ACADEMIA ROMÂNĂ
Revue Roumaine de Chimie
http://web.icf.ro/rrch/

Rev. Roum. Chim.,
2015, *60*(7-8), 677-687

Dedicated to Professor Valer Farcasan
on the occasion of his 95th anniversary

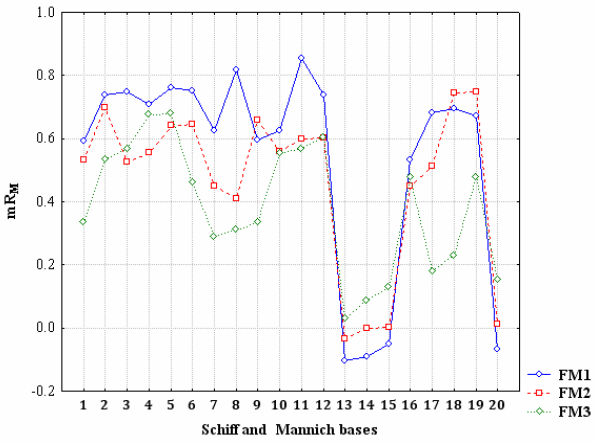
HETEROCYCLES 37. LIPOPHILICITY OF NEW POLYHETEROCYCLIC SCHIFF BASES AND MANNICH BASES ESTIMATED BY THIN-LAYER CHROMATOGRAPHY AND COMPUTATIONAL METHODS

Alexandra TOMA,[a] Denisa LEONTE,[a] Valentin ZAHARIA,[a] Costel SÂRBU[b] and Dorina CASONI[b,*]

[a] Department of Organic Chemistry, Iuliu Haţieganu University of Medicine and Pharmacy, 41 Victor Babeş, Ro-400012 Cluj-Napoca, Roumania
[b] Department of Chemistry, Faculty of Chemistry and Chemical Engineering, Babeş-Bolyai University, Cluj-Napoca, 11 Arany Janos, Ro-400028 Cluj-Napoca, Roumania

Received January, 30, 2015

Lipophilicity of novel potentially anti-inflammatory 1,2,4-triazole derivatives (Schiff bases and Mannich bases) has been estimated by thin-layer chromatographic method (TLC). The chromatographic retention was measured using phosphate buffer as aqueous component and methanol respectively methanol containing 10% of different upper alcohol (1-butanol and 1-octanol) as the hydrophobic additive of the mobile phase. Different experimental lipophilicity indices (denoted by mR_M, R_{M0} and $PC1/R_M$) were estimated using retention parameters in all cases. Various partition coefficients (log P) values were calculated by means of different software and further correlated with the experimental indices determined using the organic–aqueous eluent system and systems modified with hydrophobic alcohol addition. It was found that values measured in eluent system modified with 1-butanol better correlate with computed lipophilicity parameters. Furthermore, by applying the principal component analysis (PCA) on the experimental values, the similarity and differences of compounds from the lipophilicity point of view were highlighted.



INTRODUCTION

Heterocyclic ring systems can be commonly found in the structure of many compounds of medicinal interest, presenting a diverse array of biological activity. In particular, Schiff bases and Mannich bases derived from 1,2,4-triazole are important classes of pharmacologically and chemically useful compounds due to their therapeutic potential and to the reactivity of their functional groups. These compounds were recently reported as potent anticancer,[1,2] antimicrobial,[3,4] anti-inflammatory and analgesic agents.[5,6] Moreover polyheterocyclic Schiff bases and

* Corresponding author: casoni_dorina@yahoo.com

Mannich bases are becoming even more important in medicinal research. Consequently sorting out new chemical entities with inappropriate absorption, distribution, metabolism and excretion behaviour at an early stage of drug discovery is a major challenge in pharmaceutical profiling. In this area an accepted strategy to predict absorption of a drug candidate is the measurement of lipophilicity which is directly related to permeability and fraction absorbed. In biological systems lipophilicity largely determines the solubility of drugs in biological fluids, penetration through the biological membranes, rate of absorption, affinity to plasma and tissue proteins, distribution into the specific body compartments or accumulation in the body.[7,8]

For lipophilicity assessment, partition chromatographic techniques, particularly reversed-phase HPLC and reversed-phase TLC offer several practical advantages compared to the traditional shake-flask method, including speed, reproducibility, broader dynamic range, insensitivity to impurities or degradation products and reduced sample handling and sample sizes.[9] Many studies have demonstrated that the chromatographic retention behaviour of a molecule can be used as a criterion of the molecule lipophilicity and biological activity.[10,11] In this area some experimental strategies to improve lipophilicity assessment have been proposed.[12-14] Also in the HPLC separations the mobile phase composition, stationary phase nature, compound structure, the injection volume and injection solvent nature has been recently investigated[15-19] and it is now generally accepted that the mobile phase plays a dominant role in the retention process.[20] In both HPLC and TLC lipophilicity determinations different parameters such as the retention factor extrapolated to pure water,[21] the chromatographic hydrophobicity index,[22] mean of retention factors and the scores corresponding to the first principal components[23, 24] have been proposed.

