Molecular Lipophilic Potential on van der Waals Surfaces as a Tool in the Study of 4-Alkylpyrazoles

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A new method for the calculation and analysis of lipophilic properties on molecular surfaces is presented. In this method, both, the concept of molecular lipophilic potential developed by Audry and co-workers and the approach of Ghose and Crippen implying that the hydrophobicity of a molecule can be obtained as the sum of certain atomic contributions, are combined. Then, shape analysis methodology is applied to characterize the interrelation between this molecular lipophilic potential and the van der Waals surfaces of a family of pyrazoles, which are known inhibitors of the enzyme liver alcohol dehydrogenase. In this study graphical representations and analytical methods are used. The mentioned topological analysis provides certain codes which combine lipophilic and steric information and are unique and characteristic of each pyrazole. These three-dimensional codes were correlated with the inhibitory activity of this series of pyrazoles. The inhibitory power of 4-isopentylpyrazole was not known experimentally before this study was carried out. Thus, by using these combined codes, the inhibitory activity of the isopentyl derivative was suggested to be lower than that of its linear isomer. This hypothesis was confirmed after the synthesis of 4-isopentylpyrazole and its enzymatic evaluation.

INTRODUCTION

The importance of hydrophobicity in chemistry and biology has been recognized for a long time, and this property has to be considered as an essential factor in the interactions between bioactive molecules and receptors.[1,2] Hydrophobicity may be described as a combination of two effects: an energetic and an entropic one. The energetic term is associated with differences in energy between a molecule and the solvent molecules, and the entropic effect is related to the polarizability of both solvent and solute. However, there is still no simple physical model available for describing hydrophobic effects and for that reason a number of empirical[3−8] and quantum mechanical[9] approaches have been developed to define relative lipophilicity values.

Drug−receptor interactions involve basically the same intermolecular forces as those acting in the partitioning of a solute between water and an immiscible organic phase. For that reason, the logarithm of the partition coefficient *P* has been classically used to represent the hydrophobic characteristics of a molecule. However, this one-dimensional representation is not sufficient when three dimensional drug−receptor interactions are analyzed. To avoid such a limitation, Audry et al. have defined the molecular lipophilic potential (MLP)[10] which provides a better description of the hydrophobicity of a molecule. Other potentials have been developed in order to understand molecular lipophilicity, between them we should mention that proposed by Furet, Sele, and Cohen[7] (who revised that from Audry et al.) and that developed by Kellogg and Abraham (HINT)[11] that has been incorporated into comparative molecular field analysis (CoMFA) an important "tool" used for molecular modeling and drug design.

In addition, representation of physicochemical properties on molecular surfaces[12−14] allows for a complete definition of a molecule since this describes not only the physicochemical property studied but also the steric factor and how shape and physical characteristic interrelate. The topological study of this interrelation between the chosen property and the molecular surface results in certain codes that describe the molecule in a unique way and that can be correlated with biological activities.[15−17]

In this paper, we propose a new empirical approach to evaluate the lipophilic characteristic of a molecule by analyzing the topological properties of the interrelation between a molecular lipophilic potential MLP_a (proposed by Furet et al.[7]) and the van der Waals surface (VDWS). To show the possibilities of this new method we applied this approach to a series of 4-substituted pyrazoles, compounds well-known structurally and biologically and that have been the object of our interest in previous papers.[15−17]

It is well-known that 4-alkylpyrazoles are potent inhibitors of the liver alcohol dehydrogenase enzyme (LADH),[18] and it has been shown that by increasing the length of the alkyl chain the inhibitory power increases.[19] On the contrary, the branching or the cyclization of these alkyl chains lower the inhibitory activity.[20] Both effects are related to the lipophilicity of these molecules which depends directly on the characteristics of the alkyl chain. Thus, lipophilicity may play a more important role in the LADH inhibition by pyrazoles than electronic factors, and, therefore, these pyrazoles would be better described by analyzing their lipophilic characteristics. For these reasons, and to proof the ability of this new method to evaluate lipophilic contributions, we have studied the effect that a methyl group has on the lipophilicity and on the activity of some 4-alkylpyrazoles when it is introduced as a branch in their side chains. The compounds chosen for this study are the following: pyrazole (**1**), 4-methyl- (**2**), 4-ethyl- (**3**), 4-propyl- (**4**), 4-(1-methyl) ethyl- (**5**), 4-butyl- (**6**), 4-(1-methyl) propyl- (**7**), 4-(2-methyl) propyl- (**8**), 4-pentyl- (**9**), 4-(1-methyl)-butyl- (**10**), 4-(2-methyl)butyl- (**11**), and 4-(3-methyl)-

