1-OCTANOL/WATER PARTITION COEFFICIENTS OF 5 PHARMACEUTICALS FROM HUMAN MEDICAL CARE: CARBAMAZEPINE, CLOFIBRIC ACID, DICLOFENAC, IBUPROFEN, AND PROPYPHENAZONE

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Abstract. Laboratory studies were conducted to characterize the 1-octanol/water partition coefficients of pharmaceutically active substances carbamazepine, clofibric acid, diclofenac, ibuprofen, and propyphenazone. Partition coefficients determined by shake flask experiments (OECD guideline 107) varied between log K_{OW} 1.51 for carbamazepine, 2.88 for clofibric acid, 1.90 for diclofenac, 2.48 for ibuprofen, and 2.02 for propyphenazone. Comparison of these values with the literature values revealed rather significant differences for most of the compounds. The partitioning coefficients of the acidic compounds diclofenac and ibuprofen agreed much better with sorption and mobility data from previously conducted experiments, whereas K_{OW} values for carbamazepine were lower and for clofibric acid higher than expected from experiments. Only K_{OW} values for propyphenazone were in the same range as reported in the literature and expected from column experiments.

Keywords: K_{OW}, mobility, drugs, K_{OC}, sorption

1. Introduction

Pharmaceutically active compounds are repeatedly reported in surface water and in groundwater from several places worldwide (Heberer, 2002; Tixier *et al.*, 2003). After application, many pharmaceuticals from human medical care are excreted as their parent substances or water-soluble metabolites (Mutschler *et al.*, 2001) and may enter the surface waters, because they are only partially eliminated in wastewater treatment plants (Ternes, 1998). Some pharmaceuticals may even percolate into the groundwater under special circumstances (Scheytt *et al.*, 1998). Beside flow conditions and degradation, sorption is one of the key factors controlling the input, transport, and transformation of those substances in the aquatic environment and in the subsurface.

The substances tested in the present experiments have been extensively used in human medical care (Heberer, 2002; Scheytt, 2002). The molecular structures of the pharmaceutically active compounds are shown in Figure 1, the physical and

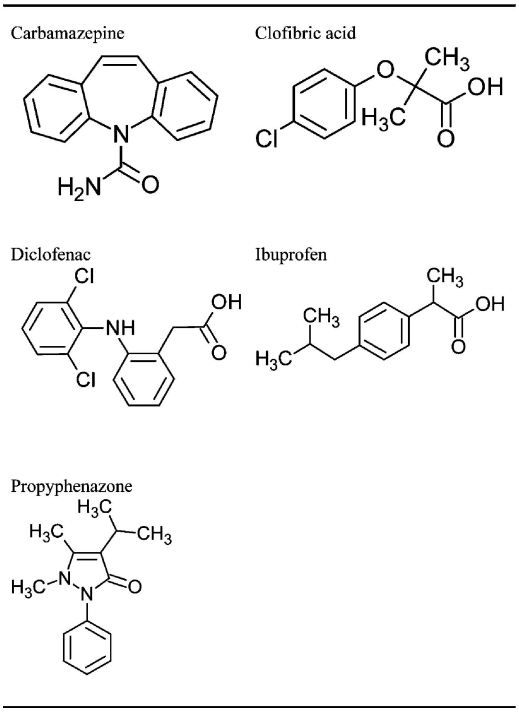

Figure 1. Molecular structures of carbamazepine, clofibric acid, diclofenac, ibuprofen, and propyphenazone.

chemical properties are presented in Table I. Carbamazepine is an anti-epileptical drug used worldwide. This drug has frequently been detected in surface water and occasionally in groundwater. Diclofenac is used in human medical care as an analgesic, antiarthritic, antirheumatic compound, belonging to the group of nonsteroidal anti-inflammatory drugs (Mutschler *et al.*, 2001). Ibuprofen is a nonsteroidal anti-inflammatory, analgesic, and antipyretic drug. It is an important nonprescription drug and used widely (Buser *et al.*, 1999). Propyphenazone is a mild analgesic (pain-killer) pharmaceutical that is normally used in combinatory drugs. Today, it is produced in Japan and Eastern Europe (Holm *et al.*, 1995).

