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Lipophilicity of zwitterions and related species: A new insight

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ABSTRACT

The experimental determination of microscopic partition coefficients for protonation isomers is elaborated for the first time, and applied for niflumic acid, an ampholytic, mainly zwitterionic drug for pains in joints and muscles. The acid–base microequilibria of niflumic acid are also characterized by NMR-pH and deductive methods using auxiliary compounds of reduced complexity. The results show that 16 times as many zwitterionic than non-charged microspecies exist in aqueous solution. Partition of the individual microspecies was mimicked by model compounds of the closest possible similarity, then correction factors were also determined and introduced. Thus the long-awaited intrinsic partition coefficients of the non-charged vs. zwitterionic species could be calculated. The non-charged microspecies is 390 times as lipophilic as its zwitterionic protonation isomer. The microscopic partition coefficients are also in line with the experimentally determined distribution coefficients. These results make evident that contribution of the zwitterionic microspecies to the overall lipophilicity is not negligible, especially at the isoelectric pH region of the compound.

1. Introduction

Lipophilicity is a molecular property of immense importance in medicinal chemistry and biochemistry. The ability of drugs to diffuse passively through biological membranes is influenced to a large extent by their lipophilicity (Fujita, 1990). In addition, lipophilicity is an improving tool to unravel biologically relevant intramolecular interactions and intermolecular forces of recognition (Testa et al., 1996). Lipophilicity, by definition, is a measure of the concentration ratio of a solute present in a single electrical state and in equilibrium between two immiscible solvents, expressed as the logarithm of the partition coefficient (log P). The organic solvent of choice is usually octanol. When more than one electrical species are present in solution, the observed ratio of concentrations is the distribution coefficient (D), which takes into account the intrinsic lipophilicity of the various electrical species present (p_i), and their mole fractions in the aqueous phase (x_i).

$$D = \Sigma x_i p_i \quad (1)$$

The lipophilicity profile (the variation of log D as a function of the aqueous pH) of a drug is a *sine qua non* to understand its pharmacokinetic, toxicokinetic and even pharmacodynamic properties (Pagliara et al., 1997).

Molecules of one acidic and one basic sites exist in solutions in four microscopic protonation forms (microspecies), including the cationic, zwitterionic, non-charged and anionic ones (Bjerrum, 1923; Noszál, 1990). Such molecules of pharmaceutical and biochemical importance are morphine, amino acids, oxicams, fluoroquinolones, niflumic acid, and numerous others.

Determination of the concentration and the concomitant acid–base microscopic equilibria in aqueous solutions became a well-established principle in terms of methodology and interpretation for the characterization of small bio- and drug molecules (Noszál, 1990).

Analogously, the idea of species-specific partition coefficient also occurred. One side-branch of such parameters, the conformation-specific partition coefficient was introduced early this century (Noszál and Kraszni, 2002), and experimental determinations were subsequently reported (Kraszni et al., 2003).

The necessity of microspecies-specific partition coefficient, the most important related parameter has long been evident (Taylor and Cruickshank, 1985; Takács-Novák et al., 1992), at least for some of the partitioning microspecies. Abraham et al. (1997) developed an equation to predict the blood-brain distribution coefficient of a drug based on the microscopic partition coefficient of the non-charged species, its hydrogen-bond acidity and its hydrogen-bond basicity. These models assumed, however, that only the non-charged species partitions into the organic phase, although the contribution of charged species to the distribution coefficient may not be insignificant in a number of cases (Irwin et al., 1988; Pagliara et al., 1997; Takács-Novák and Szász, 1999; Mandić and Gabelica, 2006). Later on these models were refined by incorporating the microscopic partition coefficient of the zwitterionic protonation isomer as well (Takács-Novák et al., 1994), but no determination has so far been reported for the individual

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microscopic partition coefficient of the zwitterion and its non-charged protonation isomer.

Rather, computational programs were published for the prediction of microscopic partition coefficients (Csizmadia et al., 1997; Marvin program by Chemaxon), lacking the control of experimental determination of these parameters.

In order to gain species-specific insight into the partition micro-equilibria of amphoteric drugs, we elaborated a method in which the partition properties of the parent compound and its synthetic derivatives, closely mimicking each microspecies, were studied. Our parent compound of choice was niflumic acid, an important non-steroidal anti-inflammatory drug (Reynolds, 1989), which has four microscopic states of protonation and an inherently large overall lipophilicity. Here we report the aqueous acid–base micro-equilibrium analysis of niflumic acid, and – for the first time for any compound – experimental microscopic partition coefficients for protonation isomers.

