34. Lipophilicity Behavior of Model and Medicinal Compounds Containing a Sulfide, Sulfoxide, or Sulfone Moiety

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This study was designed to unravel lipophilicity changes associated with the oxidation state of the S-atom in model compounds, drugs, and metabolites, special attention being given both to intermolecular and intramolecular effects. The methods used were experimental (potentiometry, CPC, and shake-flask techniques to measure lipophilicity, ^{13}C-NMR spectroscopy to investigate tautomeric equilibria) and computational (quenched molecular dynamics and molecular lipophilicity potential). Simple, monofunctional model compounds were used to assess intermolecular forces, as revealed by the $\Delta\log P_{oct-alk}$ and $\Delta\log P_{oct-chf}$ parameters. Drugs and their metabolites proved to be good probes to study intramolecular effects in both neutral and anionic forms, as revealed by the difference between calculated and experimental log P_{oct} values (the *diff*(log $P^{exp-calc}$) parameter). Sulindac and its metabolites showed a normal partitioning behavior, whereas the lipophilicity of sulfinpyrazone and its metabolites was markedly affected by tautomeric and conformational equilibria.

1. Introduction. – During metabolism, organic sulfides can undergo oxidation to sulfoxides and then to sulfones, whereas sulfoxides but not sulfones can undergo reduction [1]. The changes in lipophilicity behavior and body distribution resulting from such metabolic reactions are not straightforward to predict. In an attempt to understand better the changes in lipophilicity associated with the oxidation state of the S-atom, we undertook a systematic study designed *a*) to examine the intermolecular forces influencing the partitioning of model compounds (see *Fig. 1* for chemical structures) in various solvents systems, *b*) to discover whether intramolecular effects could affect the lipophilicity of sulfides, sulfoxides, and sulfones of medicinal relevance, and *c*) to rationalize any change in lipophilicity elicited by such intramolecular effects.

Two drugs containing a sulfoxide group were examined, namely the uricosuric agent sulfinpyrazone (**15**) and the anti-inflammatory drug sulindac (**18**; see *Fig. 2* for chemical structures). Both drugs undergo sulfoxide reduction to an active sulfide metabolite (**14** and **17**, resp.), and sulfoxide oxidation to an inactive sulfone metabolite (**16** and **19**, resp.) [1]. Sulindac (**18**), an isostere of indomethacin [2], may be considered a rather special prodrug, since it is in metabolic equilibrium with its active metabolite [1]. Sulfinpyrazone (**15**), an analogue of phenylbutazone (**13**), has received much attention due to the antithrombotic and antiplatelet properties of its sulfide metabolite which acts as an inhibitor of arachidonate-induced platelet aggregation [3].

Sulfinpyrazone **15** and sulindac **18** being strongly acidic compounds, the relationship between lipophilicity and ionization had to be considered when studying their partitioning behavior [4]. The pH-dependent distribution profiles [5] of the drugs and metabolites under study (**14**–**19**) were thus determined in various solvent systems, leading to the partition coefficients of their neutral (log P^N) and anionic form (log P^A) (*Table 1*).

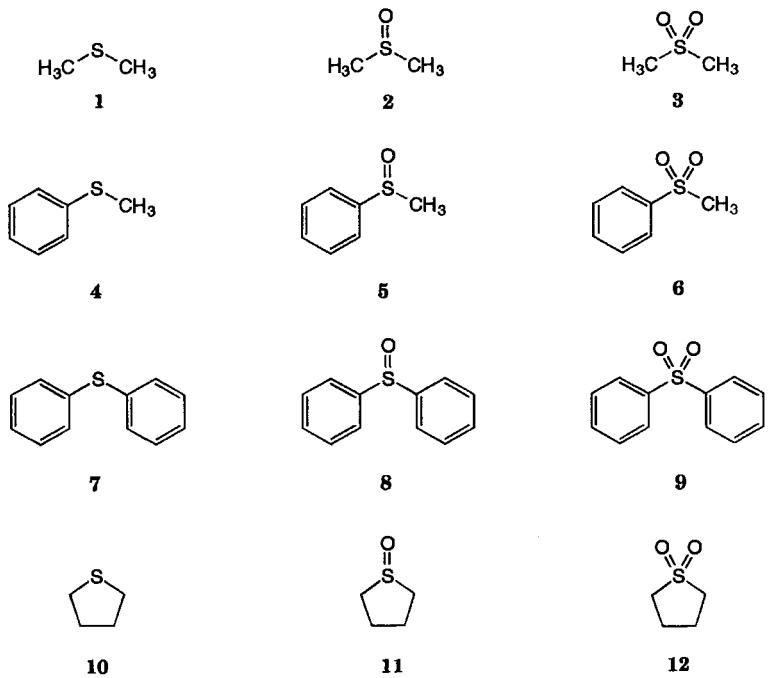
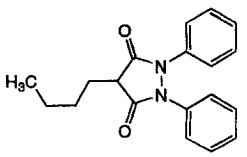


Fig. 1. *Chemical structures of investigated model compounds.* **1**: Dimethyl sulfide, **2**: dimethyl sulfoxide, **3**: dimethyl sulfone, **4**: methyl phenyl sulfide, **5**: methyl phenyl sulfoxide, **6**: methyl phenyl sulfone, **7**: diphenyl sulfide, **8**: diphenyl sulfoxide, **9**: diphenyl sulfone, **10**: 2,3,4,5-tetrahydrothiophene, **11**: 2,3,4,5-tetrahydrothiophene 1-oxide, **12**: 2,3,4,5-tetrahydrothiophene 1,1-dioxide.

Because of the large value of the *diff*(log $P^{exp-calc}$) parameter (*i.e.*, the difference between experimental log *P* and that calculated by an incremental algorithm [6]), intramolecular effects acting on **15** and its metabolites were postulated. Indeed, the involvement of tautomeric and conformational equilibria was confirmed by ^{13}C-NMR spectroscopy and Quenched Molecular Dynamics (QMD) [7], respectively, and their influence on lipophilicity was revealed by the Molecular Lipophilicity Potential (MLP) [8].