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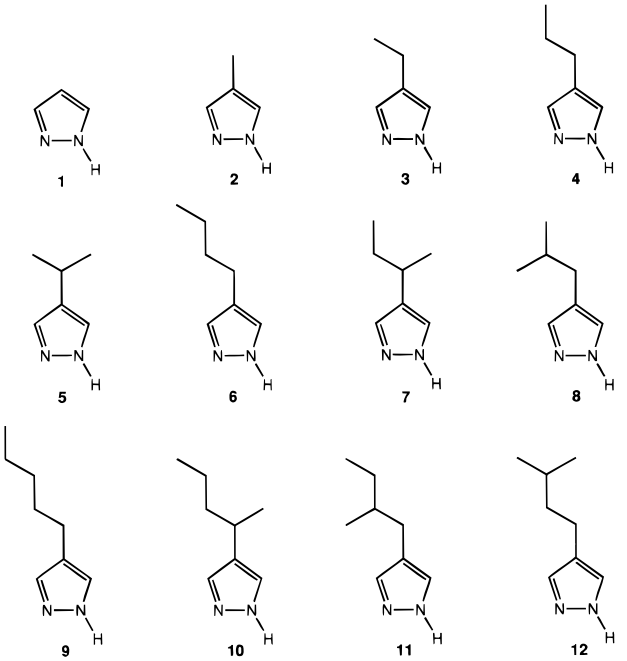

Figure 1. Pyrazoles studied in this work. One methyl group is introduced as a branch and moved along linear chains of different length.

butylpyrazole (**12**) (Figure 1). Although there is no possibility for branching in compounds **1**, **2**, and **3**, they have been studied to consider the effect of introduction and elongation of the alkyl chain and for the sake of comparison. Compounds **7**, **10**, and **11** have a chiral C atom in the alkyl chain and only the *S* enantiomer has been studied, because the activity values found for some of them correspond to that enantiomer. Most of these molecules can present different conformations, and, for that reason, in a previous paper,[16] we carried out an exhaustive conformational analysis of compounds **2**−**12**. All the local and global minima obtained for the 11 derivatives in this former study were the structures analyzed in this work. The activity parameter used for comparing to the lipophilicity has been the inhibition constant of the enzyme (K_I, in μM), which is known for the most of the compounds.[21]

This article is organized as follows: First, the molecular lipophilic potential MLP_a and the theoretical log *P* are described and then calculated for this set of pyrazoles. Next, the interrelations between the MLP_a and the VDWS for this family of compounds are topologically characterized and the resulted three-dimensional codes correlated with the corresponding inhibitory power. Finally, some concluding remarks are made.

MOLECULAR LIPOPHILIC POTENTIAL AND CALCULATED LOG *P*

Considering the approach that a molecule can be regarded as a set of **j** fragments, Fujita et al.[22] showed that the partition coefficient (*P*) can be estimated from its chemical structure. Thus, the log *P* has been defined as the summation of certain hydrophobic fragment constants

$$\log P = \sum_{i=1}^{j} f_i \qquad (1)$$

where f_i is the lipophilic contribution of a molecular fragment to the total lipophilicity of the molecule. Rekker[23,24] introduced this system of hydrophobic fragment constants, and, later on, Hansch and Leo[25] performed more detailed studies. The value of f_i provides a numerical estimation of the affinity of a fragment for an organic phase. In the same way the log *P* evaluates this affinity of a whole molecule.

Considering a molecule S, constituted by **j** fragments, inside an organic phase, Audry et al.[10] defined the molecular lipophilic potential (MLP) in any point *M* of the space around the molecule S as

$$\mathrm{MLP} = \sum_{i=1}^{j} \frac{f_i}{1 + d_i} \qquad (2)$$

where d_i is the distance (in Å) between the point M and the "i" fragment of the molecule S.

Other important hydrophobic field developed in the last years has been HINT by Kellogg and Abraham[11] which uses the fragmental system of Hansch and Leo[25] and an exponential distance function to represent lipophilicity in a 3D manner.