2. Material and methods

2.1. Materials

Niflumic acid (**1**) was generously supplied by its manufacturer (Gedeon Richter Chemical Works, Budapest). Methanol of HPLC grade was purchased from Aldrich, whereas octanol from Fluka. All other reagents were of analytical grade (Reanal). All solutions were prepared from freshly boiled distilled water.

2.2. Synthesis of niflumic acid derivatives

While the synthesis of niflumic acid amide (**2**) was feasible starting from the parent compound, the synthesis of *N*-methyl niflumic acid (**5**) required first the methyl ester (**3**) of the parent compound and its *N*-methylated derivative (**4**) as well. The structure of all the compounds was confirmed by 1D (^{1}H, ^{13}C) and 2D NMR (HSQC, HMBC) spectra in $CDCl_3$ on a 600 MHz Varian Inova Unity spectrometer. Chemical shifts are reported in parts per million (ppm) downfield from the $CHCl_3$ residual line (δ = 7.27 ppm) and were assigned as singlet (s), doublet (d), or doublet of doublets (dd). Analytical thin layer chromatography (TLC) was performed on glass plates (Merck Kieselgel 60 F254). Visualization was accomplished by UV light (254 nm). The structure of these molecules can be seen in Fig. 1.

2.2.1. 2-[3-(trifluoromethyl)anilino]nicotinamide (2) ($C_{13}H_{10}F_3N_3O$, 281.237)

Thionyl chloride (5 ml) was slowly added to 1 g (3.54 mmol) of niflumic acid and the suspension was stirred for 5 min. The mixture was heated to reflux at 60 °C for 180 min until a clear solution was obtained. The excess of thionyl chloride was evaporated in vacuo. To the residue anhydrous benzene (10 ml) was added and evaporated. The remaining solid was dissolved in dry tetrahydrofuran (10 ml) and 5 ml of 25% aqueous ammonium hydroxide solution was added while keeping the temperature below 20 °C. The reaction mixture was stirred at RT for 3 h. Tetrahydrofuran was removed in vacuo and the water solution was extracted with ethyl acetate (3 × 10 ml). The combined organic phase was washed with water (2 × 10 ml) and dried over Na_2SO_4. The solvent was removed in vacuo, and the solid residue was crystallized from isopropanol. Yield: 0.93 g (91%). Mp. 287–289 °C (Mp. 284–285 °C reported by Criddle et al. (2002)). ^{1}H NMR ($CDCl_3$): 8.38 (d, 1H), 8.09 (s, 1H), 7.82 (d, 1H), 7.75 (d, 1H), 7.40 (dd, 1H), 7.25 (d, 1H), 6.77 (dd, 1H).

2.2.2. Methyl 2-{[3-(trifluoromethyl)phenyl]amino}nicotinate (3) ($C_{14}H_{11}F_3N_2O_2$, 296.25)

Thionyl chloride (5 ml) was slowly added to 1 g (3.54 mmol) niflumic acid under stirring and the suspension was stirred for 5 min. The mixture was heated to reflux at 60 °C for 180 min till a clear solution was obtained. The excess of thionyl chloride was evaporated under vacuum. To the residue anhydrous benzene (10 ml) was added and evaporated. The remaining solid was dissolved in methanol (10 ml) containing 0.5 ml (3.54 mmol) triethylamine keeping the temperature at about 20 °C. The mixture was left to stand for 5 h. The solvent was evaporated under vacuum. To the residue 20 ml chloroform was added and extracted with water (2 × 10 ml), 10 ml of 5% aqueous sodium hydrogen carbonate solution and water (10 ml), dried over Na_2SO_4, filtered and concentrated under vacuum to remove the solvent. The residue was crystallized from methanol. Yield: 0.96 g (92%). Mp. 74–75 °C (Mp. 72–74 °C reported by Criddle et al. (2002)). ^{1}H NMR ($CDCl_3$): 8.41 (d, 1H), 8.27 (d, 1H), 8.10 (s, 1H), 7.88 (d, 1H), 7.43 (dd, 1H), 7.28 (d, 1H), 6.79 (dd, 1H), 3.95 (s, 3H).