2. Results and Discussion. – 2.1. *Model Compounds.* 2.1.1. *Intermolecular Effects as Revealed by Partitioning in Various Solvent Systems.* To extract information about intermolecular interactions influencing the partitioning of model compounds **1**–**12** (*Fig. 1*), their log *P* values were compiled for the octanol/H_2O system, and measured in dodecane/H_2O and $CHCl_3/H_2O$, from which $\Delta\log P_{oct-alk}$ and $\Delta\log P_{oct-chf}$ were calculated (*Table 2*). The latter parameters, it is recalled, are measures of the H-bonding capacity of a solute [9–11]. Two different patterns emerge from *Table 2*: the $\Delta\log P$ values of sulfides (when measurable) are close to zero, whereas for sulfoxides and sulfones $\Delta\log P_{oct-alk}$ values are largely positive and $\Delta\log P_{oct-chf}$ values negative. Thus, the $\Delta\log P_{oct-alk}$ of the *S*-oxygenated compounds is governed by their H-bond acceptor basicity in agreement with the large coefficient of β (1.96 ± 0.42) in solvatochromic equations [10]. In addition, the $\Delta\log P_{oct-chf}$ parameter of the *S*-oxygenated compounds



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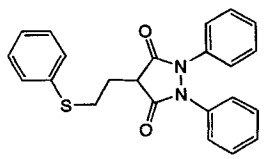

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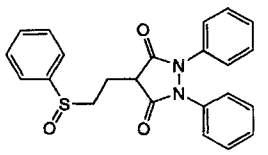

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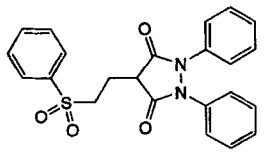

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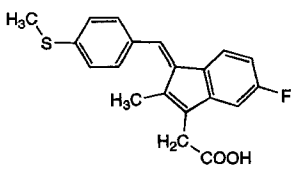

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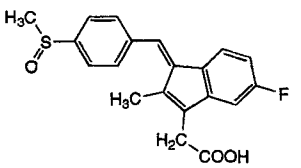

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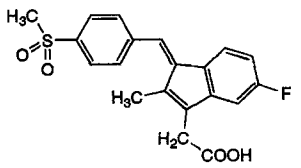

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Fig. 2. *Chemical structures of investigated drugs and their metabolites.* **13**: 4-Butyl-1,2-diphenyl-2,3,4,5-tetrahydropyrazole-3,5-dione, **14**: 1,2-diphenyl-4-[2-phenylthio)ethyl]-2,3,4,5-tetrahydropyrazol-3,5-dione, **15**: 1,2-diphenyl-4-[2-(phenylsulfinyl)ethyl]-2,3,4,5-pyrazole-3,5-dione, **16**: 1,2-diphenyl-4-[2-(phenylsulfonyl)ethyl]-2,3,4,5-tetrahydropyrazole-3,5-dione, **17**: 5-fluoro-2-methyl-1-[4-(methylthio)phenyl]methylideneindene-3-acetic acid, **18**: 5-fluoro-2-methyl-1-[4-(methylsulfinyl)phenyl]methylideneindene-3-acetic acid, **19**: 5-fluoro-2-methyl-1-[4-(methylsulfonyl)phenyl]methylideneindene-3-acetic acid.

Table 1. *Comparison of the Partition Coefficients* (log *P*) *of Phenylbutazone* (**13**) *as Measured by Centrifugal Partition Chromatography (CPC) and Potentiometry (pM)*

	log P^N (CPC)[a]	log P^N (pM)[b]	log P^A (CPC)[c]	log P^A (pM)[d]
Octanol/H_2O	3.04	3.10	0.22	0.13
Dodecane/H_2O	2.60	2.42	−0.19	−0.04

[a]) log $P^{(\text{neutral form})}$ measured by CPC.
[b]) log $P^{(\text{neutral form})}$ measured by potentiometry.
[c]) log $P^{(\text{anionic form})}$ measured by CPC.
[d]) log $P^{(\text{anionic form})}$ measured by potentiometry.

also appears sensitive to their polarizability properties, in keeping with the large negative coefficient of π^* in solvatochromic equations (-0.79 ± 0.36) [10].

Furthermore, the difference in H-bond acceptor basicity between sulfoxides and sulfones [9][12] is revealed by their different $\Delta\log P_{\text{oct-alk}}$ values. The $\Delta\log P_{\text{oct-alk}}$ of the sulfoxide **5** and sulfone **6** are 2.07 and 1.37, respectively, whereas their H-bond acceptor basicity (β) is 0.91 and 0.76 [9]. This fact accounts for the solvent-sensitive partition

Table 2. *Partition Coefficients of Model Sulfides* (**1**, **4**, **7**, and **10**), *Sulfoxides* (**2**, **5**, **8**, and **11**), *and Sulfones* (**3**, **6**, **9**, and **12**) *in Various Solvent Systems*

	$\log P_{oct}$ [a])	$\log P_{alk}$ [b])	$\Delta\log P_{oct-alk}$ [c])	$\log P_{chf}$ [d])	$\Delta\log P_{oct-chf}$ [e])
1	1.05[b])	0.98	0.07	0.72	0.33
2	−1.35	−4.41[a])	3.06	−1.11	−0.24
3	−1.34	< −3	–	−0.60	−0.74
4	2.74	2.81	−0.07	2.38[a])	0.36
5	0.55	−1.52	2.07	1.14	−0.59
6	0.50	−0.87	1.37	1.81	−1.31
7	4.45	> 3	–	> 3	–
8	2.06	0.67	1.39	3.02	−0.96
9	2.40	1.45	0.95	> 3	–
10	1.61	1.59	0.02	1.14	0.47
11	−0.96[b])	< −3	–	−0.30	−0.66
12	−0.77	< −3	–	0.31	−0.46

[a]) Taken from the *Pomona* database [13].
[b]) Measured by CPC; $n = 3$; S.D. ≤ 0.02.
[c]) $\log P_{oct}$ minus $\log P_{alk}$.
[d]) Measured by the shake-flask method except when indicated otherwise; S.D. ≤ 0.05 except for **8** (S.D. = 0.10).
[e]) $\log P_{oct}$ minus $\log P_{chf}$.

coefficients of *S*-oxygenated compounds, as shown in *Table 2*. Surprisingly, the octanol/H_2O system yields similar lipophilicities for sulfoxides and sulfones.