All these authors used the f_i constants originally defined by Rekker[23,24] which are related to "molecular fragments". Thus, each f_i factor describes the lipophilicity of a particular set of atoms defining a specific functional group. However, the calculation of lipophilicity on the van der Waals surface requires those f_i factors to be referred to atoms not to functional groups.

Furthermore, atomic parameters present a number of advantages: (i) a molecule immersed in a solvent may experience different interactions in different regions of its surface and (ii) the relative orientation or conformation of a certain group may affect, for example, its interaction with a receptor, and such changes would be difficult to study with properties related to functional groups.

For these reasons, Ghose and Crippen[26−28] have developed several sets of hydrophobicity parameters referred to atoms. According to these authors, the partition coefficient of a molecule with **k** atoms can be expressed as

$$\log P = \sum_{i=1}^{k} a_i \qquad (3)$$

where **k** is the total number of atoms in the molecule and a_i is the lipophilic contribution of a certain "atom type". In a first approach,[26] these parameters a_i were defined for C, H, O, N, F, Cl, Br, I, and S in different environments according to both the hybridization and the nature of the bonds and atoms around them. Later, those parameters were extended to other atoms as Se and P and to other oxidation states for S.[27,28]

Thus, by using the atomic hydrophobic factors (a_i) first proposed by Ghose and Crippen[26] and the molecular lipophilic potential (MLP) as defined by Audry (eq 2), Furet et al. proposed the following equation to define an empirical molecular lipophilic potential (MLP_a) depending on atoms not on functional groups

$$\mathrm{MLP_a} = \sum_{i=1}^{k} \frac{a_i}{1 + d_i} \qquad (4)$$

where *k* is the total number of atoms in the molecule, a_i is

Table 1. Calculated log *P* Values Found for Compounds **1**−**12**

compd	log P_{calc}
pyrazole (**1**)	0.529
4-methylpyrazole (**2**)	1.206
4-ethylpyrazole (**3**)	1.668
4-propylpyrazole (**4**)	2.130
4-(1-methylethyl)pyrazole (**5**)	2.017
4-butylpyrazole (**6**)	2.592
4-(1-methylpropyl)pyrazole (**7**)	2.479
4-(2-methylpropyl)pyrazole (**8**)	2.479
4-pentylpyrazole (**9**)	3.054
4-(1-methylbutyl)pyrazole (**10**)	2.940
4-(2-methylbutyl)pyrazole (**11**)	2.940
4-(3-methylbutyl)pyrazole (**12**)	2.940

the lipophilic factor of certain atom type, and d_i is the distance (in Å) between the point of the VDWS where the MLP_a is calculated and the "*i*" atom of the molecule. In order to compute this MLP_a on the van der Waals surface (VDWS), we have developed a simple program that calculates a value of MLP_a (according to eq 4) in each point of a distribution on the VDWS.

Van der Waals surfaces are continuous surfaces defined as the superposition of atomic spheres which correspond to the atoms of the molecule. Thus, to get a relatively isotropic distribution of points over this VDWS the MS program of Connolly[29] was used.

Hence, the calculated log *P* and the MLP_a on the VDWS (according to eqs 3 and 4, respectively) were estimated for pyrazole **1** and for all the local and global minima found, in the previously mentioned paper,[16] for compounds **2**−**12**. The results of the topological analysis of the interrelation between MLP_a and VDWS for these structures will be discussed in the next section.

Regarding the calculated log *P* values, the results for the pyrazoles studied are shown in Table 1. The only experimental log *P* found for this set of compounds was that of pyrazole (**1**). The log *P* value reported by Hansch and Leo for this compound[25] was 0.26 that is rather smaller than the calculated one (0.53). Thus, these calculated log *P*'s should be considered with a certain care since, even though they give a good approximation to the experimental behavior, they do not reproduce totally well those values.

In Table 1 it can be observed that the calculated log *P* values increase as the number of C atoms in the alkyl chain increases. Additionally, and as expected, a linear chain is more lipophilic than a branched one in alkyl chains with equal number of C atoms. Moreover, calculated log *P* values are not affected by the position of the methyl group introduced in the alkyl chain as a branch. This occurs because the a_i hydrophobic factors, used to calculate log *P*, only reflect the nature of the atom considered and the nature of the atoms directly bonded to this one. That is, these factors do not reflect the different spatial dispositions of these atoms. Thus, calculating log *P* with the equation proposed by Ghose and Crippen does not allow to distinguish between branched isomers.