2.2.3. N-methyl 2-{[3′-(trifluoromethyl)phenyl]amino}-3-methoxycarbonyl-pyridinium methosulfate (4) ($C_{15}H_{14}F_3N_2O_2 \cdot CH_3SO_4^-$)

To a solution of **(3)** (1 g, 3.37 mmol) in dry xylene (8 ml), dimethyl sulfate (1.02 g, 6.74 mmol) was added at RT. The reaction mixture was heated to reflux and stirred at 140 °C for 8 h. The reaction mixture was cooled to RT and the precipitated crystals were filtered and washed with ether (2 × 10 ml). The product was crystallized from a methanol–ether mixture. Yield: 1.12 g (78%). Mp. 178–180 °C.

2.2.4. N-methyl 2-{[3′-(trifluoromethyl)phenyl]amino}-pyridinium-3-carboxylate (5) ($C_{14}H_{11}F_3N_2O_2$, 296.25)

To the suspension of 1.27 g (3 mmol) **(4)** in dioxane (10 ml), 40% aqueous potassium hydroxide solution (5 ml) was added and the mixture was heated in water bath at 80 °C for 1 h. The suspension

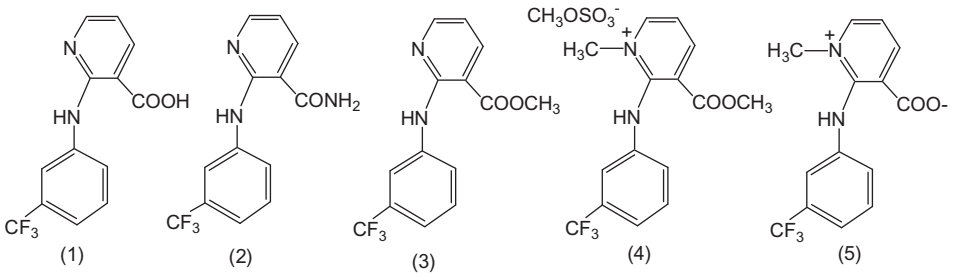

Fig. 1. Niflumic acid derivatives. (1): 2-[3-(trifluoromethyl)anilino]nicotinic acid (niflumic acid); (2): 2-[3-(trifluoromethyl)anilino]nicotinamide; (3): methyl 2-{[3-(trifluoromethyl)phenyl]amino}nicotinate; (4): N-methyl 2-{[3'-(trifluoromethyl)phenyl]amino}-3-metoxycarbonylpyridinium methosulfate; (5): N-methyl 2-{[3'-(trifluoromethyl)phenyl]amino}-pyridinium-3-carboxylate.

was slowly dissolved and the resulting dark yellow precipitate separated from the mixture. The reaction mixture was neutralized by dropwise addition of 36% hydrochloric acid while keeping the temperature under 10 °C. The separated solid was collected and washed with cold water. The product was crystallized from methanol. Yield: 0.48 g (54%). Mp. 238–241 °C. ^{1}H NMR (600 MHz, $CDCl_3$): 1H NMR ($CDCl_3$): 8.81 (d, 1H), 7.51 (dd, 1H), 7.48 (s, 1H), 7.46 (d, 1H), 7.26 (d, 1H), 7.23 (d, 1H), 6.87 (dd, 1H), 3.43 (s, 3H).

2.3. ^{1}H NMR measurements

Titrations were carried out on a 600 MHz Varian VNMRS spectrometer. All solutions contained 5% D_2O by volume, this deuterium concentration is enough for the spectrometer lock system and does not necessitate an adjustment in the pH scale. 0.5 mM DSS (2,2-dimethyl-2-silapentane-5-sulfonate) was used as the reference. ^{1}H NMR spectra were recorded at 23.5 °C, the resonance of the solvents methanol and H_2O was suppressed by WET. The pH of the partly aqueous solutions was measured by a Radiometer pHC2406 combined pH electrode and a Radiometer pHM93 reference pH meter, calibrated by aqueous NBS standard buffer solutions, and was adjusted using the δ term for each mixture (Castells et al., 2003). The pH of the solutions ($2.0 \cdot 10^{-3}$ M for the solute) was adjusted by mixing basic (containing 0.1 M NaOH) and acidic solutions (containing 0.1 M $HClO_4$) in appropriate ratios. Constant ionic strength of 0.15 M was provided by the presence of $NaClO_4$. The NMR spectra were evaluated by MestRe-C 2.3a software, and multiple fitting of the protonation constants were performed with Origin 8. The aqueous protonation constant of the poorly soluble niflumic amide was determined by the Yasuda-Shedlovsky procedure, using apparent protonation constants determined in methanol–water solutions of various proportions (Mazák et al., 1999).

