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Hepatic sequestration of chlordecone and hexafluoroacetone evaluated by pharmacokinetic modeling

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Abstract

Chlordecone (CD) and mirex (M) differ by a single carbonyl group in CD in place of two chlorines in M. Although both compounds are lipophilic, their tissue distributions differ markedly: CD concentrations are highest in liver; M concentrations are highest in fat. We used tissue time course data in rats from our laboratory for CD and M and literature data from monkeys to develop PBPK models to study differences in liver and fat partitioning. The PK model for M had partitioning in tissue without specific hepatic binding. The CD model had partitioning similar to M, and also included liver binding: the maximal binding (B_{max}) and binding affinity constant (Kd) required to describe the rat data were 370 nmol/g liver and 100 nM, respectively. To see if other ketones with electron withdrawing constituents at the alpha carbon were also preferentially distributed to liver, we developed a PBPK description for tissue distribution of hexafluoroacetone (HFA). Compared to acetone, HFA is known to be preferentially sequestered in liver and more slowly excreted unchanged from the body. Acetone is more equally distributed to tissues. HFA distribution was evaluated with a PBPK model that included hepatic binding. B_{max} and Kd were 1.58 μmol/g liver and 301 μM. In summary, liver sequestration of CD and HFA most likely represents relatively high-affinity but reversible binding of activated carbonyls in these compounds (activated by the presence of electron withdrawing substituents on the alpha-carbons) with glutathione and glutathione transferases, that are present at much higher concentrations in liver than in other tissues. Strong, but reversible hemithioketal formation with active sulfhydryls may also be associated with the toxic responses to CD and HFA.

Keywords: PBPK; Chlordecone; Kepone; Mirex; Acetone; Hexafluoroacetone

1. Introduction

Chlordecone (Kepone®, decachlorotetracyclodecanone; $C_{10}Cl_{10}O$; CD), an environmentally persistent chlorinated hydrocarbon pesticide, is no longer produced and sold in the USA (Faroon et al., 1995). In studies of hepatotoxicity, CD potentiated the responses to sublethal doses of carbon tetrachloride and chloroform (Cianflone et al., 1980; Curtis et al., 1979). A physiologically based pharmacodynamic (PBPD)

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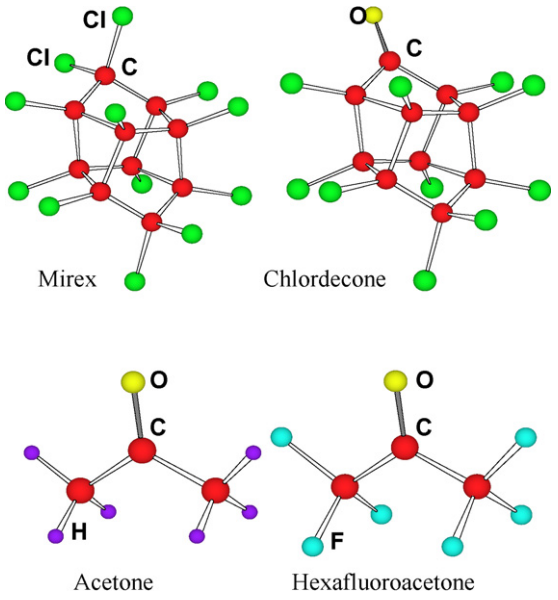

Fig. 1. Structures of mirex, chloredecone, acetone and hexafluoroacetone (C S Chem 3D Pro, version 5.0. Cambridge Soft Corp., 100 Cambridge Park Dr., Cambridge, MS, 02140).

model had previously been used to relate effects of CD on CCl_4 toxicity (El-Masri et al., 1996). This PBPD model did not consider distribution of CD into liver and various other tissues. Our work here was initially intended simply to provide a mechanistic PBPK model for CD distribution that would more accurately account for the tissue distribution of CD (Belfiore et al., 2002).

CD is structurally similar to mirex (M), having one ketone group ($C_{10}Cl_{10}O$) while M ($C_{10}Cl_{12}$) has only chlorine substitutions (Fig. 1). Both compounds are highly lipophilic (log K_{OW} between 5 and 6), and on this basis alone, both should exhibit similar tissue distribution patterns. However, CD is preferentially accumulated in liver rather than fat. Differential effects of blood and hepatic binding on the disposition of the two compounds can readily be included in PBPK models in which partition coefficients reflect the similar lipophilicity of CD and M, with specific binding of CD provided by binding maxima and dissociation binding constants in liver.

The carbonyl group in CD is more nucleophilic (i.e., has a higher partial positive charge) than the carbonyl in ketones lacking alpha-halogen substitutions. Another ketone with alpha-carbon halogen substitution, hexafluoracetone (HFA), also distributes preferentially to liver, and has dose-dependent kinetics (Borzelleca and Lester, 1965; Gillies and Rickard, 1984). Acetone, the water-miscible hydrocarbon analog of HFA, is uniformly distributed among blood, urine and tissues (Kumagai and Matsunaga, 1995; Scholl and Iba, 1997; Singer and Jones, 1997). Pharmacokinetic data describing acetone concentration in blood (Plaa et al., 1982) have already been described by PBPK modeling (Clewell et al., 2001). This current work also extends the acetone PBPK model to HFA by inclusion of tissue specific binding in liver.

For both pairs (CD versus M and HFA versus acetone), the apparently anomalous tissue disposition observed with HFA and CD indicate specific binding in liver. In this paper we develop PK descriptions of CD and HFA that include hepatic binding and assess the binding maxima and affinities consistent with the differential liver uptake of CD and HFA compared to M and acetone. A similar approach had been used to assess hepatic binding of tetrachlorodibenzo-*p*-dioxin (Leung et al., 1988; Andersen et al., 1993). Our results provide evidence of strong reversible binding with sulfhydryl groups in liver as the determinant of hepatic accumulation of CD and HFA.

2. Methods

2.1. Data collection and analysis

A group of studies was conducted in our laboratory with CD and M. Male Sprague–Dawley rats weighing approximately 200 g were obtained from Harlan Sprague–Dawley (Indianapolis, IN). Animals were tagged and acclimated for 4 weeks. Rats were housed at the Laboratory Animal Resources facility, which is accredited by the Association for Assessment and Accreditation of Laboratory Animal Care (AAALAC). Three animals per polycarbonate cage were supplied food (Harlan NIH-07 diet, Madison, WI) and water ad libitum. Lighting was set to a 12 h light/dark cycle. At treatment, average body weight of the rats was 359 g.

Animals were randomized for treatment with either CD or M. Equimolar solutions of each compound were prepared in corn oil so that each animal would receive a dose of 40.0 or 44.48 mg/kg body weight of CD or M, respectively, in approximately 1 ml oral gavage dose. These doses were chosen to correspond to that used by Egle et al. (1978). Rats were returned to normal diet and water and killed 1, 14 or 30 days after dosing.

Rats were anesthetized with inhaled isoflurane. Euthanasia was performed by exsanguination via aortic puncture; blood was collected and placed into a heparinized glass tube. Liver, fat, kidney and muscle specimens were removed from each animal, diced, placed into plastic vials and snap-frozen in liquid nitrogen. Tissue samples were stored at −80 °C and whole blood samples were refrigerated until time of assay.

Analysis of CD and M followed a procedure adapted from that of Blanke et al. (1977). Tissue samples (100–500 mg) were spiked with either 5 μg CD or M internal standard and ground with 3 ml of phosphate-buffered saline (PBS, 0.05 M, pH 7.44).