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Short communication

Quantitative structure—retention and retention—activity relationships of some 1,3-oxazolidine systems by RP-HPTLC and PCA

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Abstract

A quantitative structure–retention and retention–activity relationships investigations were performed on the lipophilicities of some 1,3-oxazolidine systems as estimated by RP-HPTLC retention parameters. The classical R_{Mo} values were compared with the factors scores obtained by principal component analysis based also onto the TLC retention data. The lipophilicities (R_{Mo} and factor scores) were correlated with the theoretical molecular descriptors of 1,3-oxazolidine derivatives providing by the ALCHEMY 2000 software package. The reliability of the factor scores values as lipophilic indices are shown by their significant correlation with the classical R_{Mo} values and other molecular descriptors. In addition, the "lipophilicity chart" described by the first two components, and/or the "lipophilicity space" described by the first three components have the effect of separating compounds from each other most effectively from the congeneric (similarity) aspect point of view. Finally, these findings support the idea that the chromatographic process of the investigated compounds in this paper and consequently their partitioning over a bio-membrane are controlled mainly by lipophilicity.

Keywords: QSRR; QRAR; PCA; Lipophilicity; Molecular descriptors; 1,3-Oxazolidine

1. Introduction

The 1,3-oxazolidine systems find important applications as platicizers, biocides [1–4] and pesticides [5]. Recently, azaoxaspiranes having an oxazolidine ring have been considered prodrugs acting as delivery systems because of their increased lipophilicity compared with that of the corresponding free β-amino-alcohol [3]. Lipophilicity plays a vital role in physicochemical, environmental and biological processes as it determines transport phenomena in vivo such as through the blood-brain membrane barrier.

The synthesis and structural aspects of two new series of 1,3-oxazolidine derivatives considered in this paper have formed the subject of recent investigations [6,7].

Quantitative structure–activity relations (QSAR) describe how the molecular structure, in terms of descriptors—lipophilic, electronic and steric—affects the biological activity of a compound [8–11].

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Similarly, quantitative structure–retention relations (QSRR) relate these descriptors to chromatographic retention. Finally, the quantitative retention–activity relations (QRAR) imply that conclusions concerning biological activity can be based on chromatographic experiments [12–16]. In context, it is considered that the same basic intermolecular actions determine the behavior of chemical compounds in both biological and chromatographic environments. As a consequence, the chromatographic approach has been quite successful in duplicating $\log P$ data derived by traditional "shake-flask" technique or other procedures [17–22]. The relations themselves are usually based on correlation analysis. For instance, the use of R_M values, obtained from various types reversed-phase thin layer chromatography, is based on the assumed linear relationship between the molecular parameter (1) and $\log P$

$$R_M = \log\left(\frac{1}{R_F} - 1\right) \quad (1)$$

The advantages of TLC methods consist in the very small amounts of sample needed for the estimation and the less strict requirement of purity because the impurities separate during the chromatographic process. They are rapid and relatively simply, low cost and easy to perform. In addition, we have to stress the dynamic aspect of the chromatographic process and the wide choice of stationary phases and developing solvents.

The R_M value (related to the molecular lipophilicity), determined by using of RP-HPTLC, generally, depends linearly on the concentration of the organic component of the mobile phase:

$$R_M = R_{Mo} + bC \quad (2)$$

where R_M values were calculated using Eq. (1) and C is the concentration of organic modifier.

Another useful form of computational analysis for the correlation of biological activity, structure, and chromatographic retention is principal component analysis (PCA) [23–32]. By using the multidimensional space described by the different mobile phases, a quantitative model is derived that transforms the axes of the system. The first principal component (PC1) defines as much of the variation in the retention data as possible. The second principal component (PC2) describes the maximum amount of residual variation after the first PC has been taken into consideration, and so on. By using only a limited number of PCs, the dimensionality of the retention data space is reduced, thereby simplifying further analysis. In chromatography two or three principal components are often sufficient to describe most of the retention data variation. Although the PCs are abstract, one of them often shows high correlation with lipophilicity, molecular size, or steric factors, whereas the other PC seems to be more strongly correlated with dipole–dipole interactions and electronic factors.

The purpose of this QSAR/QSRR study is to investigate the feasibility of the scores, obtained by PCA using RP-HPTLC retention data, as a measure of lipophilicity in correlation with partition coefficient ($\log P$) and other descriptors in the case of two new series of 1,3-oxazolidine systems. In addition, the scatterplots of the scores onto plane described by the first two components appear to be very useful having the effect of separating compounds one from each other most effectively, obtaining in this way the "congeneric lipophilicity chart" of the series; 3D scatterplots giving a "congeneric lipophilicity space".