Many times sorption coefficients or environmental risk assessments are based on the octanol/water partitioning coefficients of the respective compounds. This is a reasonable assumption if the affinity of the compound for 1-octanol reflects its affinity for dissolved organic carbon (DOC) or lipids. 1-octanol/water partition coefficients (K_{OW}) are readily available for all above-mentioned pharmaceuticals. However, the interpretation of the results from previously conducted laboratory soil column and batch experiments (Mersmann *et al.*, 2002; Scheytt *et al.*, 2004; Scheytt *et al.*, in press) revealed significant deviations from available literature on 1-octanol/water partitioning coefficients. Therefore, own laboratory

TABLE I
Physical and chemical properties of the test compounds

Compound	Vapor Pressure [mm Hg]	Solubility [mg L^{-1}]	pK_a (20 °C)	log K_{OW}	log K_{OW} (this publication)
Carbamazepine	1.4×10^{-7}[a]	17.66[b]	14.0	2.25[f]	1.51
Clofibric acid	1.13×10^{-4}[a]	582.5[b]	3.2	2.84[k]	2.88
				2.57[l]	
Diclofenac	6.14×10^{-8}[a]	2.37[c]	4.16[j]	4.51[g]	1.90
				4.02[f]	
Ibuprofen	1.86×10^{-4}[a]	21[d]	4.52[j]	3.5[h]	2.48
				3.97[g]	
Propyphenazone	5.2×10^{-6}[a]	2400[e]		2.32[i]	2.02

Temperature is 25 °C, if not otherwise stated; 1 Pa = 7.500617×10^{-3} mm Hg.
[a]Calculated value; from Neely and Blau (1985).
[b]Calculated value; from Meylan *et al.* (1996).
[c]Fini *et al.* (1993).
[d]Yalkowsky and Dannenfelser (1992).
[e]Merck Index (2001).
[f]Syracuse Science Center (2002), calculated value.
[g]Avdeef *et al.* (1998), value determined using a pH-metric technique.
[h]Stuer-Lauridsen *et al.* (2000), calculated value.
[i]Holm *et al.* (1995), determined by HPLC on reversed phase.
[j]Rafols *et al.* (1997).
[k]Henschel *et al.* (1997), calculated value.
[l]Hansch *et al.* (1995), experimental value.

experiments were conducted to investigate the partitioning behavior of the pharmaceuticals.

The objectives of this study were to determine the 1-octanol/water partitioning coefficients for carbamazepine, clofibric acid, diclofenac, ibuprofen, and propyphenazone using laboratory shake flask experiments. The experiments took place at approximately pH 7, a value commonly found in groundwater.

2. Materials and Methods

We conducted laboratory shake flasks experiments according to OECD guideline 107 (OECD, 1995) using 1-octanol from Merck (Darmstadt) and water from Millipore Purification Pak (ultra-pure-water system). All pharmaceuticals were dissolved in 1-octanol to obtain a concentration of 0.5 mg mL^{-1} for the pharmaceutical standards.

In our experiments carbamazepine, clofibric acid, diclofenac, ibuprofen, and propyphenazone were added to individual flasks to obtain different volume ratios

of water and 1-octanol. In all experiments the total amount of liquid (water plus 1-octanol) was 20 mL. To the flasks containing the solvent 1-octanol, the pharmaceuticals were added directly in a volume of 500 μL. Afterwards the flasks were filled up with water to obtain the desired 1-octanol/water ratio of either 6 mL 1-octanol plus 14 mL water (1-octanol ratio of 1:31/3) or 4 mL 1-octanol plus 16 mL water (1-octanol ratio of 1:5). A control for each compound and each ratio was made as specified by OECD Guideline 107. All samples were shaken at 20 rotations/minute for 105 min and centrifuged afterwards at 2000 rotations/minute. All experiments were conducted at ambient/laboratory temperatures of 21 °C ± 1 °C.

