ is the instantaneous concentration of TCDD, which gradually falls between the time of exposure and the time of measurement.

[1] $$\text{integrated exposure} = (\text{background TEQ H t}) + \int [TCDD]_t \cdot dt$$

The work of Rozman[15] indicates that the lethal toxicity of 1,2,3,4,6,7,8-HpCDD towards Sprague-Dawley rats could be explained on the basis of Haber's Rule. This states that for a given level of response the product of concentration (c) and time of exposure (t) is constant: $c \times t = K$. The in vivo half-life of HpCDD is sufficiently long that its whole body concentration was almost constant during the study, and so $c \times t$ represents the integrated concentration. Our objective was to estimate, on the assumption that Haber's rule applies to a link between dioxin exposure and diabetes, whether the referents' lifetime 35 ppt of TEQ might represent a significant exposure in the context of chronic toxicology. For a variable exposure, allowing consideration of uptake and depuration, the proper form of Haber's Rule is $\int c(t) \cdot dt = K$, where $c(t)$ is the instantaneous concentration of the toxicant.

We assumed, since the study subjects were all adults, that the background exposure was constant with time and took the conservative position that it had this level throughout life. The second term in Eq. [1] is the integrated concentration associated with the exposure to TCDD, which we assumed for the purpose of this analysis to be proportional to the increased probability LIP of an adverse toxicological outcome (diabetes): $LIP = \alpha \int [TCDD]_t . dt$, where the constant a relates AAUC to probability. Having LIP scale linearly with TEQ is a conservative assumption that there is no threshold to the diabetogenic effects of dioxins.

The non-background concentration of TCDD at any time subsequent to the initial exposure is given by Eq. [2]. Since [TCDD] was actually measured in 1991, earlier values can be back-calculated using first order kinetics: $[TCDD]_{1991} = [TCDD]_t e^{-kt}$. The rate constant k depends on the whole-body half-life for TCDD: i.e., $k = \ln(2)/t_{½}$.

[2] $$[TCDD]_t = [TCDD]_{1991} e^{+kt}$$

The integrated exposure to TCDD since any time t previous to the measurement in 1991 and excluding background is given by Eq. [3].

[3] $$\text{integrated exposure} = \int [TCDD]_{1991} e^{+kt}) . dt = [TCDD]_{1991} [e^{+kt} - 1]/k$$

Results

The data of refs 8 and 10 allowed us to assign numerical values to the parameters in Eq. [3] and to LIP. The exposed population consisted of 279 workers, of average age 55.4 years, who had been employed for an average of 34.5 years prior to the study at a chemical plant whose products may have been contaminated with TCDD. The incidence of diabetes was 26 out of 279 in the workers and 18 out

of 258 in the referents, the latter of which are normative values,[16] hence LIP = (26/279) – (18/258) = 0.093 – 0.070 = 0.023.

The workers' average serum lipid concentration of TCDD was 221 ppt in 1991 (total TEQ was 256 ppt). They were compared to 258 matched but unexposed referents, of average age 56 years, whose background TEQ was 35 ppt, of which 7 ppt was attributable specifically to TCDD. The elimination rate constant k was taken to be 0.11 $year^{-1}$, a mean value in the range 0.077 to 0.14 $year^{-1}$ (ref. 11), yielding an integrated TCDD burden of $[TCDD]_{1991}[e^{+kt} – 1]/k = 8.7$ H 10^4 ppt yr, for t = 34.5 years. Since $a = (kDP)/[TCDD]_{1991}[e^{+kt} – 1]$, the foregoing data gave $a = 2.6$ H 10^{-7} $(ppt\ yr)^{-1}$.

We then applied the value of a to the background exposure. The integrated TEQ is 35 ppt H 56 yr = 1925 ppt yr. On the assumption that DP also scales linearly with the integrated background TEQ, we estimate that LIP for exposure to a background TEQ would be (35 ppt H 56 yr) H (2.6 H 10^{-7} ppt $yr)^{-1}$ = 5 H 10^{-4}. This is the estimated increase in risk of diabetes due to a 35 ppt TEQ (background exposure) relative to a background risk of 0.07, using TEQ concentration in serum lipid as a surrogate for exposure.

Discussion

One can cite many reservations about the estimate concerning background exposure to dioxins as a risk factor for diabetes. It refers to a single study with a modest number of participants. The levels of exposure of both referents and subjects over the duration of the exposure required assumptions to be made, notably that Haber's Rule is valid for conceptualizing long term exposure to persistent toxicants such as members of the dioxin-like family of compounds. Nevertheless, we estimate that factoring out background exposures to dioxins would reduce the risk of diabetes in the nominally unexposed population by less than 1 % of the 0.07 actually observed. This argues strongly that present levels of dioxins are unlikely to constitute an important risk factor for type II diabetes in the general population. The parameter that contributes most to the uncertainty of the results is k, the half-life for TCDD elimination: using the extreme values for k, namely 0.077 and 0.14 yr^{-1}, gives DP(background) as 1 H 10^{-3} and 2 H 10^{-4} respectively, which does not change the conclusion of our study.

Generalizing, we believe the following to be the significant aspects of our study. First is the modification of Haber's Rule to relate the area under the exposure curve to an increase in the probability of a toxic outcome. Second is the use of this relationship to correct for the lack of an unexposed referent population. This approach should be generally applicable in epidemiological studies involving persistent, ubiquitous pollutants.

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