For this reason, this particular way to calculate the log *P* is not sufficient to explain the relation between the lipophilicity of compounds **1**−**12** and their inhibitory activity against LADH. Therefore, a deeper analysis is required to consider not only the lipophilicity of these molecules in a global way but also the particular characteristics of their structure or even their conformation. Such an analysis will be provided by the topological study of the interrelation between the MLP_a and the VDWSs.

SHAPE CHARACTERIZATION OF MOLECULAR LIPOPHILIC POTENTIAL AND VAN DER WAALS SURFACE INTERRELATIONS FOR 4-ALKYLPYRAZOLES

The inhibitory power of 4-alkylpyrazoles increases when increasing the length of the alkyl chain, this means when increasing the degree of conformational freedom. For this reason, in a previous paper[16] a systematic conformational search was carried out for compounds **2**−**12** to localize all the possible low-energy conformers (a total of 234 local and global minima for the 11 compounds). Then, the VDWSs of all these different minima were characterized by using shape analysis, and only those conformers with different shape descriptors were chosen (the number of conformers to study was then reduced to 76). These 76 conformations, local or global minima in energy, with different shape corresponding to the 11 compounds studied (**2**−**12**), were optimized by using the PM3 semiempirical program. Pyrazole (**1**) structure was built using standard distances and angles and optimized with the PM3 semiempirical program.

Surfaces combining two properties as the MLP maps (lipophilic properties) and VDWSs (steric properties) allow for three-dimensional representation of the drugs regarding both the shape and the interrelations between the surface and molecular properties. This is an essential consideration when evaluating the geometric, lipophilic, and electronic properties of the drug−receptor interaction.

The method used to characterize the interrelation of molecular properties (e.g., MEP and MLP) with the VDWS has been described extensively in the literature.[12−14,30,31] However, included is a brief explanation of the main concepts and parameters that have been used in this work.

Calculation of MLP_a for a distribution of points on a VDWS (as given by the MS program[29]) was performed using eq 4 and the first set of parameters proposed by Ghose and Crippen.[26] While the visual analysis of MLP_a distributions on VDWS within desired ranges was performed with the molecular modeling program InsightII,[32] the topological study of MLP_a-VDWS interrelations was carried out analytically with a program we developed.[33] This analysis provided the characterization of the MLP_a function on a VDWS by means of certain topological invariants.[34]

The VDWS of a molecule, in a certain conformation **K**, is defined as the cover surface $\mathbf{G}(\mathbf{K},\rho)$ resulting from the superposition of atomic spheres, where $\rho = (\rho_1, \rho_2, ..., \rho_N)$ is the set of van der Waals atomic radii. Considering a range of MLP_a below a given value $\mathbf{L_j}$, a subset $\mathbf{D_j}(\mathbf{L_j})$ of the total molecular surface $\mathbf{G}(\mathbf{K},\rho)$ containing the points associated with that MLP_a range, is defined. This $\mathbf{D_j}(\mathbf{L_j})$ surface can be considered derived from the total $\mathbf{G}(\mathbf{K},\rho)$ surface by a truncation (i.e., by separating all the points on the VDWS with MLP_a value above $\mathbf{L_j}$) and can be characterized by different topological invariants.[34] For a systematic search, $\mathbf{L_j}$ can be varied in equidistant ranges of potential. In the present study these truncated surfaces were identified by Euler−Poincaré characteristics $\chi_j(D_j)$. The MLP_a-VDWS interrelation, then, can be described by $\chi(K,d,\rho)$ which

Table 2. The χ_j Shape Descriptors of the Interrelation between MLP_a and VDWS for 4-Methylpyrazole (**2**) and the Corresponding δ_j Codes[a]

MLP_a	χ_j	δ_j
max = 0.3955		
0.3905	(0)	0
0.3855	(−3)	−3
0.3805	(1,1,1,1)	4
<u>0.3755</u>	(1,1,1,1,1)	<u>5</u>
0.3705	(1,1,1,1,1)	5
0.3655	(1,1,1,1)	4
0.3605	(1,1,1,1)	4
0.3555	(1)	1
0.2755	(1,1)	2
0.2705	(1,1)	2
0.2655	(1,1)	2
0.2605	(1,1)	2
0.2555	(1)	1
min. = 0.1443	(Ø)	

[a] The maximum δ_j code (δ_{max}) and the corresponding MLP_a range appear underlined.