2.4. Partition coefficient measurements by the stir-flask method

The distribution coefficients were calculated from the absorbance of the molecules in the aqueous phase before and after partitioning at several octanol/water phase ratios as previously reported (Mazák et al., 2003). For the pH control phosphate, citrate and MES (4-morpholineethanesulfonic acid) buffers were used with an ionic strength of 0.15 M.

3. Results

3.1. Acid–base equilibria

Niflumic acid in its most basic form bears one negative charge, the addition of one proton will convert this microspecies into a zwitterionic or a non-charged microspecies. The subsequent addition of yet another proton will result in a single, cationic microspecies. The protonation scheme of niflumic acid is depicted in Fig. 2.

Relationships between micro- and macroscopic protonation (proton association) constants have been known since Bjerrum's pioneer work (Bjerrum, 1923), and can be written for niflumic acid as follows:

$$K_1 = k^{\mathrm{N}} + k^{\mathrm{O}} \tag{2}$$

$$K_1 K_2 = k^{\mathrm{N}} k^{\mathrm{O}}_{\mathrm{N}} = k^{\mathrm{O}} k^{\mathrm{N}}_{\mathrm{O}} \tag{3}$$

where K_1 and K_2 are the stepwise macroconstants, k^{N}, k^{O}, $k^{\mathrm{O}}_{\mathrm{N}}$, $k^{\mathrm{N}}_{\mathrm{O}}$ are the microconstants, indices O and N designate oxygen and nitrogen atoms, respectively. Superscripts of the microscopic protonation constants indicate the group protonating in the given microequilibrium protonation process, whereas the subscript (if any) stands for the group holding proton during the process.

Due to the very poor water solubility of niflumic acid, the aqueous log K values were determined from the apparent protonation constants in methanol–water mixtures of various proportions by extrapolation to zero methanol content using the Yasuda-Shedlovsky procedure (Takács-Novák et al., 1994). We used their reported, highly reliable values ($\log K_1 = 4.44$ and $\log K_2 = 2.26$) in the determination of microconstants.

We chose the deductive method for the determination of microconstants, which requires an appropriate model compound to mimic the minor species (Noszál, 1990; Mazák et al., 1999; Mazák et al., 2009). It has been shown that electronic (withdrawing or sending) effects of a carboxylic, –COOH group (strictly not a carboxylate, $-COO^-$), and an amide, $-CONH_2$ group are virtually identical on the adjoining moieties (Noszál and Sándor, 1989; Boros et al., 2007). We synthesized the amide derivative of niflumic acid, determined its protonation constant by the Yasuda-Shedlovsky procedure, and introduced it into $k^{\mathrm{N}}_{\mathrm{O}}$ of niflumic acid. This model compound belongs to the minor protonation pathway, providing thus reliable conditions for the accurate calculation of the microconstants (Noszál, 1990).

The microscopic constants determined for this molecule can be seen in Table 1, that also contains the pH-independent k_z parameter ($k_z = k^{\mathrm{N}}/k^{\mathrm{O}}$, the concentration ratio of the zwitterionic to the non-charged microspecies).

3.2. Lipophilicity

The temperature dependency of the solubility of niflumic acid in buffers with pH 2.0 and 7.4, in *n*-octanol and in *n*-hexane were recently measured. The thermodynamic functions of solubility, solvation and transfer processes were also deduced (Perlovich et al., 2007).

The pH-distribution profile of niflumic acid was already investigated and some microscopic partition coefficients were also determined (Takács-Novák et al., 1994). The approximately parabolic pH-distribution profile of niflumic acid in the octanol/water system showed a maximum distribution coefficient (D) around the isoelectric point. Potentiometric titrations in the presence of octanol at 0.15 M ionic strength resulted in a log P of 3.88 for the neutral macrospecies, containing the lipophilicity contributions of both the non-charged and the zwitterionic microspecies. Assuming the zwitterion does not partition into octanol, the authors reported a partition coefficient for the non-charged microspecies ($\log p^{\mathrm{N}} = 5.14$).