Apart from the experimental methods, lipophilicity can be estimated computationally using various chemical software products based on the different mathematical algorithms.[25] Generally, a good correlation of chromatographic lipophilicity indices with those obtained by theoretical computed indices validates the elaborated experimental methodology.

The objective of this study is to investigate the influence of different hydrophobic alcohols used as mobile phase additives in the chromatographic evaluation of lipophilicity of a representative class of new potentially anti-inflammatory drugs (new Schiff bases and Mannich bases).

RESULTS AND DISCUSSION

Chromatographic indices are widely used as alternative to lipophilicity values obtained by extractive method. Generally partitioning between a non-polar stationary phase and aqueous mobile phase in chromatography seems to be similar to partitioning in membranes in biological systems. In this study RP-TLC technique was used to evaluate the lipophilicity of new 1,2,4-triazole derivatives (Schiff bases – compounds 1-6 and Mannich bases – compounds 7-20) (their structure given in Fig. 1) with potentially anti-inflammatory activity tested in our previous study.[20] Because these drugs have not yet been investigated in this way, the retention study should provide relevant information about this important physicochemical property which affects pharmacodynamics and pharmacokinetic aspects of their action. The retention parameters were determined for different mobile phases containing phosphate buffer as aqueous component and methanol (FM1) respectively methanol containing 10% of upper alcohol (1-butanol (FM2) and 1-octanol (FM3)) as the hydrophobic additive. A linear relationship ($R^2 > 0.99$) between the concentration of organic modifier (methanol, methanol with 10% 1-butanol and methanol with 10% 1-octanol) and retention R_M were observed for each drug over the examined range of organic modifier concentration (from 50% to 90%) in all cases. Based on these relationships, mR_M, extrapolated R_{M0} values, corresponding slopes (b) and new $PC1/R_M$ lipophilicity indices (Table 1) were estimated with significant statistical parameters.

Profiles of derived parameters mR_M (Fig. 2) indicate the compounds 13, 14, 15 and 20 as the less lipophilic ones, their lipophilicity increasing by 1-butanol, respectively 1-octanol addition in the mobile phase composition. For the remaining compounds the hydrophobic additive (1-butanol or 1-octanol) in mobile phase has generally the tendency to lead to lower mR_M lipophilicity values.