2.1.2. *Derivation of Fragmental Constants for the Octanol/Buffer System.* To assess the intrinsic lipophilicity of the sulfide, sulfoxide, and sulfone moieties, a systematic study on model compounds (*Fig. 1*) to derive fragmental values was performed. From their partition coefficients in octanol/buffer reported in *Table 2*, the sulfide, sulfoxide, and sulfone fragmental values were calculated according to a *Rekker*-type approach [6].

The results (*Table 3*) clearly indicate that the fragmental values are structure-dependent. For dialkyl analogues, the average values are approximately $f(S) = -0.44$, $f(SO) = -2.9$, and $f(SO_2) = -2.8$. Quite identical values were also obtained for cycloalkyl derivatives, making it possible to group the two series of values. In aryl-alkyl analogues the increase in lipophilicity is *ca.* 0.5, 0.8, and 0.7, respectively. In aryl-aryl analogues the increase in lipophilicity is *ca.* 1.1, 1.2, and 1.4, respectively. These variations are correctly taken into account in the CLOGP algorithm [13] but not in the *Rekker*

Table 3. *Fragmental Values*[a]) *in the Octanol/H_2O System*

	Flanking groups	$f(S)$	$f(SO)$	$f(SO_2)$
A[b])	alkyl-alkyl	−0.44	−2.92	−2.83
B[c])	aryl-alkyl	0.11	−2.08	−2.13
C[d])	aryl-aryl	0.65	−1.74	−1.40

[a]) Calculated with the help of the fragmental constants of *Rekker* [6] for other groups.
[b]) Calculated from the experimental values of compounds **1**, **2**, **3**, **10**, **11**, and **12**.
[c]) Calculated from the experimental values of compounds **4**, **5**, and **6**.
[d]) Calculated from the experimental values of compounds **7**, **8**, and **9**.

approach [6]. When examining the lipophilicity of medicinal compounds containing an aryl and an alkyl flanking group (see below), the fragmental constants derived from compounds **4**–**6** will be used.

2.2. *Medicinal Compounds.* 2.2.1. *Ionization Constants.* For phenylbutazone (**13**) analogues, there exists a relation in humans between acidic pK_a, half-life, urinary excretion, and uricosuric activity [14]. Thus, compounds with pK_a of 4.5 to 5.5 were found to be excreted only to a small extent, to have a relatively long half-life and a slight uricosuric activity, and generally to be potent antirheumatic agents. In contrast, analogues with pK_a of 2.3 to 3.1 are rapidly excreted in urine, have short half-lifes, but are potent uricosuric agents.

To enhance the acidity of **13** to obtain a potent uricosuric agent, a S=O group was introduced, yielding sulfinpyrazone (**15**) [14]. This group delocalizes the negative charge of the anion due to its strong inductive effect [15], producing a two-unit decrease in pK_a (*Table 4*).

The trend of a greater acidity of sulfones compared to sulfoxides and even more to sulfides [16] is confirmed here for **15** and its metabolites **14** and **16**, but not in the sulindac (**18**) series where the parent compound **18** is almost as acidic as its sulfone metabolite **19**. The ionizability of a solute obviously has a major impact on its partitioning behavior. Recent studies [17] show that the distribution of ionizable compounds in biological systems depends not only on their neutral form, but also on the ionized form. As a consequence, it is important when studying the partitioning behavior of ionizable compounds to measure their complete lipophilicity profile (distribution coefficient log *D vs.* pH) [5] in order to reach the partition coefficient of both the neutral and ionized form.

The pH-dependent distribution profile of acids can be described by *Eqn. 1* which is adapted from the general distribution function [5]:

$$D = P^N \cdot \left(\frac{1}{1 + 10^{pH - pK_a}}\right) + P^A \cdot \left(\frac{10^{pH - pK_a}}{1 + 10^{pH - pK_a}}\right) \quad (1)$$

where P^N and P^A are the partition coefficients of the neutral form and anion, respectively.

Table 4. *Dissociation Constants and Partition Coefficients of Neutral and Anionic Forms of Medicinal Compounds in the Octanol/H_2O System*

	pK_a[a])	log P^N[b])	S – SO[c])	log P^A[d])	S – SO[e])	*diff*(log P^{N-A})[f])
13	4.61(4.80)	3.10		0.22		2.88
14	2.55	5.31	1.75	0.93	1.03	4.38
15	2.37(3.25)	3.56		−0.08		3.64
16	2.09	3.51		−0.10		3.61
17	4.88	4.76	1.47	1.85	2.80	2.91
18	4.03(4.50)	3.29		−0.95		4.24
19	4.16	3.29		−0.79		4.08

[a]) Measured by potentiometry; MeOH as cosolvent was used except for **13**; $n = 4$, S.D. < 0.02. The *Pomona* database value [13] is given in parentheses.

[b]) Determined by potentiometry.

[c]) log $P^{(\text{neutral form})}$ of the sulfide minus log $P^{(\text{neutral form})}$ of the correspondent sulfoxide.

[d]) log $P^{(\text{anionic form})}$ of the sulfide minus log $P^{(\text{anionic form})}$ of the correspondent sulfoxide.

[e]) log $P^{(\text{neutral form})}$ minus log $P^{(\text{anionic form})}$.