depends on the width **d** of those intervals, and the VDW's radii ρ

$$\chi(K,d,\rho) = (\chi_1, \chi_2, ..., \chi_i, ..., \chi_k) \qquad (5)$$

where χ_j is the Euler−Poincaré characteristic of each truncated surface $\mathbf{D_j(L_j)}$, which is numerically equal to two minus the number of discontinuities or truncations on that surface. These vectors $\chi(K,d,\rho)$ allow for the comparison between different conformations or different molecules with given configurations. In the present work, the topological analysis of the different local minima obtained for each pyrazole derivative was carried out using Gavezotti radii[35] as the ρ set and a width **d** of 0.005 units of MLP in all cases.

The program that performs this analysis starts calculating the Euler−Poincaré characteristics χ_j of the truncated surface after removing those points within a MLP_a interval of 0.005 units below the maximum MLP_a value. For example, in the case of 4-methylpyrazole (**2**) this range will be between 0.3955 and 0.3905 units of potential, and after removing the points within this range the remaining surface will have two holes, therefore the corresponding Euler−Poincaré characteristic will be χ_j: $2-2 = (0)$ (see Table 2). Then, the program continues analyzing topologically the surface that remains after removing those points with a MLP_a value within 0.010 units below the maximum MLP_a value. In the mentioned case of compound **2**, the resulting truncated surface will present five holes and therefore χ_j: $2-5 = (-3)$ (see Table 2). The last step will be the analysis of the surface remaining after truncating all those points associated with a MLP_a interval between the maximum and minimum values of MLP_a (i.e., the total surface), that is the empty set that has an Euler−Poincaré characteristic of χ_j: (Ø).

In the case of the global minimum of 4-methylpyrazole (**2**) the total descriptor $\chi_j(D_j)$ for the truncated VDWSs is given in Table 2, and the surfaces associated with some of the different MLP_a ranges are shown in Figure 2. The black triangles and gray stars represent clusters of points on the Connolly surface within and out of the MLP_a interval considered, respectively. The graphical representations were carried out by using the InsightII package,[32] and the graphics here presented are a simplification of those obtained in the screen.

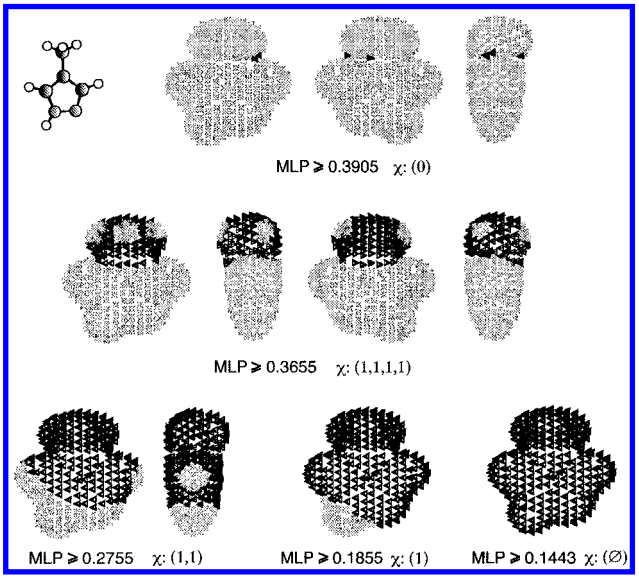

Figure 2. Graphical representation of the interrelation MLP_a-VDWS. Black triangles and gray stars, respectively, represent clusters of points on the Connolly surface with MLP_a values within and out of the potential range considered. Each surface is associated with a range of potential between the maximum of MLP_a and the values represented in the figure. The corresponding Euler−Poincaré characteristic of the remaining surface after removing the points within the range considered appears under each associated surface. Upper part: views $+x$, $-x$, and $-y$; middle part: views $+x$, $-y$, $-x$, and $+y$; lower part: views $+x$ and $-y$, view $+x$, and view $+x$.

The topological analysis of the MLP_a-VDWS interrelations for the 77 different local minima found for compounds **1**−**12** was carried out in the same analytical way. A width **d** of 0.005 units of potential was used for the analysis of all the minima with different shape.