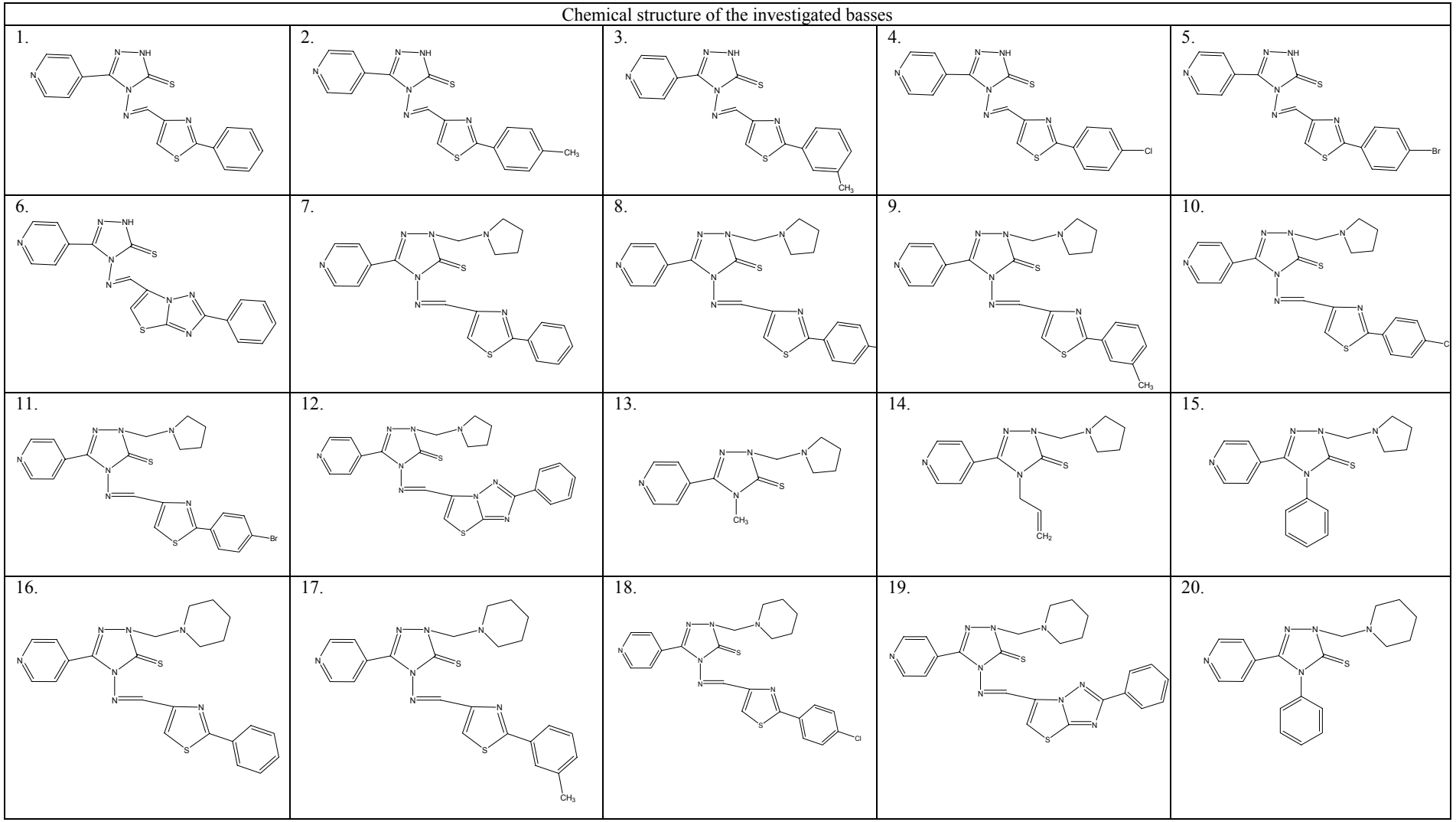


Fig. 1 – Chemical structure of the Schiff basses (compounds 1-6) and Mannich basses (compounds 7-20).

Table 1

Chromatographic lipophilicity parameters for the investigated Schiff bases and Mannich bases (FM1 – phosphate buffer: methanol; FM2 – phosphate buffer: methanol with 10% 1-butanol; FM3 – phosphate buffer: methanol with 10% 1-octanol)

No.	FM1				FM2				FM3			
Cpd.	mR_M	R_{M0}	b	$PC1/R_M$	mR_M	R_{M0}	b	$PC1/R_M$	mR_M	R_{M0}	b	$PC1/R_M$
1	0.59	3.06	-3.44	-0.19	0.53	2.39	-2.66	-0.34	0.33	2.91	-3.68	-0.32
2	0.74	3.75	-4.30	-0.55	0.70	3.04	-3.35	-0.94	0.53	3.57	-4.33	0.78
3	0.75	3.70	-4.22	-0.60	0.53	3.18	-3.78	-0.02	0.57	3.76	-4.56	0.93
4	0.71	3.45	-3.92	-0.52	0.55	3.32	-3.96	-0.10	0.68	4.30	-5.19	1.38
5	0.76	3.55	-3.98	-0.70	0.64	3.20	-3.69	-0.61	0.68	4.30	-5.17	1.43
6	0.75	3.46	-3.87	-0.69	0.64	2.95	-3.30	-0.70	0.46	3.23	-3.95	0.43
7	0.63	3.16	-3.63	-0.30	0.45	1.99	-2.21	-0.06	0.29	2.34	-2.92	-0.34
8	0.82	3.97	-4.50	-0.81	0.41	1.80	-1.98	0.05	0.31	2.41	-2.99	-0.23
9	0.60	3.18	-3.68	-0.19	0.66	3.06	-3.43	-0.73	0.34	2.63	-3.28	-0.15
10	0.63	3.30	-3.82	-0.27	0.56	2.67	-3.02	-0.37	0.55	3.90	-4.78	0.71
11	0.86	4.15	-4.71	-0.90	0.60	2.86	-3.22	-0.51	0.57	3.89	-4.75	0.82
12	0.74	3.32	-3.70	-0.67	0.60	2.86	-3.22	-0.57	0.60	3.78	-4.52	1.08
13	-0.10	0.92	-1.47	1.89	-0.04	0.63	-0.95	1.91	0.03	1.75	-2.45	-1.90
14	-0.09	1.28	-1.96	1.94	-0.00	0.78	-1.12	1.79	0.09	2.10	-2.87	-1.69
15	-0.05	1.48	-2.19	1.84	0.00	0.90	-1.28	1.81	0.13	2.24	-3.01	-1.46
16	0.53	3.21	-3.82	0.06	0.45	2.37	-2.74	0.08	0.48	3.49	-4.31	0.36
17	0.68	3.65	-4.26	-0.41	0.51	2.50	-2.84	-0.20	0.18	1.55	-1.96	-0.65
18	0.69	3.82	-4.47	-0.39	0.74	3.12	-3.40	-1.15	0.23	1.68	-2.07	-0.36
19	-0.07	1.48	-2.22	1.91	0.01	0.95	-1.34	1.78	0.15	2.07	-2.74	-1.17
20	0.67	3.12	-3.50	-0.48	0.75	3.27	-3.61	-1.11	0.48	3.65	-4.54	0.33