2. Experimental

The chromatographic behavior of the 1,3-oxazolidine derivatives depicted in Fig. 1 was studied on the C_{18} silica gel bonded plates. RP-HPTLC plates (20 cm × 20 cm) were obtained as a gift from Macherey-Nagel (Düren, Germany). Methanol for chromatography was supplied from "Reactivul" (Bucharest, Romania). 3 μl in duplicate of each solution in methanol (1 mg ml^{-1}) was spotted to origin of the plate by hand. Chromatography was performed in a normal developing chamber at room temperature, the developing distance being 8 cm. Methanol was used as the organic modifier of the mobile phase in the concentration range 50–70% (v/v) in steps of 5%, as the studied 1,3-oxazolidine compounds differed considerably in their retention; very well-defined spots were also obtained for all the observed compounds. However, we have to mention that the compound (**8**) has not been identified being presumably the least lipophilic. In contrast, compound (**16**) which has been moved only for the ratio methanol–water (70:30, v/v) and as a consequence it appeared to be the most lipophilic of the two series considered.

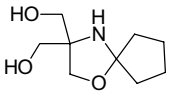
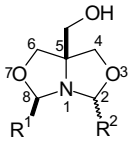
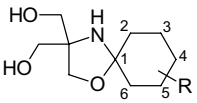
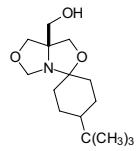
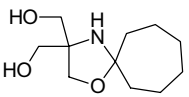
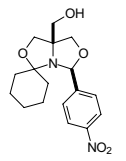
Series I			Series II			
Chemical structure	No.	Substituent, R	Chemical structure	No.	R^1	R^2
	**1**	Without		**8**	H	H
				9	C_6H_5	C_6H_5
				10	*p*-O_2N- C_6H_4 (*cis*)	*p*-O_2N- C_6H_4(*trans*)
				11		
				12		
	2	H		**13**		
	**3**	4-CH_3		**14**		C_6H_5-CO
	4	4-$C(CH_3)_3$		**15**	C_6H_5-CO	$CH_2C(C_6H_5)_3$
				16	$CH_2C(C_6H_5)_3$	*p*-O_2N- C_6H_4
	5	5- CH_3		**17**	H	
	6	6- CH_3		**18**	-	-
	**7**	Without				
				**19**	-	-

Fig. 1. The chemical structure of the investigated 1,3-oxazolidine derivatives.

After being developed, the dried plates were examined under UV lamp ($\lambda = 254$ nm).

3. Principal component analysis of TLC retention data

The PCA results obtained from the initial chromatographic data (R_F values) using covariance matrix (without autoscaling), considering only the proportion higher than one, suggested a significant three component model which can explain 99.71% of the total variance (information). The first component explains 93.02% of the total variance, the second 5.63% and the third only 1.06%; the subsequent eigenvalues are just sampling noise.

It is interesting also to mention that when the significance of the component model retained was tested applying the Bartlett's statistics [24], testing the hypothesis that (p–k) eigenvalues in variance–covariance matrix are equal, a model with three components was also selected.

4. Results and discussion

The results of regression analysis using Eq. (2) are compiled in Table 1. The statistics obtained (see also Table 1) illustrate that the linear equation fits in a very good way the experimental data, the linear model explaining over 97% of the total variance (see R^2 values) in the majority of cases. As usual, a good correlation has been also found between the R_{Mo} and b values of Eq. (2) as it is shown by the following linear relationship

$$R_{Mo} = -0.158 - 0.751b$$
$$(r = 0.971, n = 17, F = 251, s = 0.166) \qquad (3)$$

This finding indicates that the intercept R_{Mo} (lipophilicity) and slope b (specific hydrophobic surface area) for the majority of these compounds are high correlated and, in that case, they might form a homologous series of compounds as has been suggested by some authors [17–22]. Additionally, a significant correlation was obtained also between R_{Mo}

Table 1
Regression data and scores on the first three principal components of the 1,3-oxazolidine derivatives studied and the partition coefficient ($\log P$)