2.2.2. *Partitioning in Octanol/H_2O.* 2.2.2.1. *Calculated log* P *Values.* To investigate possible intramolecular effects, a standard procedure is to compare calculated log *P* values (*e.g.*, by the CLOGP algorithm [13]) with experimental values [18]. The CLOGP values of the investigated medicinal compounds are shown in *Table 5*. For sulfinpyrazone (**15**) and its metabolites **14**–**16**, *Rekker*'s approach was also applied to calculate log *P* values [6] with the revised fragmental values of S, SO and SO_2 listed in *Table 5*. This latter method is of interest, because it allowed the log *P* values of both the neutral (log P^N) and anionic (log P^A) forms to be calculated, starting from the experimental log P^N and log P^A values of phenylbutazone (**13**) (*Table 1*) and the fragmental values of the relevant S moiety (*Table 3*).

Table 5. *Comparison between Experimental and Calculated log* P *Values in the Octanol/H_2O System*

	log P[a])	CLOGP[b])	*Rekker*[ca])	*diff*(log $P^{exp-calc}$)[d])
14	5.31	3.58	3.82	1.49[f])
15	3.56	1.43	1.59	1.97[f])
16	3.51	1.39	1.65	1.86[f])
17	4.76	4.92	–	−0.16[g])
18	3.29	2.77	–	0.52[g])
19	3.29	2.72	–	−0.57[g])
14 (an)[e])	0.93	–	0.94	−0.01[f])
15 (an)[e])	−0.08	–	−1.35	1.27[f])
16 (an)[e])	−0.10	–	−1.23	1.13[f])

[a]) See *Table 4*.
[b]) Taken from the *Pomona* database [13].
[c]) Fragmental value coming from [6].
[d]) log $P^{(experimental)}$ minus log $P^{(calculated)}$.
[e]) Anionic form of the indicated compound.
[f]) The calculated value is from *Rekker*'s system.
[g]) The calculated value is from CLOGP.

As shown in *Table 5*, there is a good agreement between the log *P* (*i.e.*, log P^N) values calculated by the two methods (difference always < 0.3).

2.2.2.2. *Experimental log* P *Values.* The partition coefficients and related parameters of compounds **14**–**19** are shown in *Table 4*, demonstrating again that sulfides are more lipophilic than sulfoxides and sulfones, whose partition coefficients are similar. Later discussions will, therefore, focus mostly on sulfides and sulfoxides.

In the partition coefficient of neutral forms (log P^N), the differences between sulfides and *S*-oxygenated compounds (*Table 4*) are smaller than in model compounds. This would suggest that the S-containing moieties express their lipophilic increment differently depending on the rest of the molecule.

The parameter *diff*(log P^{N-A}) (*i.e.*, the difference between the lipophilicity of the neutral and anionic form) (*Table 4*) contains important structural information, being *a priori* a function of solvent system and various intramolecular effects (*Eqn. 2*):

$$diff(\log P^{N-A}) = f(\text{solvent system}) + f(\text{conformational effects}) + f(\text{tautomeric effects}) + \ldots \quad (2)$$

Whereas such contributions cannot be deconvoluted directly from *diff*(log P^{N-A}) values, this parameter can point to the existence of intramolecular effects.

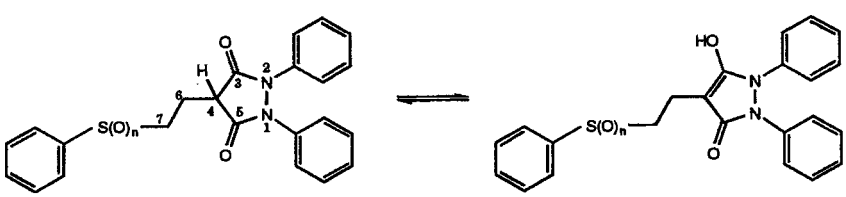
As shown in *Table 5*, there is a fair agreement between calculated values (using CLOGP) and experimental values for sulindac (**18**) and its metabolites **17** and **19**, with differences smaller than 0.6 unit. This rules out any major conformational effects that would affect lipophilicity. In contrast, the differences are significant (around 2.0 units) for sulfinpyrazone (**15**) and its metabolites **14** and **16** (using *Rekker*'s system). The *diff*(log $P^{exp-calc}$) of the anionic form of compounds **15** and **16** is also remarkable. Studies were, therefore, undertaken to unravel the nature of the effects that must affect lipophilicity.

2.2.2.3. *Tautomeric Equilibria.* A starting hypothesis to understand intramolecular factors affecting lipophilicity is the diketo/keto-enol tautomerism (see *Table 6*) existing in the neutral form of compounds **14**–**16**, but not in their anions [19][20]. In fact, the log *P* values calculated by *Rekker*'s method are based on the experimental log *P* of phenylbutazone (**13**) (see above) where the percentage of enolic and ketonic forms in H_2O is 1.8% and 98.2% [21], respectively, whereas the compound is mostly diketonic in DMSO [22]. In other words, the calculated values of **14**–**16** mostly neglect a contribution of the enolic tautomer which is more lipophilic than the ketonic tautomer as verified for acetone by the CLOGP algorithm (keto: −0.21; enol: 0.83).

To explore the diketo/keto-enol equilibrium in the neutral form of **14**–**16**, ^{13}C-NMR spectroscopy was used (*Table 6*). Both tautomers can easily be identified in the spectra [22], and their proportion assessed from the ratio of intensities of significant peaks. To avoid problems caused by different relaxation times, at least three pairs of peaks were considered. Interestingly, the diketo/keto-enol ratio of the sulfide **14** is *ca.* 3:1 and larger than that of the sulfoxide **15** (*ca.* 1.5:1) and the sulfone **16** (*ca.* 1:1). This difference could explain the small influence of the tautomeric equilibrium on the lipophilicity of **14** and the more accurate calculation of its log *P* compared to oxygenated compounds.