The influence of the mobile phase composition on the grouping of the investigated compounds from the lipophilicity point of view was evaluated by PCA analysis. According to the lipophilicity charts (Fig. 3) obtained by representation of the scores corresponding to the first two principal components ($PC1/R_M$ and $PC2/R_M$), different classes could be observed depending on the hydrophobic additive of the mobile phase. Also using methanol as organic component (Fig. 3(a)) the investigated bases are derived in two main groups according to their structural similarities. The first group contains the most lipophilic compounds including Schiff bases (compounds 1-6), pyrrolidine Schiff-Mannich bases (compounds 7-12) and piperidine Schiff-Mannich bases (compounds 16-19) while the second group is formed by Mannich bases (compounds 13-15 and 20). By using hydrophobic additive (1-butanol, Fig. 3(b) or 1-octanol, Fig. 3(c)) a representative group of compounds according to their structural particularities could not be observed.

The influence of mobile phase additive on the retention mechanism could be better observed by loadings profiles obtained by applying PCA directly on retention values (Fig. 4). According to these representations, a good linear dependence of loadings values on the organic additive fractions was observed (R^2 = 0.9824 for FM1; R^2 = 0.9981 for FM2; R^2 = 0.9917 for FM3) with the best statistical parameters being observed in case of 1-butanol addition.