CORRELATION OF THE SHAPE CODES AND THE INHIBITORY ACTIVITY OF 4-ALKYLPYRAZOLES

In a previous work[16] an almost linear relationship was found between maximum and minimum MEP values on the VDWS of substituted pyrazoles. Two different clusters were formed. On the one hand, those pyrazoles with high inhibitory power presented the most negative MEP minima and lower MEP maxima, while, on the other hand, low power inhibitors exhibited less negative MEP minima and higher MEP maxima.

In the present study and for the 77 conformations studied (corresponding to compounds **1**−**12**), a linear relationship between MLP_a minima and MLP_a maxima was found (see Figure 3). It can be observed that as the number of C atoms of the alkyl chain increases, the value of the maxima and minima of MLP_a increases. Pyrazole derivatives with larger maximum and minimum MLP_a values are those with higher inhibitory activity, which confirms the important role of lipophilicity in the inhibition of LADH.

However, for branched compounds with an equal number of C atoms the relationship between maximum MLP_a/minimum MLP_a and the biological activity is not clear. For this reason, some "shape codes" obtained from the topological analysis of MLP_a-VDWS interrelations are needed in order to find a correlation between lipophilicity and the inhibitory activity.

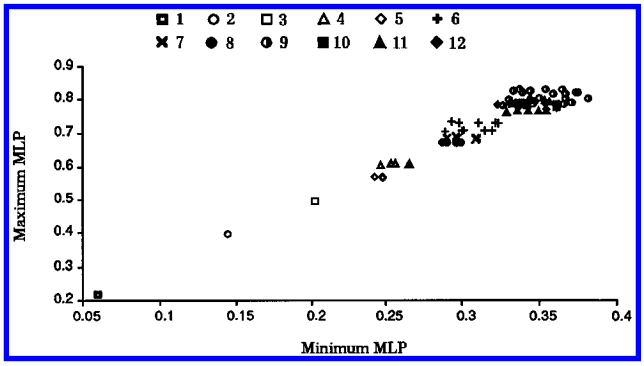

Figure 3. Representation of the maximum *vs* the minimum MLP_a for all the local minima (same symbol) of each pyrazole studied.

The parameter δ_j was previously defined[15] as the sum of Euler−Poincaré characteristics

$$\delta_j = \sum_{i=1}^{n_j} \chi_{ji} \qquad (6)$$

Thus, for the conformation of minimum energy of the 4-methylpyrazole (**2**) we obtain a δ_j value for each interval of MLP_a considered (see Table 2), and the maximum δ_j value obtained ($\delta_j = 5$) appears at a MLP_a value of 0.3755. This maximum δ_j parameter represents the maximum number of truncations that a 4-alkylpyrazole can show in a certain conformation. Therefore, each conformer can be represented by the MLP_a value in which the maximum δ_j appears: MLP-(δ_j). In the case of the minima of 4-methylpyrazole (**2**) the representative MLP(δ_j) code will be 0.3755 (see Table 2). The MLP(δ_j) values obtained for every minimum for every pyrazole derivative studied are gathered in Table 3.

For a certain 4-alkylpyrazole derivative, that conformation with a more extended chain (more H atoms exposed, more truncations in the surface) would be the one that better characterizes the hydrophobic properties of the molecule. For that reason, for compounds with more than one minimum (**3**−**12**), we could represent this 4-alkylpyrazole by using the largest value of the MLP(δ_j) code (largest hydrophobic contribution), with larger δ_j (larger number of truncation, therefore more extended disposition of the chain), obtained from within all the local minima of that 4-alkylpyrazole. From now on, such a code will be referred to as $MLP(\delta_j)_{max}$ (see underlined values in Table 3 and see, as well, Table 4).

Thus, comparing this code representing each pyrazole it is observed that $MLP(\delta_j)_{max}$ increases as the length of the alkyl chain increases (see Table 3). On the contrary, branching of the chain decreases the value of $MLP(\delta_j)_{max}$ compared to the linear chain derivative with the same number of C atoms. Both observations are in agreement with the hydrophobic character of linear and branched alkyl chains.

For pyrazole and its linear derivatives (compounds **1**, **2**, **3**, **4**, **6**, and **9**) a good correlation was found between MLP-$(\delta_j)_{max}$ and the ln(K_I) as a measure of the activity ($\ln(K_I) = -0.117 - 10.196\ MLP(\delta_j)_{max}$; $r^2 = 0.978$; sd = 0.104; $F = 180.742$; $t = -13.444$). Cross-validation is a statistical technique extensively used on the computation of quantitative structure−activity relationships (QSAR). Although the number of derivatives in this sample is small to develop a quantitative relationship, cross-validated statistics will provide a better measurement of the quality of the correlation between $MLP(\delta_j)_{max}$ and ln(K_I). Thus, cross-validations have been carried out for derivatives **1**, **2**, **3**, **4**, **6**, and **9** by omitting one of them each time and predicting the corresponding ln-(K_I) from the equation obtained in each case.