Nevertheless, tautomeric equilibria alone cannot account completely for the observed *diff*(log $P^{exp-calc}$) values. In fact, anionic forms that are not affected by tautomeric

Table 6. ^{13}*C-NMR Chemical Shifts of Sulfinpyrazone* (**15**) *and Its Metabolites* **14** *and* **16** *in* (D_6)*DMSO.* A non-systematic numbering of C-atoms is used.



	n = 0 (**14**)[a])	n = 1 (**15**)[a])	n = 2 (**16**)[a])
C(3)	170.14 (165.92)	169.69 (165.83)	169.40 (165.59)
C(4)	44.37 (85.61)	44.10 (84.08)	43.48 (82.87)
C(6)	29.05 (30.70)	19.02 (13.92)	20.37 (14.96)
C(7)	26.28 (21.21)	51.43 (54.11)	51.73 (52.87)

[a]) Chemical shifts (δ in ppm) relative to (D_6)DMSO. In parentheses the values of the keto-enol tautomer.

equilibria also show a remarkable *diff*(log $P^{exp-calc}$). The conformational behavior of sulfinpyrazone (**15**) and its metabolites could also affect their lipophilicity and was investigated by classifying the conformers according to their virtual lipophilicity.

2.2.2.4. *Conformational Equilibria.* Following an exploration of the conformational space by QMD, the molecular lipophilicity potential (MLP) of all retained conformers was calculated, from which their virtual log *P* values were obtained [18].

The range of virtual log *P* values covered by sulfinpyrazone (**15**) was *ca.* 0.8 log *P* unit (*Table 7*), while those of the sulfone **16** and the sulfide **14** are *ca.* 1 and 0.4 unit, respectively. The MLPs of the conformers of highest and lowest virtual log *P* are represented in *Fig. 3* for sulfinpyrazone (**15**), its sulfone **16**, and its sulfide **14**. In the folded conformers of **15** and **16**, both the S-containing moieties and the pyrazolidine-dione ring are prevented from expressing their full polarity (hydrophilicity) due to the masking effect of the Ph groups. In the folded conformers of **14**, masking of polarity involves only the pyrazolidine-dione ring, since the sulfide group is not polar. This explain the smaller lipophilicity range covered by compound **14** relative to **15** and **16**.

Table 7. *Classification of Conformers According to Their Lipophilicity Behavior: the Lipophilic Range Values*

	log P^{low} [a])	log P^{high} [b])	Lipophilicity range [c])	*Average* log P [d])
13	2.47	2.64	0.17	2.55 ± 0.08
14	2.73	3.10	0.37	2.92 ± 0.19
15	1.20	1.96	0.76	1.58 ± 0.38
16	1.05	2.04	0.99	1.55 ± 0.50

[a]) The lowest virtual log *P*.
[b]) The highest virtual log *P*.
[c]) Lipophilicity range (see text for definition) calculated as log P^{low} – log P^{high}.
[d]) Calculated as follows:

$$\text{average log } P = \frac{\log P^{low} + \log P^{high}}{2} \pm \frac{\text{range}}{2}$$

Such a masking effect on the polarity of the pyrazolidine-dione ring does not exist in phenylbutazone (**13**) due to the absence of a third Ph group.

It must be noted that the conformational effects reported here concern the neutral forms only. Indeed, the MLP in its current state of development cannot yet handle the lipophilicity of this type of delocalized anions. However, it is reasonable to expect some intramolecular folding effects also in the ionized forms of **14**–**16**. Indeed, the higher value of *diff*(log P^{N-A}) for the sulfide **14** compared to **15** and its metabolite **16** confirms that the anionic forms of **15** and **16** experience a polarity-decreasing masking effect.

In summary, the *diff*(log $P^{exp-calc}$) parameter brings intramolecular effects to light as schematized in *Fig. 4*. Tautomeric and conformational effects influence the lipophilicity of neutral sulfinpyrazone (**15**) and its metabolites, whereas the anionic forms of **14**–**16** experience only conformational effects.

2.2.3. *Partitioning of Compounds* **13**–**16** *in the 1,2-Dichloroethane/H_2O System.* Partition coefficients obtained in the alkane/H_2O system (log P_{alk}) are of interest in drug design, since they encode a larger H-bonding contribution of solutes than octanol/H_2O partition coefficients [10]. However, the total insolubility of sulindac and metabolites in

