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Use of Physiologically Based Pharmacokinetic (PBPK) Models in Marine Mammal Toxicology

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Abstract—Physiologically based pharmacokinetic (PBPK) models are mathematical models that are largely based upon the physiological characteristics of the species and the biochemical properties of the chemical of interest. They quantitatively describe and predict the kinetics of pollutants inside the body and can be of major importance for risk assessment of chemicals in marine mammals. PBPK models which consist of five compartments (liver, blubber, kidney, brain, and the rest of the body) were made for selected polychlorinated biphenyls (PCBs) and polybrominated diphenyl ethers (PBDEs) in order to address the bioaccumulation of these compounds in tissues of harbour porpoises (*Phocoena phocoena*). Harbour porpoises have relatively long life spans, are common cetaceans in the North Sea, a heavily polluted area, and are known to be very sensitive to pollution. Models developed for all compounds (some PCBs and PBDEs) were evaluated using existing datasets from the literature and from analyses performed by GC-MS, the latter being obtained from stranded porpoises in the Black Sea and the North Sea over a period of 18 years (1990–2008) to assess spatial and temporal trends in bioaccumulation of the respective PCBs and PBDEs. We demonstrate that PBPK models are a feasible computational approach that can be used as a non-destructive tool for predicting the chemical pollution status of the marine mammals.

Keywords: PBPK modeling, harbour porpoises, PCBs, PBDEs, spatial trends, temporal trends

BACKGROUND

Physiologically based pharmacokinetic (PBPK) modeling is an *in silico* or computer-based technique that allows to show the distribution and kinetics of chemicals in the body of an organism (Andersen, 1995; Reddy *et al.*, 2005;

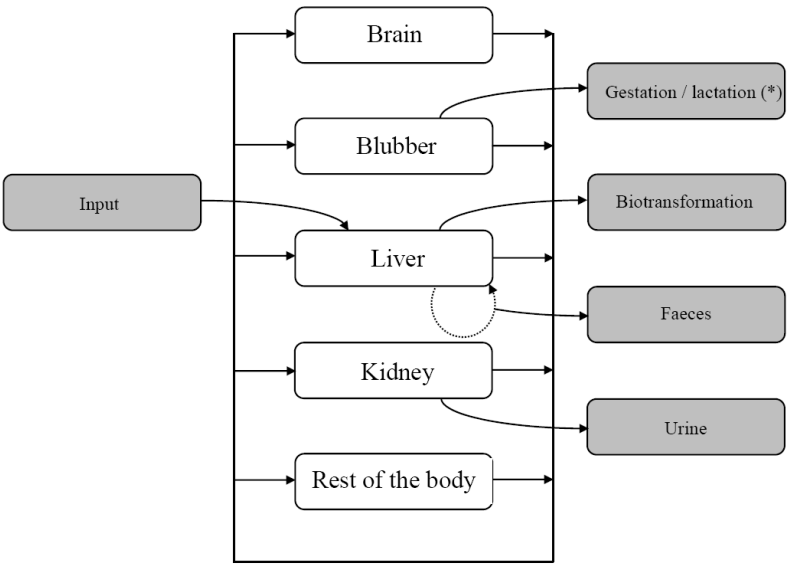

Fig. 1. Conceptual diagram of the PBPK model for PCB 153 in male and female ((*) included) harbour porpoises. Figure taken from Weijs *et al.* (2010).

Clewell and Clewell, III, 2008). Divided into several compartments, the body represents a mass-balanced system in which the movements of the compound obey the biochemical properties of the compound and the physiological characteristics of the organism of interest (Fig. 1). Usually, a PBPK model follows on or is strongly attached to a certain exposure experiment in organisms like rodents (Lee *et al.*, 2002, 2007; Emond *et al.*, 2010). Both compound-specific and species-specific parameters are then largely deduced from the experimental set-up.

However, this approach is not possible as *in vivo* exposure experiments are undesired and prohibited in marine mammals. For decades, toxicological research in marine mammals has focused mainly on the analyses of chemicals in tissues of dead, stranded animals and sometimes also in blood taken from live animals (e.g., Law *et al.*, 2002; Houde *et al.*, 2006; Lebeuf *et al.*, 2007; Weijs *et al.*, 2009a, b). Currently, *in vitro* exposure experiments in blood or liver cell lines of marine mammals are gaining more and more attention (McKinney *et al.*, 2006; Das *et al.*, 2008; Frouin *et al.*, 2010). This has the potential to go deeper into the toxic mechanisms of pollutants, but lacks the knowledge to put the observed effects into a broader perspective as it focuses on one tissue only. For marine mammals, PBPK models can be seen as a non-destructive replacement for the traditional *in vivo* investigations that can combine results of recent *in vitro* research and on-going biomonitoring efforts.

