DIOXIN AND THE AHR: THE BEGINNINGS AND NO END IN SITE

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Otto Hutzinger – The Early Days

In the late 1960's, Otto Hutzinger was a newly hired Research Officer at the National Research Council (NRC) Regional Laboratory on the campus of Dalhousie University in Halifax, Nova Scotia. His interest in PCBs and Dioxins resulted from a conversation with Vlado Zitko, a scientist with the Fisheries Research Board in St. Andrews New Brunswick. Vlado indicated his concern regarding organochlorine environmental contaminants and suggested that environmental chemistry and impact of these compounds would be a "hot" area of research. I was also a Research Officer at NRC and Otto persuaded me to get involved in this new area of environmental research which would give us independence from our immediate supervisors and also be lots of fun. I reluctantly agreed, and from 1971-1974, we coauthored 35 refereed publications, at least 10 review articles, and two books, "The Chemistry of PCBs" (Hutzinger, Safe and Zitko) and "Mass Spectrometry of Pesticides and Pollutants" (Safe and Hutzinger) and founded a small company. Some of this early work described the first studies showing that PCBs could be photodegraded and metabolized (1-3), and these collaborative studies were continued after Otto was appointed Professor Environmental Chemistry at the University of Amsterdam in 1975 and I joined the University of Guelph in 1973. One of the lessons I learned from Otto was his attitude toward senior authorship and credit – he always favored his colleagues over himself.

After leaving NRC, Otto continued his studies on organohalogen pollutants and was the first to discover the formation and emissions of polychlorinated dibenzo-*p*-dioxins (PCDDs) and dibenzofurants (PCDFs) from municipal waste incinerators. Otto was an original thinker and mentor to a generation of environmental scientists at the Universities of Amsterdam and Bayreuth, and he was continually organizing research conferences (starting with Dioxin 1 in Rome), editing journals (e.g. *Chemosphere*), and books (e.g. *The Handbook of Environmental Chemistry*). Otto was one of a kind and is greatly missed.

Dioxin and the Ah Receptor

The combination of several poisoning incidents due to occupational and accidental release of 2,3,7,8-tetrachlorodibenzo-*p*-dioxin (TCDD) and contaminated PCBs in Fukuoka (Yusho poisoning), Taiwan (Yucheng poisoning), and Seveso (TCDD) coupled with their widespread environmental contamination, stimulated scientific, regulatory and legal concerns about halogenated aromatics. All of these issues and their resolution were presented and discussed during the annual "Dioxin Symposia". Although some occupational exposures and the Seveso accident primarily involved a single toxicant, TCDD, other accidents and environmental exposures to persistent organic pollutants (POPs) involved TCDD and many other individual PCDD, PCDF and PCB congeners. The analytical approaches included development of high resolution separation and detection approach which are now routinely used to quantity POP congeners from multiple sources at the sub-parts-per-trillion level. Since human and other biota are exposed to complex mixtures of POPs, it was imperative to develop hazard and risk assessment paradigms that could quantitatively assess mixture-induced responses. Pioneering work by Dr. Alan Poland (4)

showed that the mechanism of action of TCDD and structurally related PCBs, PCDDs and PCDFs involved initial binding to an intracellular protein designated as the aryl hydrocarbon receptor (AhR). This was later confirmed in AhR knockout mouse models where the characteristic AhR-mediated toxicities for TCDD and structurally related compounds were observed in wild-type but not AhR-knockout (AhRKO) mice. Thus, it was possible to develop a dioxin or toxic equivalents (TEQ) approach for risk assessment of TCDD- or dioxin-like compounds (DLCs), where

$$TEQ = \sum[(DLC_i) \times (RP_i)]$$

the dioxin equivalents is equal to the summation of the individual concentrations of the DLC (DLC_i) times their relative potency (RP_i) compared to TCDD (arbritarily set at 1.0) (5). This initial approach has been continually refined and modified as new data become available and has been used extensively by regulatory agencies to reduce emissions and environmental/human exposures to DLCs. The TEQ approach was invaluable for estimating the potential toxicity of an important sub-class of POPs (i.e. DLCs); however, evaluation of the potential adverse effects of non-DLCs is still a major regulatory problem

The AhR and Its Ligands: Health and Therapy

Although AhRKO mice are viable and reproduce, initial studies by Bradfield and others have identified an increasing number of defects in these animals, demonstrating that the receptor may play a role in organ/tissue homeostasis (6,7). For example, AhR-deficient mice exhibit a decreased liver size but only during development due to defects in closure of the ductus venosus. AhRKO mice have difficulty in maintaining pregnancy, they have an increased susceptibility to infection indicating a role for the AhR in the immune system function and the AhR also plays a role in stem cell development. Moreover, there is extensive evidence from transgenic animal studies that the AhR plays a critical role in intestinal health via interactions with microbiota-derived AhR metabolites and the AhR exhibits tissue-specific promotion or protection from cancer (8). Ongoing research continues to add to the growing list of AhR functions in animal models and to a lesser extent in humans and this makes the AhR an ideal target for drug development.

Receptors such as the estrogen receptor are among the most well developed targets for drug development since receptor-mediated adverse effects can be treated with receptor antagonists and receptor-mediated health benefits can be enhanced by receptor agonists. Although AhR-interactions with TCDD and related compounds lead to well characterized toxicities, the AhR also binds several endogenous biochemicals, health promoting phytochemicals, AhR-active pharmaceuticals, multiple microbiota-derived AhR ligands and synthetic AhR antagonists (9). These compounds are selective AhR modulators (SAhRMs) that exhibit tissue-specific AhR agonist or antagonist activities and can be exploited for treatment of multiple diseases including cancer, inflammatory bowel disease, immune and autoimmune diseases and expansion of stem cells. Development of AhR-active drugs has been a "cautionary tale" due to concerns of "dioxin-like" side effects; however, some of the current AhR-based drugs in clinical trials include Laquinimod for treatment of multiple sclerosis and StemReginin 1 (SR1), an AhR antagonist use for production of hemapoietic stem cells.

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