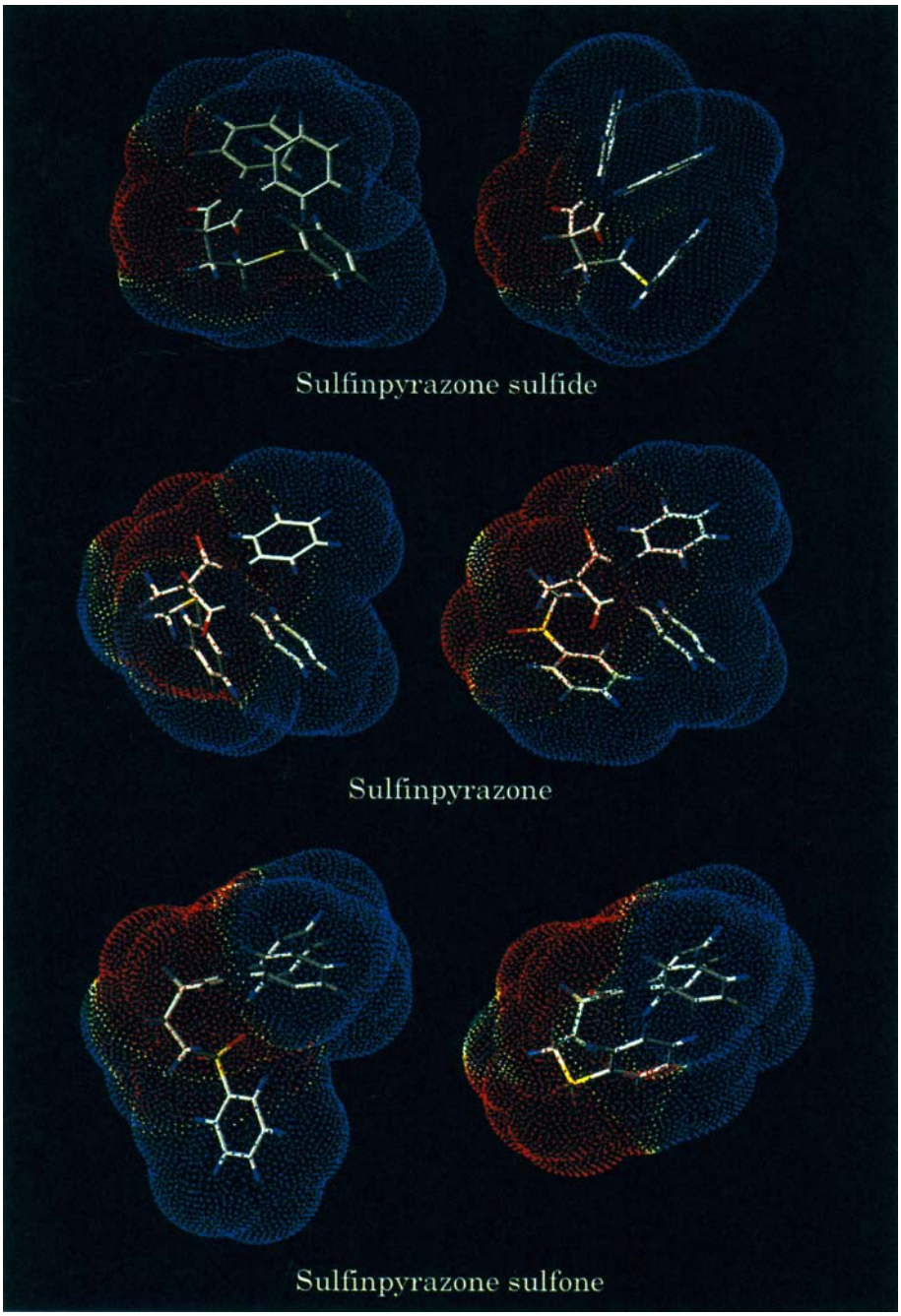


Fig. 3. *Molecular Lipophilicity Potential (MLP) of conformers involved in the calculation of the virtual log* P *range of sulfinpyrazone* (**15**; middle), *and its sulfide* (**14**; top) *and sulfone* (**16**; bottom) *metabolites.* The most lipophilic compounds are on the left and the most hydrophilic on the right. The blue dots represent the lipophilic regions and the red the polar regions of the molecules.

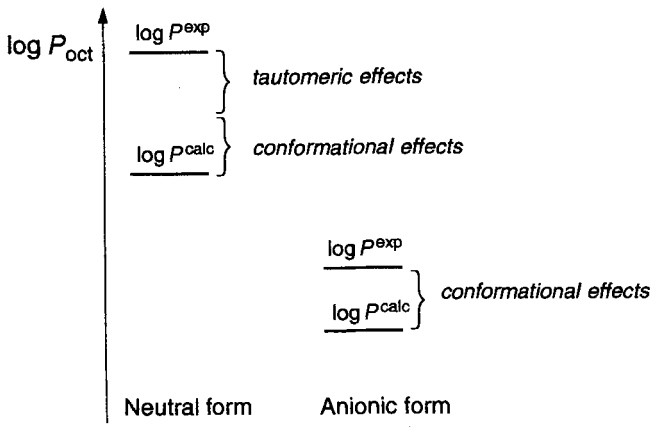


Fig. 4. *A schematic representation of intramolecular effects acting on the lipophilicity of neutral and anionic forms of sulfinpyrazone* (**15**) *and its metabolites* **14** *and* **16**

dodecane, and the very low solubility of sulfinpyrazone (**15**) and its metabolites, made it impossible to obtain reliable values. Partition coefficients of **13**–**16** were, therefore, determined in the 1,2-dichloroethane/H_2O system (log P_{dce}), since a recent solvatochromic analysis has demonstrated that log P_{dce} values give the same information as log P_{alk} as far as H-bonding properties of solutes are concerned [23]. The difference between log P_{dce} and log P_{alk} is mainly due to the property of 1,2-dichloroethane to stabilize compounds with a large polarizability (π^* term).

For the 1,2-dichloroethane/H_2O system, no algorithm exists to calculate log P values and hence to derive the *diff*(log $P^{exp-calc}$) parameter. However, the difference in log P^N_{dce} between sulfinpyrazone (**15**) and its metabolites (*Table 8*) is 1 unit larger than in log P_{oct} (*Table 4*); this suggests a different balance in tautomeric equilibria.

Δlog P Parameters are of interest in pharmacokinetics, since they are often negatively correlated with membrane permeability [11]. The values of Δlog $P_{oct-dce}$ for the neutral and anionic forms of compounds **13**–**16** are reported in *Table 8*. Whereas the Δlog $P_{oct-dce}$ of dimethyl sulfoxide (**2**) is positive [23], negative values of Δlog $P_{oct-dce}$ were found for the neutral form of phenylbutazone (**13**), sulfinpyrazone (**15**), and its

Table 8. *Partition Coefficients of Compounds* **13**–**16** *in the 1,2-Dichloroethane/H_2O System*

	log P^{N}[a])	log P^{A}[b])	*diff*(log P^{N-A})[c])	Δlog $P^{N}_{oct-dce}$[d])	Δlog $P^{A}_{oct-dce}$[e])
13	4.72	−0.77	5.49	−1.70	0.99
14	7.70	1.28	6.42	−2.02	−0.35
15	4.69	−1.24	5.93	−1.22	1.16
16	5.53	−1.08	6.61	−2.05	0.98

[a]) log P of the neutral form measured by potentiometry.
[b]) log P of the anionic form measured by potentiometry.
[c]) log $P^{(neutral\ form)}$ minus log $P^{(anionic\ form)}$.
[d]) log $P^{(neutral\ form)}_{oct}$ − log $P^{(neutral\ form)}_{dce}$.
[e]) log $P^{anionic\ form)}_{oct}$ − log $P^{(anionic\ form)}_{dce}$.

metabolites. This is due to the fact that log P^{N}_{dce} values (*Table 8*) are greater than the corresponding log P^{N}_{oct} values (*Table 4*), *i.e.*, that 1,2-dichloroethane attracts these solutes more than does octanol, presumably due to marked *van der Waals* interactions (π^* term).