PBPK MODELING IN MARINE MAMMALS

In the past, several attempts were made to develop PBPK models for some pollutants in marine mammals. PBPK models for hydrophobic contaminants were presented by Hickie *et al*. (1999) and applied to the sum of PCBs in beluga whales (*Delphinapterus leucas*). The same was done for PCBs in killer whales (*Orcinus orca*; Hickie *et al*., 2007) and for individual PCB congeners in ringed seals (*Phoca hispida*; Hickie *et al*., 2005). PBPK models have species-specific parameters which make that models developed for one species are of limited value to assess bioaccumulation in tissues of other species (Clewell and Clewell, III, 2008). Thus, we have developed new models for harbour porpoises, one of the most abundant marine mammal species in the European North Sea. Harbour porpoises accumulate high loads of contaminants and are considered to be sensitive to pollution because of their limited capacities for metabolic biotransformation of some pollutants (e.g., PCBs and PBDEs) compared to other species such as harbour seals (Weijs *et al*., 2009a). Linking body burdens to *in vitro* toxic effects would enhance our understanding of the pollution status in these animals and would create opportunities towards their conservation. Therefore, assessing body burdens through PBPK modeling of some of the most persistent PCBs (PCB 99, 101, 118, 153, 149, 170 and 180) and PBDEs (PBDE 47, 99, 100 and 153) was performed for harbour porpoises recently (Weijs *et al*., 2010, 2011, 2012).

In these studies, tissues included were the blubber, brain, kidneys, muscle and liver to cover processes such as storage, elimination through the production of feces and urine and through metabolic biotransformation. Parameters in the models were either taken from the literature or were fitted to the data. Datasets used for the evaluation of the models were from harbour porpoises from the Black Sea and the North Sea (Weijs *et al*., 2010, 2011, 2012). All models run over the entire lifetime (approximately 20 years) of the harbour porpoises.

RESULTS AND APPLICATIONS OF PBPK MODELS IN HARBOUR PORPOISES

Results reveal that the highest and lowest concentrations of the selected PCBs and PBDEs were consistently found in the blubber and brain, respectively. All models show that there is a huge peak in the concentrations of PCBs and PBDEs for animals from birth until the age of 1 year mostly because of the high concentrations of these congeners in milk (Weijs *et al*., 2010, 2011, 2012). After that, concentrations decrease until the age of 3 or 4 years followed by an increase in levels of all modeled congeners until the end of their lives. For some congeners (e.g., PCB 101, PBDE 47), the maximum levels observed overall are the concentrations in calves at the end of lactation. Other congeners reach a maximum in animals at the age of 20 years (e.g., PCB 99, PCB 149, PCB 170, PCB 180, PBDE 99, PBDE 100, PBDE 153) thereby even exceeding the peak concentrations in calves. Reverse dosimetry modeling, thus reconstructing exposure scenarios from a given concentration in the tissues, was also applied using data of PCBs and PBDEs in blubber of North Sea harbour porpoises from 1990–2008. During this

time period of 18 years, all concentrations of PCBs and PBDEs decreased, although not at the same rate. For PBDEs, levels of PBDE 99 and PBDE 100 seemed to decrease the fastest and slowest, respectively (Weijs *et al.*, 2012). For PCBs, levels of PCB 101 and PCB 149 decreased the fastest and slowest, respectively (Weijs *et al.*, 2011). Levels of PCBs in harbour porpoises in 2008 were on average 2.8 times lower than in animals from 1990, whereas PBDEs were on average 2.6 times lower in 2008 compared to 1990. Overall, decreases are going slowly, thereby leaving the harbour porpoises at risk for several years to come.

POTENTIAL APPLICATIONS OF PBPK MODELS IN MARINE MAMMAL TOXICOLOGY

In the pharmaceutical industry, PBPK models are often used to predict and evaluate the PK characteristics of candidate drugs, to understand and compare the fate and disposition of chemicals in animals and humans, to help with the design and the interpretation of toxicological studies, to calculate exposure based safety margins and to compare the bioavailability for different dosage schemes or administration routes (Andersen, 2003; Theil *et al.*, 2003; Khalil and Läer, 2011; Rowland *et al.*, 2011). This particular industry mainly uses typical model organisms such as rodents or rabbits for these applications and can thus control several parameters (e.g., dosage schemes, acute versus chronic exposure) in the experiments. It is not possible to control exposure conditions for wild marine mammals. Nevertheless, some of the applications may still be a possibility in marine mammal toxicology. A better understanding of the fate, kinetics and disposition of pollutants like PCBs and PBDEs has been achieved using PBPK models for harbour porpoises (Weijs *et al.*, 2010, 2011) and other marine mammal species (Hickie *et al.*, 1999, 2005, 2007). Together with *in vitro* toxicity data, these models could be useful to set up safety margins which may consequently be a useful tool for the conservation of a species. Following the example of Verwei *et al.* (2006), a combination of *in vitro* and *in silico* techniques has already been proven to work for the prediction of *in vivo* embryotoxic effect levels.

Currently, on-going research is focusing on the extrapolation of the PBPK models for several PCBs and PBDEs to other new lipophilic compounds with similar properties. This aspect of modeling has been tested already by Wang *et al.* (2000) for dioxins and deserves further attention as it would give the opportunity to deduce the potential for bioaccumulation of an unknown compound in an organism based on previous knowledge of the behavior of similar-looking compounds. However, more information about the bioaccumulation processes of these compounds in mammals is needed before such extrapolations are possible.

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