2.1. CHEMICAL ANALYSIS

For the analysis of the pharmaceutical compounds the water was cautiously separated from the shake flaks. The pharmaceutical compounds were determined in the water phase only. The concentrations of pharmaceuticals in 1-octanol were calculated by subtraction of the total amount and the amount in the water phase. The water sample was then adjusted to a pH of 2 and afterwards extracted by solid-phase extraction (SPE) using a non-endcapped reversed phase adsorbent (RP-C18 Bakerbond Polar Plus). Before extraction, the samples were spiked with 100 ng of 4-chlorophenoxy-butyric acid (100 μL of a 1 ng μL^{-1} solution in methanol) or dihydrocarbamazepine used as surrogate standards for analytical quality control (Reddersen and Heberer, 2003). Then the acidic analytes clofibric acid, diclofenac, ibuprofen, and propyphenazone and the surrogate standard were derivatized with pentafluorobenzyl bromide (2% in toluene), carbamazepine was derivatized with N-tert-Butyldimethylsilyl-N-methyltrifluoroacetamide (MTBSTFA) making them amendable to gas chromatographic separation using 5%-phenylmethylpolysiloxane column (HP5MS, 30 m × 0.25 mm inner diameter, 0.25 μm film thickness) from Agilent Technologies (Waldbronn, Germany) (Reddersen and Heberer, 2003). 2 μL of the sample extracts (100 μL for each sample) were analyzed by capillary gas chromatography-mass spectrometry (GC-MS) with selected ion monitoring (SIM) using an HP 5890 gas chromatograph and an HP 5970B quadrupol mass spectrometer from Agilent Technologies (Waldbronn, Germany). The solvents used were Methanol (PESTANAL®, solvent for residue analysis, ≥99.9% (GC)), Acetone (PESTANAL®, solvent for residue analysis, ≥99.8% (GC)), and Toluene (PESTANAL®, solvent for residue analysis, ≥99.7% (GC)) ordered from Sigma-Aldrich, Steinheim (Germany). Depending on the sample volume (100 to 200 mL), the limits of determination were between 1 and 10 ng/L, and the limits of quantitation were between 5 and 25 ng/L. The analytical recoveries range between 80% and 120%. For further analytical details refer to Heberer *et al.* (1998) or Reddersen and Heberer (2003).

2.2. PARTITIONING COEFFICIENTS

The distribution of neutral substances between the aqueous phase and a hydrophobic phase (1-octanol) is defined by the ratio between the concentration of the substance in *n*-octanol and in water.

$$K_{\mathrm{OW}} = \frac{C_{n-\mathrm{Octanol}}}{C_{\mathrm{Water}}} \tag{1}$$

where K_{OW} is the *n*-octanol/water partition coefficient, $C_{n\text{-Octanol}}$ the concentration of a chemical in *n*-octanol [mg L^{-1}] and C_{Water} the concentration of the compound in water [mg L^{-1}].

For compounds with dissociable groups, like in the case of the carboxylic acids clofibric acid, diclofenac, and ibuprofen the general equation leads to:

$$\mathrm{RH} \Leftrightarrow R^- + H^+ \tag{2}$$

where RH refers to the neutral compound and R^- refers to the negatively charged compound. On the basis of this equation the following equations can be derived:

$$K_{\mathrm{a}} = \frac{[R^-]_{\mathrm{Water}} \cdot [H^+]_{\mathrm{Water}}}{[\mathrm{RH}]_{\mathrm{Water}}} \tag{3}$$

$$D_{\mathrm{OW}} = \frac{[\mathrm{RH}]_{\mathrm{Octanol}}[R^-]_{\mathrm{Octanol}}}{[\mathrm{RH}]_{\mathrm{Water}} + [R^-]_{\mathrm{Water}}} \tag{4}$$

where K_{a} is the acid dissociation constant and D_{OW} the apparent partitioning coefficient. The following pH-dependent partitioning coefficient can be derived analogously to the Henderson Hasselbalch equation (Lützhoft *et al.*, 2000a):

$$D_{\mathrm{OW}} = \frac{K_{\mathrm{OW}}}{1 + 10^{\mathrm{pH-pKa}}} \tag{5}$$

$$D_{\mathrm{OW}} = \frac{K_{\mathrm{OW}} + D_{\mathrm{R}} \cdot 10^{\mathrm{pH-pKa}}}{1 + 10^{\mathrm{pH-pKa}}} \tag{6}$$

K_{OW} is the partitioning coefficient for the neutral species and D_{R}^- the distribution coefficient for the negatively charged species. Hence, Equation (5) only considers distribution of neutral species whereas Equation (6) takes the neutral as well as the negatively charged species into account. However, Lützhoft *et al.* (2000b) found that Equation (5) did not fit their data on the distribution of oxolinic acid significantly better than Equation (6). The above equations do not take distribution of ion pairs into account, but only distribution of the neutral and the charged species.