In contrast, positive values of $\Delta\log P_{oct-dce}$ were found for the anionic forms of compounds **13**, **15**, and **16**, but not **14**, indicating that 1,2-dichloroethane has less affinity for the anions (except **14**) than octanol. As a result of such differences, the *diff*(log P^{N-A}) parameter is 2–3 units larger in 1,2-dichloroethane than in octanol.

Thus, many differences become apparent between the log P_{oct} and log P_{dce} values of compounds **13**–**16**. Such differences are intriguing and suggest differences in intramolecular and intermolecular interactions, but they cannot be interpreted further at present.

3. Conclusion. – This study serves to illustrate the changes in lipophilicity associated with the oxidation state of the S-atom. Because intermolecular forces influence the partitioning of model compounds in various solvents systems, the difference in basicity between sulfoxides and sulfones will be revealed by their different $\Delta\log P_{oct-alk}$ value. In addition, intramolecular effects acting on sulfides, sulfoxides, and sulfones of medicinal relevance were uncovered, again demonstrating the effects of tautomeric and conformational equilibria on lipophilicity.

Such dynamic relations between structure and properties are expected to have a marked influence on the distribution of drugs and metabolites in the body, but a much better understanding of such relations is required before quantitative permeability predictions can be ventured.

Experimental Part

1. *Solvents.* Anal. grade octanol, $CHCl_3$, and $ClCH_2CH_2Cl$ were purchased from *Fluka* (Buchs, CH). Anal. grade dodecane was obtained from *Aldrich* (Steinheim, D). Sodium phosphate (*Fluka*) was used for buffers of pH 7.4 and 4.5. Glycine (*Fluka*) was used for buffers of pH 2.0. *He 57* (*i.e.*, 99.9997% purity) for GC/MS analysis was obtained from *Carbagas* (Liebefeld-Bern, CH). The deuterated DMSO (of 99.8% isotopic purity) was purchased from *Armar* (Döttingen, CH).

2. *Solutes.* Compounds **1**–**11** were purchased from *Fluka*. Compound **12** was obtained from *Aldrich*. Compound **13** was purchased from *Siegfried*, **15** and its metabolites **14** and **16** were kindly donated by *Ciba-Geigy Ltd* (Basel, CH), and **18** and its metabolites **17** and **19** were kindly donated by *Merck Research Laboratories* (Rahway, NY, USA).

The purity of the compounds was checked by HPLC and GC/MS. The HPLC equipment consisted of a *Kontron MT1* chromatograph equipped with a *MSI T-660* autosampler, an HPLC pump model *420*, a column oven 480, an oven controller 480, and an UV/VIS detector model *430* with variable wavelength (*Kontron AG*, Zürich-Müllingen, CH). The column was a *Supelcosil LC-ABZ* (150 × 4.6 mm ID; *Supelco*, Bellefonte, PA, USA) of 5 μm packing and 100-Å pore size, the eluent was MeOH/buffer (0.02M phosphate pH 7.4) 70:30 *v/v*, and the flow-rate was 1.0 ml · min^{-1}. The GC/MS equipment consisted of a *HP 5890 Series II* gas chromatograph with a *HP 7673* autosampler, and an *HP-1* column (100% dimethylpolysiloxane; 325° upper temp. limit), a *HP 5971A* mass-selective detector with a *HP G1034A* MS *ChemStation Software*. The following analysis conditions were used: carrier gas He 57, initial temp. 40°, final temp. 280°, rate of 50.00 (°C · min^{-1}). The split inlet system and the SIM mode of acquiring the MS data were used.

3. pK_a *Determinations.* Potentiometric titrations of compounds **13**–**19** were performed with the *PCA101* apparatus [24] (*Sirius Analytical Instruments Ltd*, Forrest Row, East Sussex, UK) equipped with a semi-micro *Ross*-type double junction combination pH electrode (*Orion 8103SC*), a temp. probe, an overhead stirrer, a precision dispenser, and a six-way valve for distributing reagents and titrants (0.5M HCl, 0.5M KOH, 0.15M KCl, and MeOH). A weighted sample (1–10 mg) was supplied manually, whereas the diluent and all other reagents were added automatically. Except for **13**, the low aqueous solubility of compounds required pK_a measurements to be performed in the presence of MeOH as cosolvent. Four separate 20-ml semiaqueous solns. of *ca.* 1 mM, in 30–60

(%*w*/*w*) MeOH were initially basified to pH 12.2 with KOH. The solns. were then titrated with standardized HCl to pH 2. The titrations were conducted under Ar at 25.0 ± 0.1°. The initial estimates of the $p_s K_a$ values, which are the apparent ionization constants in the mixed solvent, were obtained by *Bjerrum* plots. These values were then refined by a weighted nonlinear least-squares procedure. The refined values were then extrapolated to zero by the *Yasuda-Shedlovsky* procedure [25].

4. *Measurement of Partition Coefficients of the Non-ionizable Model Compounds* **1**–**12**. The partition coefficients in the $CHCl_3$/buffer system (log P_{chf}) were measured at r.t. by the shake-flask method [26]. The pH values was 7.4. The two layers were shaken for *ca.* 8 h, separated, and centrifuged (10 min). The concentration of solutes was measured in the aq. phase by HPLC, when log P_{chf} was greater than 0, and in the org. phase by GC/MS, when log P_{chf} was smaller than 0 or for UV-inactive compounds. All values were obtained at least in quadruplicate.