The most widely used metric for evaluating the accuracy of the QSAR prediction is the value of the squared of the regression coefficient, r^2 (defined as $(SD_{original} - SD_{remaining\ error})/SD_{original}$), and this parameter has been suggested[36] to be named q^2 in the case of cross-validated statistics. In the case of the pyrazoles studied here $q^2 = 0.920$. This is in agreement with what was previously observed[16] for the interrelations between molecular electrostatic potential (MEP) and VDWSs for this family of compounds.

Yet, in the case of branched 4-alkylpyrazoles (compounds **5**, **7**, **8**, **10**, **11**, and **12**) there is no a linear relationship with their activity (see Table 4). Activity of compounds **5**, **7**, and **8** only can be explained in relation to their linear isomers. Thus, for a pyrazole with a chain with three C atoms, the branched derivative (**5**) will be less active than the linear one (**4**) as it is reflected in the smaller value found for MLP-$(\delta_j)_{max}$ (K_I (**4**): 0.004 *vs* K_I (**5**): 0.008; $MLP(\delta_j)_{max}$ (**4**): 0.550 *vs* $MLP(\delta_j)_{max}$ (**5**): 0.500). In the same way, one can expect a lower activity for the branched derivatives **7** and **8** than for the linear isomer **6** by looking at the smaller values of $MLP(\delta_j)_{max}$ found in both **7** and **8** than for **6**. That is, although the model is not quantitative for branched chains, it provides a general tendency for the activity thereby establishing a relative order of K_I for 4-alkylpyrazoles.

Thus, according to Table 4, derivatives **10**, **11**, and **12**, for which K_I was previously unknown, were predicted to have an inhibitory power smaller than the linear isomer (**9**). This prediction is in agreement with the MEP-VDWS previous results[16] and was confirmed for the 4-isopentyl derivative **12**. This 4-(3-methylbutyl)pyrazole (**12**) was synthesized, and its inhibitory activity was measured in our laboratory ($K_I = 0.002\ \mu M$)[37] following the protocol established in ref 21.

CONCLUSIONS

Although hydrophobic properties are very difficult to quantify, empirical approaches, combining shape, and lipophilicity parameters provide a good idea of this effect. Surfaces combining two properties as the MLP_a maps (lipophilic properties) and VDWSs (steric properties) allow a deeper understanding not only of the characteristics of biomolecules but also of the nature of the interaction with the corresponding receptor, than those surfaces representing only one property.

Thus, the topological characterization of the MLP_a-VDWS interrelations on a series of 4-alkylpyrazoles (by analytical and graphical methods) yielded certain shape codes that represent a three-dimensional view of the lipophilicity of the molecules and correlate well with the inhibitory activity. In all the cases studied, it was observed that the minimum MLP_a is located on the H atom bonded to the pyrazolic N atom (hydrophilic area). The maximum MLP_a value (lipophilic area) was always located on the atoms at the end of the alkyl chain, and, when a branch was introduced, the maximum appears there as well.

The $MLP(\delta_j)_{max}$ code represents the lipophilicity of the conformer of each 4-alkylpyrazole which has more H atoms

Table 3. MLP_a Values at Which the Maxima δ_j Parameters Were Found for Every Minima of Compounds **1**−**12**[a]