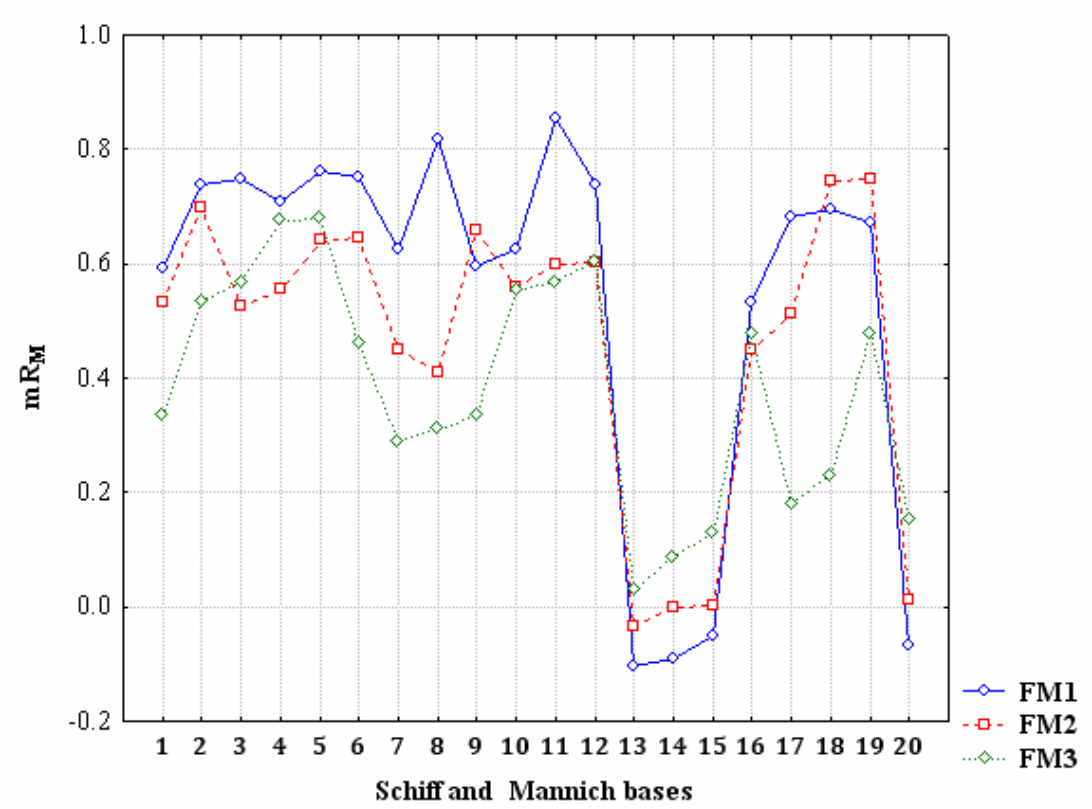
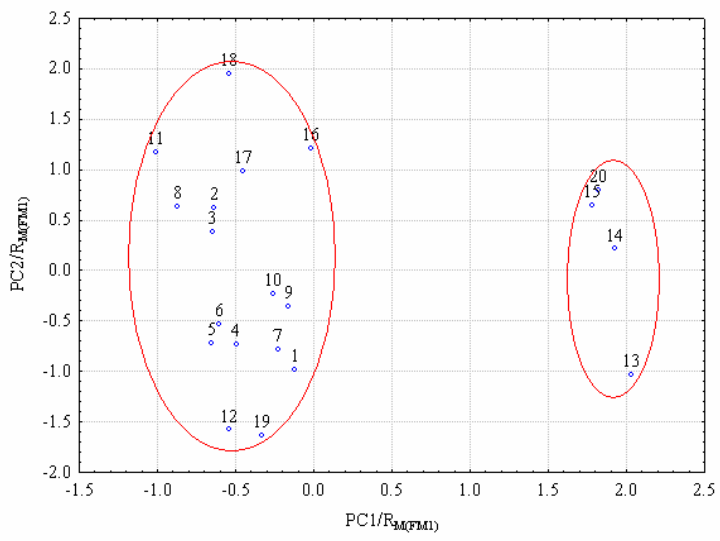
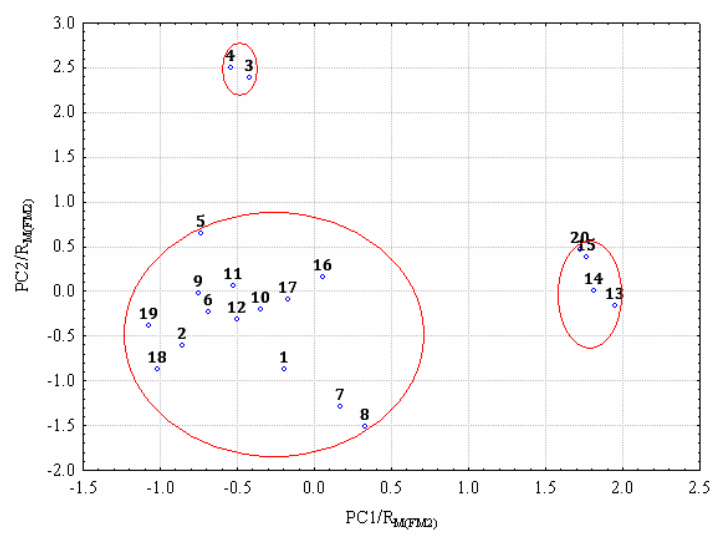


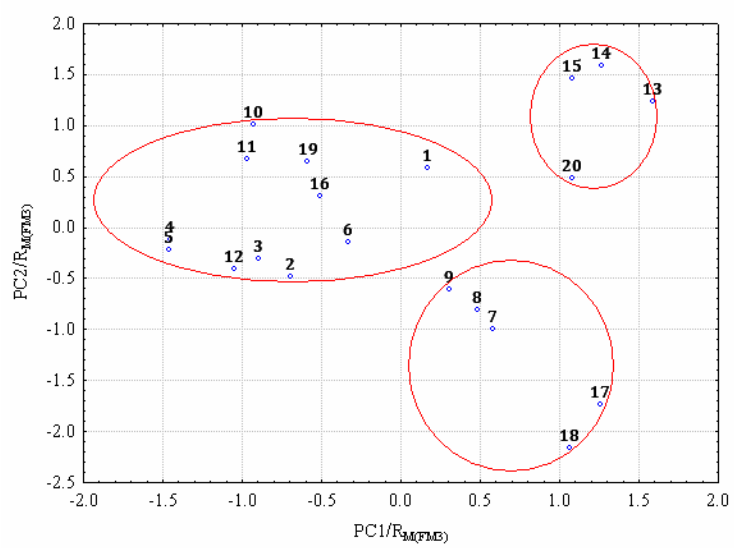
Fig. 2 – Profiles of the chromatographic retention parameters mR_M for the investigated mobile phases: FM1 – phosphate buffer : methanol; FM2 – phosphate buffer: methanol with 10% 1-butanol; FM3 – phosphate buffer: methanol with 10% 1-octanol.