The partition coefficients in octanol/buffer and in dodecane/buffer (0.02M phosphate pH 7.4 and 4.5, 0.02M glycine/HCl pH 2) were determined by flow-through CPC with a coil planet type centrifuge (*Ito Multi-layer Coil Separator-Extrator, P.C. Inc.*, Baltimore, USA), as described in detail elsewhere [27][28].

5. *Measurement of Partition Coefficients by the pH-Metric Method.* Three separate acid titrations of *ca.* 1 mn for compounds **13**–**19**, initially basified to pH 12.2 with KOH, containing various volumes of octanol or $ClCH_2CH_2Cl$ (from 1 ml of org. solvent/15 ml of H_2O to 8 ml of org. solvent/8 ml H_2O), were performed in the pH range 12.2 to 1.8 with the *PCA101* apparatus. The titrations were conducted under Ar at 25.0 ± 0.1°.

6. *Validation of Techniques.* To verify that the partition coefficients of neutral and anionic forms (log P^N and log P^A, resp.) obtained by potentiometry and by CPC [28] in various solvent systems were in good agreement, validation experiments were performed. Phenylbutazone (**13**) was chosen as the reference compound, and octanol/H_2O and dodecane/H_2O as reference solvent systems.

From the distribution profiles of **13** obtained both by CPC and by potentiometry the values of log P^N and log P^A were extrapolated by a nonlinear regression of log *D* vs. pH. The values of log P^N and log P^A are reported in *Table 1* and are indeed in very good agreement both in octanol/H_2O and in dodecane/H_2O (differences between 0.06 and 0.18). It is interesting to note that, despite the different ionic strengths used in the two techniques, a negligible difference was obtained in the partition coefficients of anionic forms. This suggests that, for acidic compounds, the contribution of the counterion in the partitioning of the anionic form is negligible.

7. *Quenched Molecular Dynamics (QMD).* A simplified conformational search [7][29] strategy was adopted which is able to describe efficiently the main valleys of a conformational space. Various starting geometries were used, among them those obtained from the crystallographic structure of phenylbutazone (**13**) [19] and of sulfinpyrazone (**15**) [30]. Additional starting geometries (three or four) were built by the CONCORD algorithm [31] and energy-optimized using the *Tripos* force field [32] with *Gasteiger-Marsili* formal atomic charges [33] in order to remove initial high-energy interactions. In both cases, the diketonic structure was retained.

High-temp. molecular dynamics (MD) calculations were carried out at 2000 K. Each simulation was run for 100 ps with steps of 1.0 fs. The frame data were stored every 0.05 ps, giving 2000 frames. The starting velocities were calculated from a *Boltzmann* distribution. Finally, 10% of all conformers were randomly selected and saved in a database ultimately containing about 200 conformers.

All conformers in the database were then subjected to energy-minimization using the same force field as for the MD calculations. The *Powell* minimization method was applied with the gradient value of 0.001 to test for convergence. The maximum number of iterations was set at 3000. The energy-minimized conformers were then classified according to increasing energy.

The conformational similarity of the 200 energy-minimized conformers was investigated by comparing all pairs of conformers. The two criteria of comparison were the force-field energy and the RMS distance difference calculated by the option MATCH of SYBYL over all heavy atoms and polar H-atoms. An *ad hoc* Fortran program then calculated the mean and standard deviations of the RMS values. Two conformers were considered identical, when their energy difference was ≤ 3 kcal/mol, and their RMS distance difference less than or equal to the RMS mean minus the standard deviation. When this was the case, one of the two conformers was eliminated from the database, and it was always the one of higher energy.

The selected conformers were minimized a second time at the semi-empirical level with the AM1 [34] parametrization without the keyword PRECISE. The AM1-minimized conformers were again classified according to increasing heat of formation and selected by heat of formation and RMS distance difference. Identical conformers were again eliminated from the database using the same criteria as above. Finally, the AM1 calculations were repeated on the retained conformers, but with a higher level of precision (inclusion of the keyword PRECISE).

All calculations were run on *Silicon Graphics Personal Iris 4D-35*, *Indigo R4000*, or *Indy R4400* workstations. SYBYL 6.2 molecular modeling package (*Tripos Associates*, St. Louis, MO, USA) and MOPAC (*QCPE*, No. 445) were used.

8. *Calculation of Partition Coefficients from the Molecular Lipophilicity Potential (MLP).* The Solvent-Accessible Surface Area (SASA) [18] of the conformers generated by QMD (compounds **13**–**16**) was utilized as the space for integrating the MLP back to log P_{oct} values using the following equation [8]:

$$\log P_{oct} = 2.86 \cdot 10^{-3} (\pm 0.24 \cdot 10^{-3}) \sum MLP^{+} + 1.52 \cdot 10^{-3} (\pm 0.22 \cdot 10^{-3}) \sum MLP^{-} - 0.10 (\pm 0.23) \quad (3)$$
$$n = 114;\ r^2 = 0.94;\ s = 0.37;\ F = 926$$

where $\sum MLP^{+}$ and $\sum MLP^{-}$ represent the hydrophobic and polar parts of the molecule, respectively, *i.e.*, the regions of the surface where positive and negative atomic values of lipophilicity are expressed. The MLP calculation were performed with the CLIP 1.0 software [35].

The most lipophilic and hydrophilic conformers were retained and the difference between their *virtual log* P *values* (virtual log *P* being the log *P* calculated for a given conformer [18]) were considered as the lipophilicity range accessible to a given solute in the neutral state. The lipophilicity range encompasses the ensemble of all virtual log *P* values of a solute, whereas the experimental log *P* is the weighted average of an unknown number of virtual log *P* values of the molecule [18].

9. *^{13}C-NMR Spectroscopy.* The ^{13}C-NMR spectra in (D_6)DMSO were recorded at 50 MHz on a *Varian VXR-200* spectrometer.

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