comp	MLP(δ_j)	comp	MLP(δ_j)	comp	MLP(δ_j)	comp	MLP(δ_j)
1	0.167 (3)	**7**:119	0.615 (6)	**9**:8207	0.720 (6)	**11**:3396	0.675 (8)
2:12	0.375 (5)	**7**:147	0.605 (5)	**9**:8228	0.725 (5)	**11**:3402	0.675 (7)
3:28	0.446 (5)	**7**:1491	0.605 (5)	**9**:11780	0.695 (6)	**11**:8183	0.680 (7)
4:45	0.555 (6)	**7**:1862	0.605 (6)	**9**:11809	0.690 (6)	**11**:8204	0.675 (7)
4:66	0.545 (6)	**7**:2177	0.600 (5)	**9**:11980	0.715 (5)	**11**:11956	0.680 (6)
4:164	0.545 (6)	**8**:311	0.580 (5)	**9**:12001	0.680 (6)	**11**:11971	0.670 (7)
4:241	0.550 (7)	**8**:486	0.610 (5)	**9**:15210	0.685 (6)	**11**:11977	0.670 (7)
4:311	0.550 (6)	**8**:1172	0.575 (6)	**9**:15235	0.685 (7)	**11**:15386	0.680 (7)
5:24	0.500 (9)	**8**:1711	0.610 (5)	**9**:15410	0.715 (5)	**11**:15407	0.675 (5)
5:49	0.500 (8)	**8**:2201	0.575 (5)	**9**:15431	0.720 (5)	**12**:1344	0.685 (6)
6:315	0.635 (5)	**9**:2176	0.700 (6)	**10**:833	0.680 (4)	**12**:2177	0.680 (6)
6:343	0.635 (5)	**9**:2201	0.700 (6)	**10**:1001	0.645 (5)	**12**:3206	0.695 (5)
6:462	0.620 (5)	**9**:2380	0.710 (5)	**10**:1029	0.685 (4)	**12**:5799	0.680 (6)
6:490	0.625 (6)	**9**:2397	0.715 (6)	**10**:2352	0.675 (6)	**12**:8008	0.695 (5)
6:1148	0.625 (6)	**9**:3205	0.685 (5)	**10**:3381	0.685 (6)	**12**:11977	0.675 (5)
6:1176	0.625 (6)	**9**:3234	0.695 (5)	**10**:10437	0.675 (5)	**12**:15211	0.645 (6)
6:1687	0.620 (5)	**9**:3409	0.675 (6)	**10**:13006	0.640 (5)	**12**:15746	0.685 (5)
6:1715	0.620 (6)	**9**:3430	0.715 (5)	**10**:13034	0.675 (5)		
6:2177	0.620 (6)	**9**:8007	0.710 (5)	**10**:15239	0.645 (5)		
6:2205	0.620 (6)	**9**:8036	0.690 (7)	**11**:2030	0.660 (5)		

[a] Underlined is shown the largest MLP_a value for the largest of the δ_j maxima for each pyrazole derivative. The notation for each minima corresponds to **compound number**:conformer number (local or global minima).

Table 4. Largest MLP_a Values at Which the Largest of the δ_j Maxima for the Maxima δ_j Parameters Were Found for Every Minima of Compounds **1**−**12** (values underlined in Table 3), and the Inhibitory Constants K_I (μM) for Each Pyrazole Derivative

compd	$MLP(\delta_j)_{max}$	K_I
1	0.167 (3)	0.22
2	0.375 (5)	0.013
3	0.446 (5)	0.007
4	0.550 (7)	0.004
5	0.500 (9)	0.008
6	0.625 (6)	0.0018
7	0.615 (6)	0.014
8	0.575 (6)	0.013
9	0.690 (7)	0.0008
10	0.685 (6)	
11	0.675 (8)	
12	0.685 (6)	

exposed, meaning a more extended chain and more interaction with the solvent. Accordingly, the alkyl chain of any 4-alkylpyrazole should be as extended as possible for a better interaction with the enzyme since this improves the inhibitory activity. Nonbranched 4-alkyl derivatives showed a linear correlation between the $MLP(\delta_j)_{max}$ code and the K_I against LADH. This fact was in agreement with previous results.[16]

The MLP_a-VDWS interrelation provides a three-dimensional view of the lipophilicity of a molecule. In this sense, the $MLP(\delta_j)_{max}$ code of the 4-isopentylpyrazole (**12**) was calculated, and its activity was predicted. Then, this compound was synthesized, and its inhibitory activity was measured (K_I(**12**) = 0.002 μM), being lower than the linear derivative **9** (4-pentylpyrazole, K_I = 0.0008 μM) as had been predicted by the calculations.

In the case here studied of pyrazoles, lipophilic and electronic effects seem to have the same effect on the inhibitory activity of these compounds, and, therefore, both approaches MEP-VDWS and MLP_a-VDWS provided similar results. However, there are systems where lipophilicity plays the most important role in the activity and for that reason this new approach to the study of lipophilicity can be applied to any other family of compounds and explain how lipophilicity and shape influence their biological activity.

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