While it is plausible that the microscopic partition coefficient of the non-charged microspecies exceeds those of the other microspecies by several orders of magnitude (Avdeef, 1996; Bouchard et al., 2002), no direct experimental evidence excludes the partitioning of the zwitterionic microspecies. Also, no determination has so far been reported for the individual microscopic partition coefficient of the zwitterion (p^{Z}) and its non-charged protonation isomer (p^{N}). The distribution coefficient measured at the isoelectric point by any previous methods characterizes the partition behavior of all the microspecies collectively.

Eq. (1) for niflumic acid gets the form as follows:

$$D_{(\mathrm{pH})} = x_{\mathrm{A}} p^{\mathrm{A}} + x_{\mathrm{N}} p^{\mathrm{N}} + x_{\mathrm{Z}} p^{\mathrm{Z}} + x_{\mathrm{C}} p^{\mathrm{C}} \tag{4}$$

where x_{i} stands for the aqueous mole fractions of the anionic (A), non-charged (N), zwitterionic (Z) and cationic (C) species

The aqueous mole fractions include protonation microconstants and acidity values, expressing thus the pH-dependence of the D distribution coefficient. For example,

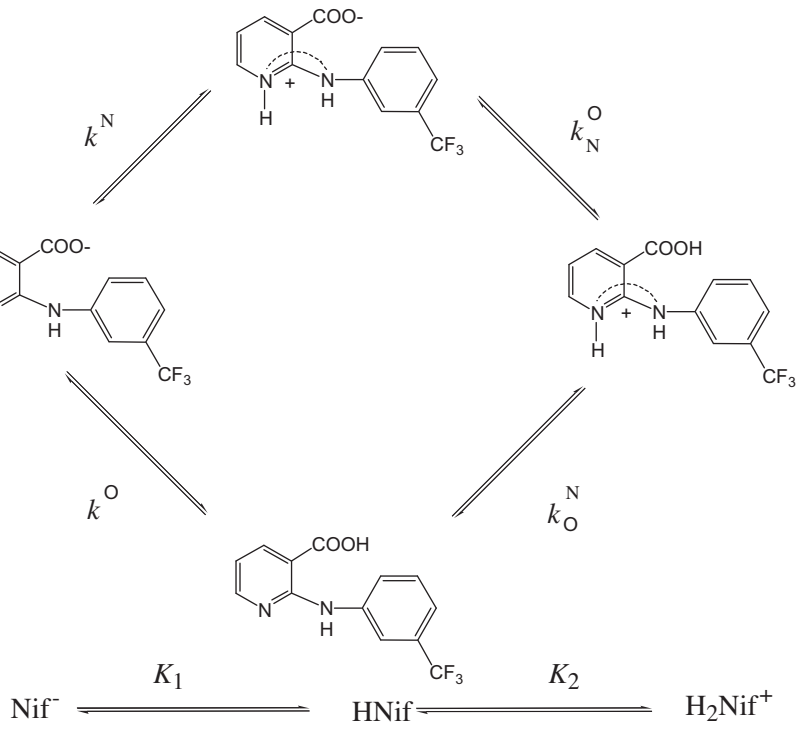

Fig. 2. The protonation equilibria of niflumic acid in terms of macroscopic (K_1, K_2) and microscopic (k^N, k^O, k^O_N, k^N_O) protonation constants.

Table 1
Protonation microconstants of niflumic acid and comparison with literature data (the standard deviation of the measurements is in parenthesis). The ionic strength is 0.15 M.

log k^O	log k^N	log k^O_N	log k^N_O	log k_z	Reference
3.21	4.41	2.29	3.49 (0.03)	1.20	This study
3.18	4.42	2.28	3.52	1.24 (0.01)	Takács-Novák et al. (1994)
2.92	4.86	2.29	4.23	1.94 (0.03)	Takács-Novák and Tam (2000)

$$x_A = \frac{1}{1 + (k^N + k^O)[H^+] + k^N k^O_N [H^+]^2} \quad (5)$$

$$x_Z = \frac{k^N [H^+]}{1 + (k^N + k^O)[H^+] + k^N k^O_N [H^+]^2} \quad (6)$$

The lipophilicity of the cationic and anionic species depends on the type and concentration of the background electrolyte that contributes to the establishment of potential difference between the two immiscible phases (Johnson and Westall, 1990; Escher and Schwarzenbach, 1996; Takács-Novák and Szász, 1999; Reymond et al., 2001; Bouchard et al., 2001).

