

## Cell bioassays (DR-CALUX<sup>®</sup> and Micro-EROD) for Ah receptor agonists

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Recently, several countries agreed to adopt the recommendations of the Stockholm Convention on persistent organic pollutants (POPs). One of the obligations will be to add new POPs to the list of restricted substances as new scientific evidence becomes available. Newly developed screening methods for dioxin-like POPs have made it possible to evaluate these substances with greater time and cost efficiency. Micro-EROD (measuring CYP1A1 induction) and DR-CALUX<sup>®</sup> (measuring luciferase induction) are two state-of-the-art technologies that can be used to evaluate Ah receptor activity. As a first step, we began a cross-validation study in June 2000 to analyze samples and mixtures of PCBs and PXDD/Fs, fly ashes, emission gases, PCB capacitor oils, mineral oils, sediments and feedstuffs. As a second step, we applied these cell bioassays to several dioxin-like compounds to validate our assays and to establish bio-TEF (or REP) values relative to 2,3,7,8-TCDD. We tested several mixed polyhalogenated dibenzodioxins and furans (PXDD/Fs; X = Br, Cl, F), polyhalogenated biphenyls (PXBs; X = Br, Cl), polybrominated diphenylethers (PBDEs), and technical mixtures of brominated flame retardants (Firemaster BP6, Dow FR250). For the PXDDs, similar REPs were found between the chlorinated and brominated congeners, while for the PXDFs significant differences were measured. A decrease in dioxin-like-potency with increasing halogenation was confirmed.

The following compounds did not show TCDD-like activity in the DR-CALUX<sup>®</sup> (EC<sub>50</sub>: 13.9 pM; LOQ: 1.1 pM; R<sup>2</sup>: 0.984; n: 108) or Micro-EROD (EC<sub>50</sub>: 5.0 pM; LOQ: 1.2 pM; R<sup>2</sup>: 0.955; n: 150) bioassays in the applied concentration ranges: 2,2',4,5',6-PBB; p-bromophenol; 2,4-bromophenol; TBBPA; 2,2,4,4'-T4BDE; 2,2,4,4',5-P5BDE; 2,2,4,4',5,5'-H6BDE; 2,3,3',4,4',5,6-H7BDE. So far, of the PBDE congeners, only PBDE congener 126 has shown activity.

Several PAHs were analysed and showed strong Ah-receptor-binding capacities at much higher concentrations, resulting in lower bio-TEF values. Of sixteen polychlorinated naphthalene congeners (PCNs) tested, only the 1,2,3,4,6,7-hexa-naphthalene congener showed an *in vitro* firefly luciferase induction. Further research is needed to understand the impact of these Ah receptor agonists on the potency of dioxin-like compounds in wastes and environmental samples.