(a)



(b)



(c)

Fig. 3 – Lipophilicity chart of the investigated bases according to the score plots of the first two principal components ($PC1/R_M$ and $PC2/R_M$): (a) FM1 – phosphate buffer: methanol; (b) FM2 – phosphate buffer: methanol with 10% 1-butanol; (c) FM3 – phosphate buffer: methanol with 10% 1-octanol.

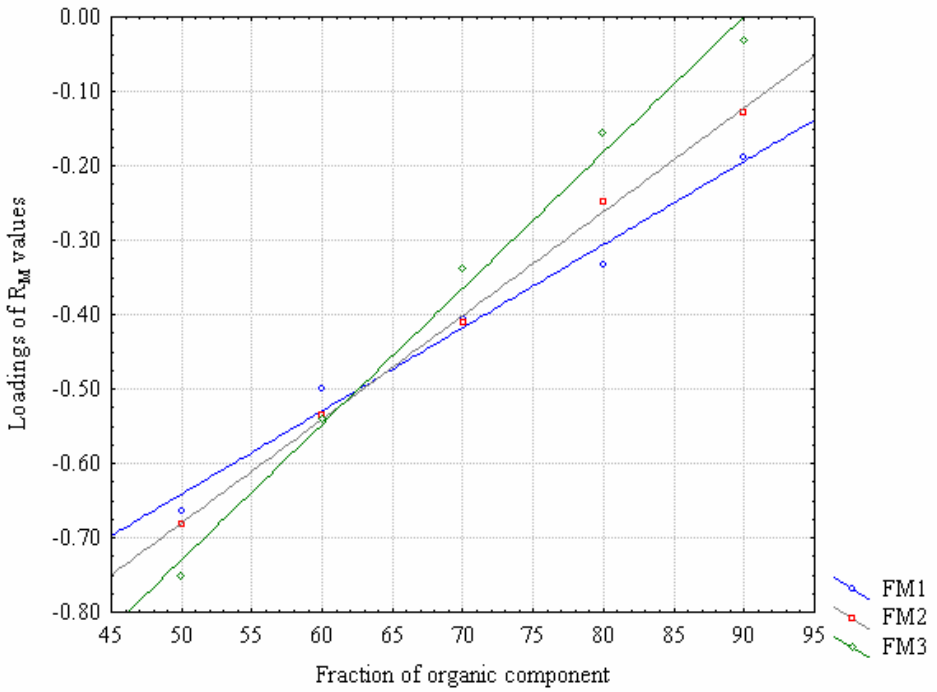


Fig. 4 – Loadings profiles of R_M values for the investigated bases (FM1 – phosphate buffer: methanol; FM2 – phosphate buffer: methanol with 10% 1-butanol; FM3 – phosphate buffer: methanol with 10% 1-octanol).

Besides experimental results, methods deriving log P from molecular structure are highly desired. Taking into account this aspect the experimental lipophilicity indices (mR_M, R_{M0}, b, and $PC1/R_M$) were compared with various theoretical log P values and also with the distribution coefficients (log D) estimated for the working pH values of the used mobile phases. While strong correlations ($r > 0.99$) were revealed between R_{M0} and b in all cases (Table 2), a significant correlation between lipophilicity parameters was observed for phosphate buffer: methanol (FM1) and phosphate buffer : methanol containing 10% 1-butanol (FM2) ($r = 0.92$ for mR_M and respectively $PC1/R_M$). These findings are supported also by correlations of experimental lipophilicity parameters with different computed lipophilicity indices (XLOGP2, PSA) that are significant only for the case of the first mobile phases FM1 and FM2. Surprisingly the 1-octanol as hydrophobic additive in the mobile phase has no positive contribution in the lipophilicity estimation of the investigated compounds.

Considering the studied bases (Table 1) it is difficult to observe structural differences between Schiff bases (compounds 1- 6) and Mannich bases (compounds 7-20) especially the possibility of dissociation. It is known that dissociable compounds may be partially dissociated depending on the pH value. Since the neutral and ionic species exhibit different polarities, the log D values of dissociable compounds are thus pH dependent. Such compounds could be susceptible to the secondary mechanisms of interaction with more or less nonpolar stationary phase and thus, their lipophilicity determination can be affected. According to the theoretical computation of log D values, the Schiff bases (compounds 1-6) seems to be susceptible for dissociation (Fig. 5) while the Mannich bases (compounds 7-20) show no significant dependence of log D values with mobile phase pH in the domain used in this study. These considerations are supported by the new correlations obtained for the Mannich bases (compounds 7-20) (Table 3).