The knowledge of each microscopic partition coefficient is essential for a thorough understanding of the pharmacokinetic and pharmacodynamic properties of a drug. This is especially important for drugs acting in the central nervous system, a particularly lipophilic medium. Furthermore, the p^N/p^Z ratio of the non-charged vs. zwitterionic partition coefficients is an important structural parameter to gain insight into intramolecular effects (Pagliara et al., 1997; Caron et al., 1999).

In order to characterize the partition microequilibria of niflumic acid, we decided to use the deductive method, namely mimicking the partition behavior of its protonation isomers with appropriate model compounds. The amide of niflumic acid is isoelectronic, isocoulombic and largely isosteric with the non-charged microspecies. We also introduce a correction factor to further improve its mimicking capacity, as it is known from literature data (Takács-Novák and Avdeef, 1996) and theoretical computations with the Marvin program, that carboxylic acids are more lipophilic than their amides. For the zwitterionic microspecies the *N*-methyl derivative of niflumic acid is certainly the best choice, especially if the effect of methylation at the cationic nitrogen can be taken into correction. The p^Z of piroxicam was characterized using a similar strategy by measuring the partition coefficient of its *N*-methyl derivative, but without any correction (Takács-Novák et al., 1995).

We also intended to secure a medium where all the three compounds, niflumic acid, its *N*-methyl derivative and niflumic acid amide are in a uniform state of ionization. Alkaline pH values were ruled out for several reasons: the *N*-methyl derivative undergoes a tautomeric conversion above pH 7, the aqueous solubility of niflumic acid amide is extremely poor, not to mention the charge differences between the parent compound and the mimicking ones. Thus we determined the lipophilicities in highly acidic solutions where all the three compounds exist overwhelmingly in their cationic form. We chose a standardized HCl solution of 0.15 M, as the majority of log *K* and log *P* values are determined at this physiological ionic strength. The results are shown in Table 2.

Due to its low water solubility, the partition coefficient of the neutral form of the amide was calculated from its distribution coefficients in various acidic solutions, and resulted in a log *P* value of 3.75 (±0.03). The predicted value of this constant by the Marvin program is 3.82, which shows a remarkably good agreement.

The water solubility of *N*-methyl niflumic acid allowed its log *D* determination in weakly acidic solutions up to pH 7, where the contribution of the zwitterionic form dominates (Table 3). The

Table 2
Log *D* values of the investigated compounds in the octanol/water system at pH 0.82 (I = 0.15 M). The standard deviation of the measurements is in parentheses.

Compound	log *D*
Niflumic acid	2.02 (0.01)
Niflumic amide	1.08 (0.02)
N-methyl niflumic acid	−0.58 (0.03)

Table 3
Log *D* values of *N*-methyl niflumic acid in the octanol/water system at various pH values (I = 0.15 M). The standard deviation of the measurements is in parentheses.

pH	Buffer	log *D*	
3.55	citrate	−0.49	(0.01)
5.03	citrate	−0.46	(0.02)
6.27	MES	−0.55	(0.03)
6.71	phosphate	−0.51	(0.02)

log *D* value is fairly constant in the different buffer systems of identical ionic strengths. Thus, the log *P* value of the zwitterionic form of *N*-methyl niflumic acid can be regarded equal to −0.50 (±0.04), largely irrespective of the identity of the counter ions. The predicted value of this constant by the Marvin program is −0.01, with less good agreement, due to computation difficulties in handling permanently charged centers (Koufopoulou et al., 2006).

4. Discussion

4.1. Acid–base equilibria

Our results in Table 1 show that there are approximately 16 times as many zwitterionic microspecies than non-charged ones, irrespective of the pH of the solution. Our deductive method confirms the validity of the k_z determination by the method reported by Takács-Novák et al. (1994).

The determination of microconstants allows the construction of a microspecies distribution diagram (Fig. 3). In the pH range 2.30–4.40 niflumic acid mainly exists with mole fraction >0.50 in zwitterionic form. At duodenal pH (4.60) the zwitterion mole percent is still 38.1%, whereas that of the neutral form is 2.4%. At higher pH values, the anionic form predominates, its mole percent in the blood plasma (pH 7.40) reaches 99.9%. The cationic form dominates at highly acidic pH values (pH < 2.30) that can occur in the stomach.

4.2. Lipophilicity

Based on our data in Table 2, the apparent lipophilicity of niflumic acid is higher than its amide or *N*-methyl derivative at that pH and ionic strength, the differences are 0.94 and 2.60 in log *D* units, respectively.