Compound	R_{Mo}	b	R^2	PC1	PC2	PC3	$\log P$
1	0.690	−1.220	0.888	−1.15	−0.18	−0.07	0.914
2	0.800	−1.200	0.951	−1.01	−0.18	−0.05	1.002
3	1.280	−1.840	0.970	−0.88	−0.22	−0.04	1.227
4	2.430	−3.200	0.982	−0.53	−0.24	−0.03	2.508
5	1.230	−1.780	0.986	−0.91	−0.22	−0.05	1.233
6	1.410	−2.130	0.994	−0.93	−0.26	−0.07	1.202
7	0.710	−0.940	0.999	−0.92	−0.15	−0.05	1.228
8	0.670[a]						0.763
9	2.830	−4.040	0.962	−0.65	−0.34	−0.03	2.620
10	2.580	−3.510	0.892	−0.56	−0.28	−0.14	2.292
11	1.250	−2.020	0.937	−1.05	−0.26	−0.03	1.651
12	1.390	−2.430	0.982	−1.17	−0.31	−0.06	1.331
13	1.510	−2.430	0.993	−1.03	−0.29	−0.07	1.376
14	2.210	−3.210	0.988	−0.76	−0.31	−0.05	2.434
15	2.040	−3.110	0.953	−0.89	−0.33	−0.04	1.990
16	7.668[a]						7.570
17	1.390	−2.380	0.984	−1.14	−0.29	−0.06	1.393
18	2.430	−3.230	0.995	−0.54	−0.25	−0.05	2.709
19	2.110	−2.580	0.978	−0.49	−0.19	−0.05	2.297

[a] Values estimated by using the prediction equation (Eq. (7)).

values and the scores of the same compounds on the first principal component as it is described by the linear equation (4)

$$R_{Mo} = 3.690 + 2.359\text{PC1}$$
$$(r = 0.801, n = 17, F = 27, s = 0.419) \quad (4)$$

Moreover, the single-parameter correlation equation (Eq. (4)) can be significantly improved by considering the scores corresponding to the first two components in a two-parameter equation (Eq. (5))

$$R_{Mo} = 1.888 + 2.315\text{PC1} - 6.976\text{PC2}$$
$$(r = 0.990, n = 17, F = 347, s = 0.102) \quad (5)$$

The correlation of RP-HPTLC retention parameters with other molecular descriptors available in the ALCHEMY 2000 programs [33] has been also computed. The partition coefficient ($\log P$), the first-order ($^1\chi$) and the third-order ($^3\chi$) connectivity index, the zero-order ($^0\chi^v$) and the first-order ($^1\chi^v$) valence order connectivity index, the third-order shape index for molecule ($^3K_\alpha$), the Wiener (W) index based on the graph of the molecule, and also the molecular polarizability (MP) were calculated for all compounds by means of the QSAR option of ALCHEMY 2000. The other descriptors: surface area (SA), volume (V), ovality (Ov), dipole moment (DM), and sum of absolute charges (SAC), respectively, formed the output of SciLogP option of the molecular modeling computer programs ALCHEMY 2000. The values obtained are presented in Table 2.

An examination of correlation data revealed that the partition coefficient ($\log P$) is the most significant contributory factor for the lipophilicity (estimated by R_{Mo}) of the compounds studied in this work, besides to majority of other descriptors, and with a lower contribution the dipole moment and the sum of the absolute charges. It was interesting also to observe that the majority of the computed descriptors are highly correlated for these series of compounds. As a consequence the single-parameter correlation equation (Eq. (6)) is not significantly improved by including all the other descriptors as independent variables in a multiple regression model. By applying the well-known stepwise regression method, after the examination of each of the independent variable (descriptors in Table 2), only R_{Mo} was kept in the model

$$\log P = 0.289 + 0.866 R_{Mo}$$
$$(r = 0.943, n = 17, F = 122, s = 0.213) \quad (6)$$

Table 2
The descriptors computed for the 1,3-oxazolidine derivatives investigated in this paper