3. Results and Discussion

For best comparison with other data we chose the shake flask method according to OECD guideline 107 (OECD, 1995) rather than other approaches (e.g. Hanna *et al.*, 1998). In all cases the samples and their controls yielded good agreement. The samples and the control samples were used to calculate the mean value for the partitioning coefficients. For carbamazepine and propyphenazone the 1-octanol/water partitioning coefficient was 1.51 and 2.02, respectively. Clofibric acid showed a value of 2.88, whereas diclofenac and ibuprofen had lower values of 1.90 and 2.48, respectively. In the case of carbamazepine with a pKa of 14 this partitioning coefficient is the log K_{OW} value, because this compound is present in its non-dissociated form in the pH range of the experiment. For the other compounds these values are the apparent partitioning coefficients log D_{OW}, as these compounds are present in their non-dissociated forms as well as anions. Due to the acidification of the samples during GS/MS analysis, not only the neutral compounds but also the previously ionized form of the compounds were included in the measured value.

Reported K_{OW} values and the coefficients presented in this manuscript vary significantly for carbamazepine, diclofenac, and ibuprofen (Table I). The values for clofibric acid and propyphenazone were in the same range as reported K_{OW} values. However, only the K_{OW} value for propyphenazone was also determined by a shake flask experiment (Holm *et al.*, 1995). For diclofenac and ibuprofen, the partitioning coefficients presented here agree much better with the results from previously conducted laboratory sorption and transport experiments (Table II), whereas for

TABLE II

Comparison of log K_{OC}-coefficients based on literature K_{OW} values and based on log K_{OW} values from this publication

	log K_{OC} values based on equations by Karickhoff[a]		log K_{OC} values from experiments	
Compound	Using literature K_{OW}	Using K_{OW} from this publication	Batch experiments[b]	Column experiments[c]
Carbamazepine	2.04	1.30	2.00–3.42	2.24–2.27
Clofibric acid	2.36–2.63	2.67		0.9–1.36
Diclofenac	3.81–4.30	1.69	2.45–3.74	1.90–2.10
Ibuprofen	3.29–3.76	2.27	2.14–2.21	2.19*
Propyphenazone	2.11	1.81		1.70–1.98

*Based on interpolation.

[a]Karickhoff *et al.* (1979): log $K_{OC} = 1.0 \log K_{OW} - 0.21$.

[b]Scheytt *et al.* (in press).

[c]Based on retardation factors from breakthrough curves of soil column experiments under water saturated conditions; see Mersmann *et al.* (2002) and Scheytt *et al.* (2004).

clofibric acid a much lower partitioning coefficient was expected based on the column and batch experiments. In the case of clofibric acid neither the values from own experiments nor the literature values approached the values from the batch and sorption experiments. For carbamazepine finally, the partitioning coefficient from the shake flask experiment was significantly lower than expected from the column and batch experiments.

K_{OC} values are often estimated based on the K_{OW} values. In the case of pharmaceuticals the sorption, especially of diclofenac and ibuprofen, would have been overestimated using literature partitioning coefficients, while our results from the shake flaks experiments agreed quite well with the batch and column experiments. In the case of clofibric acid the column and batch experiments showed very low sorption despite the relatively high partitioning coefficients found in our shake flask experiments and despite the high log K_{OW} values reported in literature. The five pharmaceutically active substances may be ranked in order of increasing 1-octanol/water partitioning coefficients as follow:

Carbamazepine > Diclofenac > Propyphenazone > Ibuprofen > Clofibric acid.

The results show that the prediction of sorption behavior or the environmental risk assessment based on partitioning coefficients of ionic and neutral xenobiotics may lead to wrong results. Likewise, correlation equations predicting K_{OC} based on K_{OW} should be used cautiously because they could be misleading regarding the mobility of those substances.

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