Using the experimentally determined partition coefficients of the model compounds, and taking into account their differences as correction factors, we calculated the logarithm of the microscopic partition coefficients (log *p*) of all the niflumic acid protonation isomers. For the non-charged form log p^N is 4.69 = 3.75 (log *P* of the amide) + 0.94 (correction factor for the acid); whereas for the zwitterionic form log p^Z can be calculated as −0.50 (log *P* of the *N*-methyl derivative) + 2.60 (correction factor for the acid) = 2.10.

The lipophilicity of the anionic form was measured in a standardized NaOH solution of 0.15 M and resulted in a log p^A value of 1.44. Table 4 shows all the experimentally determined microscopic partition coefficients of niflumic acid alongside the predicted values by the Marvin program.

The log (p^N/p^Z) value is 2.59 in this study, thus the non-charged microspecies is only around 390 times as lipophilic as its zwitterionic counterpart. The agreement between the experimentally determined and predicted values is remarkably good. Marvin predicted an even smaller difference between the lipophilicity of the protonation isomers, due to the difficulty of handling permanently charged centers in the computation. Our experimental method delivers a result much closer to the predicted value of the non-charged species, than the reported value of 5.14 by Takács-Novák et al. (1994), where the partition of the non-charged species was taken into account only.

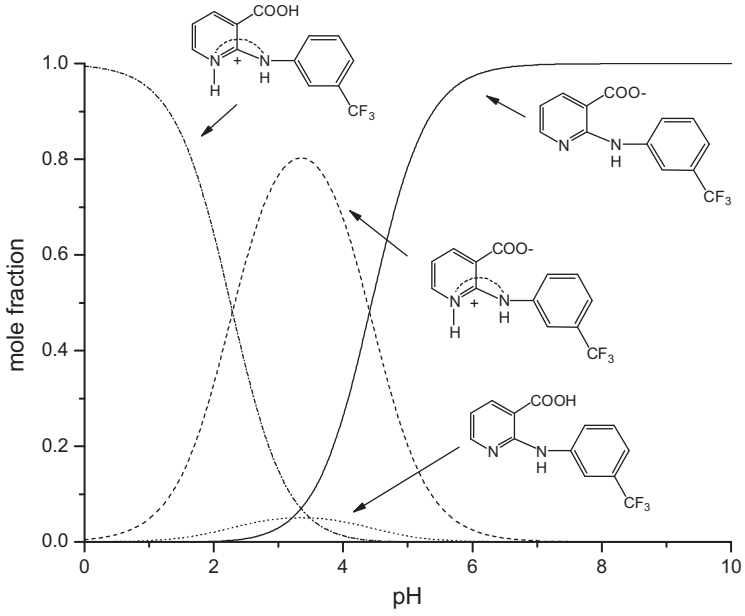

Fig. 3. Distribution of the niflumic acid microspecies in aqueous solution.

Table 4
The logarithm of microscopic partition coefficients of niflumic acid microspecies in the octanol/water system at 0.15 M ionic strength.

	Experimental log p	Predicted log p
Non-charged	4.69	4.63
Zwitterionic	2.10	2.95
Anionic	1.44	1.24

Using the mole fractions and microscopic partition coefficients of the non-charged and zwitterionic species, their pH-independent contribution ratio to the distribution coefficient can also be quantified. This $(x_N\, p^N)/(x_Z\, p^Z)$ value is only 24.5, which shows that for niflumic acid (and possibly many compounds of medicinal interest) the contribution of the zwitterionic microspecies can not be neglected during the calculation of the microscopic partition coefficient of the non-charged, most lipophilic species.

Our method for experimentally quantifying the lipophilicity difference between the non-charged and zwitterionic microspecies can be extended to other compounds of medicinal interest, such studies are in progress in our department. Zwitterionic compounds with unexpectedly high lipophilicity include morphine-6-glucoronide (Stain et al., 1995), where a hypothesis has been proposed for the existence of conformers with intramolecular hydrogen bonds displaying increased apparent lipophilicity in a lipid-like environment (Gaillard et al., 1995). The sedating side-effects of certain zwitterionic antihistamines, like acrivastine and cetirizine (Mann et al., 2000), could also be explained by quantifying the lipophilicity of the zwitterionic species. A possible low lipophilicity could indicate the presence of carriers or transporters facilitating the entrance of these drugs into the central nervous system.

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