Compound	$^1\chi$	$^3\chi$	$^0\chi^v$	$^1\chi^v$	$^3K_\alpha$	W	MP	SA	V	Ov	DM	SAC
1	6.29	4.77	7.7	4.97	1.59	230	19	231	181	1.492	2.631	3.075
2	6.79	5.02	8.41	5.47	1.96	286	20.84	243	197	1.483	2.822	3.181
3	7.18	5.44	9.27	5.86	2.18	353	22.67	264	213	1.529	2.832	3.276
4	8.40	6.38	11.78	7.07	2.9	605	28.18	315	263	1.589	2.74	3.562
5	7.19	5.29	9.28	5.86	2.18	345	22.67	263	213	1.523	2.783	3.277
6	7.21	5.66	9.28	5.87	1.91	337	22.67	255	213	1.483	2.955	3.273
7	7.29	5.27	9.11	5.97	2.33	345	22.67	257	213	1.493	2.786	3.287
8	4.87	4.02	5.75	3.39	0.81	102	13.5	171	131	1.364	1.729	2.827
9	10.84	8.79	12.26	7.56	2.19	960	32.82	301	271	1.486	1.431	3.953
10	13.45	11.01	14.71	8.6	3.25	2034	37.29	366	318	1.623	3.092	4.722
11	10.84	8.79	12	7.26	2.16	960	31.4	296	263	1.493	7.8	4.202
12	10.84	8.79	12	7.28	2.16	960	31.4	312	266	1.564	4.802	4.325
13	10.84	8.79	12	7.26	2.16	960	31.4	312	266	1.559	5.633	4.227
14	9.84	8.29	12.4	8.29	1.98	722	31.86	305	258	1.557	3.739	3.704
15	12.66	10.42	14.08	8.47	2.8	1558	36.66	359	311	1.616	3.313	5.1
16	24.69	19.88	28.22	17.65	5.62	8242	78.8	638	620	1.815	1.466	6.788
17	9.160	7.54	10.23	5.99	2	699	25.39	274	226	1.527	3.222	3.775
18	8.920	7.68	12.44	7.52	2.04	657	29.24	322	271	1.593	1.786	3.516
19	11.60	9.76	13.56	8.52	2.47	1243	33.79	349	298	1.617	2.01	4.082

Furthermore, the results are not significantly changed by setting R_{Mo} as a dependent variable and keeping as independent variables all other descriptors in Table 2. The prediction equation (Eq. (7)) in that case has the following form:

$$R_{Mo} = -0.114 + 1.028 \log P \quad (r = 0.943, n = 17, F = 122, s = 0.232) \qquad (7)$$

It is also stimulating to observe that the correlation between the scores corresponding to the first two principal components and the partition coefficient $\log P$ values, considering a two-parameter equation, is absolutely the same with the correlation in the single-parameter equation (Eq. (6)) as it is indicated by the following linear multiple regression equation:

$$\log P = 2.492 + 2.251\mathrm{PC1} - 4.623\mathrm{PC2} \quad (r = 0.943, n = 17, F = 56, s = 0.222) \qquad (8)$$

By the stepwise regression analyses of the factor scores corresponding to PC1 and PC2 with the above mentioned descriptors the most dominant properties are found by the highest regression coefficients in Eqs. (9) and (10)

$$\mathrm{PC1} = -0.497 - 0.522\mathrm{SAC} + 0.170\,{}^3K_\alpha - 0.032\mathrm{DM} + 0.009V \quad (r = 0.986, n = 17, F = 60, s = 0.048) \qquad (9)$$

$$\mathrm{PC2} = -1.086 + 0.244\,{}^1\chi + 0.207\,{}^1\chi^v + 0.140 \log P - 0.095\mathrm{MP} + 0.025\mathrm{DM} \quad (r = 0.993, n = 17, F = 32, s = 0.120) \qquad (10)$$

The results suggest that the most important descriptors in PC1 are SAC and $^3K_\alpha$, i.e. the electronic parameter and the shape of the molecules and the most dominant feature in PC2 is the size and branching of the molecule ($^1\chi$, χ^v, $\log P$ and with a lower contribution MP and DM).

On the basis of these findings and from data provided in Table 1, the scores on the first principal components can be used efficiently besides the R_{Mo} values in the estimation experiments of the lipophilicity of these compounds directly from RP-HPTLC data or via $\log P$. In addition, as it is shown in Fig. 2, scores plots are very useful as a display tool for examining the relationships between compounds, looking for trends, groupings or outliers. Hence, graphing scores onto the plane described by PC1 and PC2 we obtain "the