Table 2

Correlation between chromatographic lipophilicity indices of the investigated Schiff bases and Mannich bases and the theoretical partition coefficients computed by different methods (n=20 compounds; FM1 – phosphate buffer: methanol; FM2 – phosphate buffer: methanol with 10% 1-butanol; FM3 – phosphate buffer: methanol with 10% 1-octanol)) (bolded value are statistically significant)

Experimental/ Computed parameters	**FM1**				**FM2**				**FM3**			
	mR_M	R_{M0}	b	$PC1/R_M$	mR_M	R_{M0}	b	$PC1/R_M$	mR_M	R_{M0}	b	$PC1/R_M$
$mR_{M(FM1)}$	1.00	**0.98**	**-0.95**	**-1.00**	**0.92**	**0.89**	**-0.85**	**-0.92**	0.75	0.56	-0.49	**0.83**
$R_{M0(FM1)}$	**0.98**	1.00	**-0.99**	**-0.97**	**0.89**	**0.85**	**-0.82**	**-0.89**	0.70	0.49	-0.42	0.79

Table 2 (continued)

$b_{(FM1)}$	**-0.95**	**-0.99**	1.00	**0.94**	**-0.86**	**-0.82**	**0.79**	**0.86**	-0.66	-0.44	0.37	-0.75
$PC1/R_{M(FM1)}$	**-1.00**	**-0.97**	**0.94**	1.00	**-0.92**	**-0.89**	**0.86**	**0.92**	-0.76	-0.57	0.50	**-0.83**
$mR_{M(FM2)}$	**0.92**	**0.89**	**-0.86**	**-0.92**	1.00	**0.96**	**-0.92**	**-1.00**	0.72	0.54	-0.48	0.79
$R_{M0(FM2)}$	**0.89**	**0.85**	**-0.82**	**-0.89**	**0.96**	1.00	**-0.99**	**-0.93**	**0.81**	0.66	-0.60	**0.87**
$b_{(FM2)}$	**-0.85**	**-0.82**	0.79	**0.86**	**-0.92**	**-0.99**	1.00	**0.88**	**-0.83**	-0.69	0.64	**-0.89**
$PC1/R_{M(FM2)}$	**-0.92**	**-0.89**	**0.86**	**0.92**	**-1.00**	**-0.93**	**0.88**	1.00	-0.67	-0.49	0.43	-0.75
$mR_{M(FM3)}$	0.75	0.70	-0.66	-0.76	0.72	**0.81**	**-0.83**	-0.67	1.00	0.95	**-0.92**	**0.99**
$R_{M0(FM3)}$	0.56	0.49	-0.44	-0.57	0.54	0.66	-0.69	-0.49	**0.95**	1.00	**-1.00**	**0.89**
$b_{(FM3)}$	-0.49	-0.42	0.37	0.50	-0.48	-0.60	0.64	0.43	**-0.92**	**-1.00**	1.00	**-0.85**
$PC1/R_{M(FM3)}$	**0.83**	0.79	-0.75	**-0.83**	0.79	**0.87**	**-0.89**	-0.75	**0.99**	**0.89**	**-0.85**	1.00
ALOGPs	**0.83**	**0.88**	**-0.89**	**-0.82**	**0.85**	**0.85**	**-0.84**	**-0.84**	0.65	0.45	-0.38	0.74
AC logP	0.78	**0.84**	**-0.86**	-0.77	0.76	0.75	-0.72	-0.75	0.58	0.38	-0.31	0.67
ALOGP	0.73	0.79	**-0.81**	-0.71	0.73	0.67	-0.63	-0.73	0.45	0.25	-0.19	0.54
MLOGP	0.50	0.49	-0.48	-0.50	0.55	0.47	-0.43	-0.57	0.31	0.18	-0.13	0.36
KOWWIN	0.39	0.50	-0.55	-0.37	0.38	0.32	-0.29	-0.39	0.14	-0.02	0.06	0.22
XLOGP2	**0.87**	**0.90**	**-0.91**	**-0.86**	**0.86**	**0.82**	-0.79	**-0.85**	0.63	0.42	-0.36	0.72
XLOGP3	0.74	**0.81**	**-0.83**	-0.73	0.72	0.67	-0.64	-0.73	0.47	0.26	-0.20	0.56
$LogP^{C}$	0.79	**0.80**	**-0.80**	-0.78	**0.80**	0.74	-0.70	**-0.81**	0.54	0.35	-0.29	0.62
$LogP^{V}$	0.79	**0.80**	-0.79	-0.79	**0.81**	0.74	-0.70	**-0.82**	0.54	0.35	-0.29	0.62
CLogP	0.48	0.57	-0.61	-0.46	0.47	0.39	-0.35	-0.48	0.19	0.01	0.03	0.27
PSA	**0.88**	**0.80**	-0.74	**-0.90**	**0.90**	0.87	**-0.84**	**-0.89**	0.75	0.60	-0.55	**0.80**
MSA	0.37	0.46	-0.51	-0.35	0.26	0.17	-0.13	-0.29	0.03	-0.17	0.23	0.12
$Log\ D_{(pH=7.90)}$	0.74	0.76	-0.76	-0.73	0.74	0.66	-0.62	-0.76	0.45	0.25	-0.20	0.53
$Log\ D_{(pH=8.20)}$	0.70	0.73	-0.73	-0.69	0.71	0.62	-0.57	-0.72	0.41	0.22	-0.16	0.49
$Log\ D_{(pH=8.40)}$	0.68	0.70	-0.71	-0.67	0.68	0.60	-0.55	-0.70	0.38	0.20	-0.14	0.46
$Log\ D_{(pH=8.80)}$	0.64	0.67	-0.68	-0.64	0.65	0.56	-0.51	-0.67	0.35	0.17	-0.11	0.43
$Log\ D_{(pH=9.10)}$	0.63	0.66	-0.67	-0.62	0.64	0.54	-0.49	-0.66	0.33	0.15	-0.10	0.41