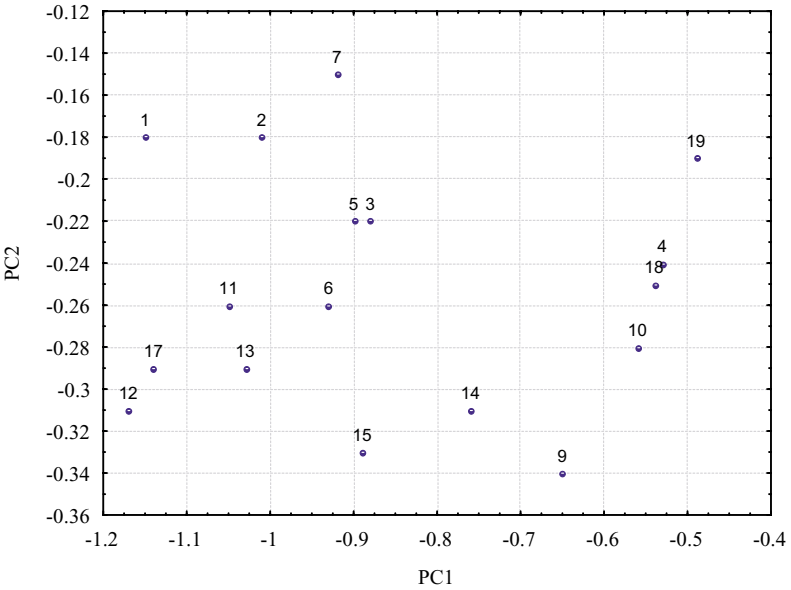


Fig. 2. PC1–PC2 score plot of the retention R_F values ("the congeneric lipophilicity chart").

congeneric lipophilicity chart". By a careful visual examination of the graph, it is possible to distinguish that the investigated compounds form practically three different congeneric classes, in a very good agreement with the selected eigenvectors when the Bartlett test was applied and with their chemical structure [34]. For example, compounds (**4**) and (**18**) are very similar because the lipophilic part of their molecule is the same and the difference between compound (**6**), and its congeners (**3**) and (**5**) by the one side, and (**11**) and its congeners (**12**) and (**13**) by the other side, can be explained by steric effects. These conclusions are also well supported by "the congeneric lipohilicity space" obtained by graphing the scores corresponding to the first three principal components (Fig. 3).

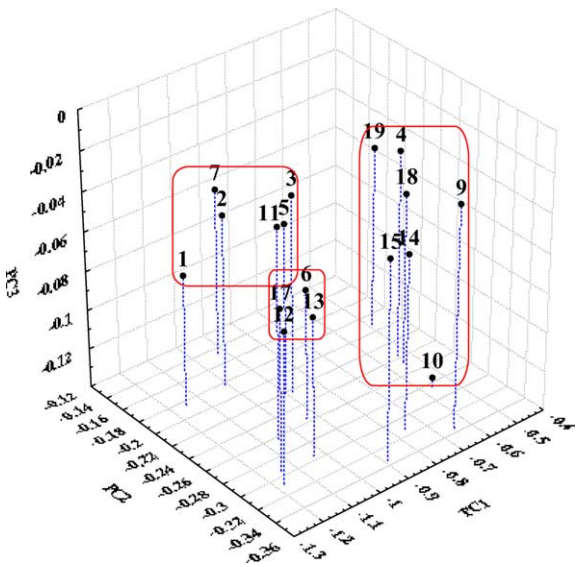


Fig. 3. The 3D score plot of the retention R_F values ("the congeneric lipophilicity space").

5. Conclusions

The lipophilic character of some 1,3-oxazolidine derivatives was studied by means of reversed thin layer chromatography using a mixture of methanol–water as the solvent system. The significant correlation between the R_{Mo} values and *b*-slopes (specific hydrophobic surface areas) indicate that these new two series of 1,3-oxazolidine derivatives could be considered as a homologous series of compounds independently of their structural heterogeneity as so far it was considered. The reliability of the factor scores values as lipophilicity indices is shown by their significant correlation with the classical R_{Mo} values. In addition, the

"lipophilicity chart" described by the first two components had the effect of separating compounds from each other most effectively from the congeneric (similarity) aspect point of view. By a careful visual examination of the graphs, it is possible to distinguish that the investigated compounds form practically three different congeneric classes, in a very good agreement with the selected eigenvectors when the Bartlett test was applied and with their chemical structure. Much more, it appears clearly that a rational interpretation of the factor scores based on the retention data could offer new insights concerning the chromatographic mechanism (QSRR) and the partitioning process over a bio-membrane (QSAR). Finally, these findings support the idea that the chromatographic process of these compounds and consequently their partitioning over a bio-membrane are controlled mainly by lipophilicity.

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