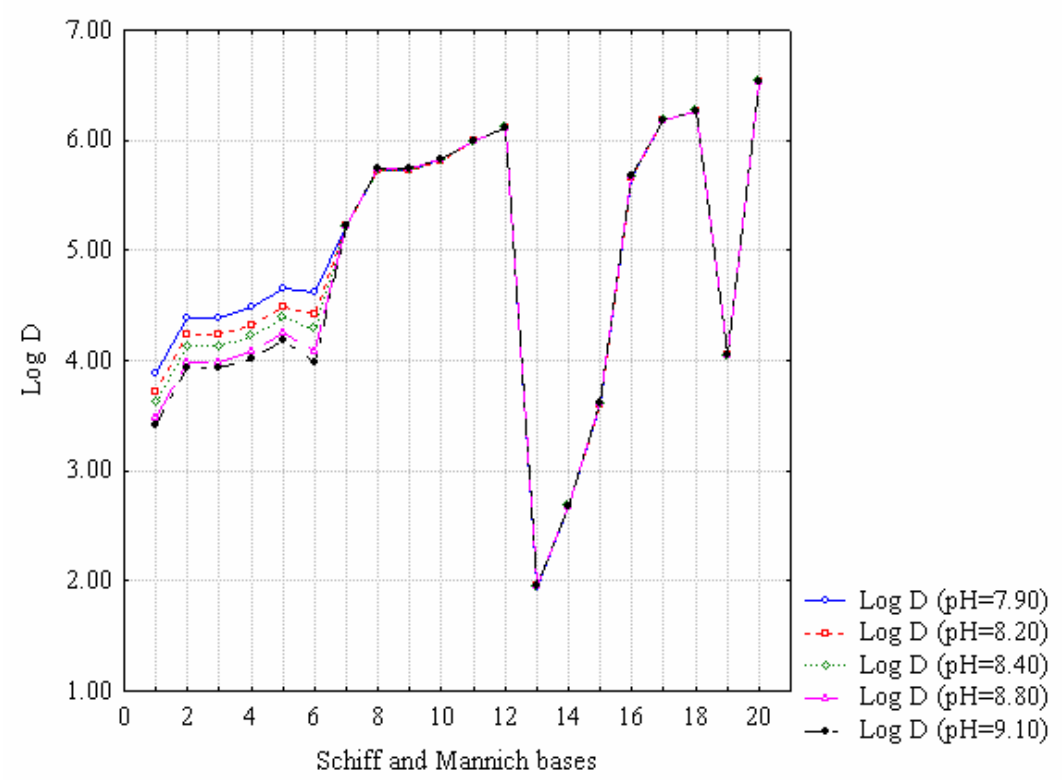


Fig. 5 – Variation of Log D values with pH for the investigated Schiff bases (compounds 1-6) and Mannich bases (compounds 7-20).

Table 3

Correlation between chromatographic lipophilicity indices of the investigated Schiff bases and Mannich bases and theoretical partition coefficients computed by different methods (n=14 compounds; FM1 – phosphate buffer: methanol; FM2 – phosphate buffer: methanol with 10% 1-butanol; FM3 – phosphate buffer: methanol with 10% 1-octanol) (bolded value are statistically significant)

Experimental/ Computed parameters	FM1				FM2				FM3			
	mR_M	R_{M0}	b	$PC1/R_M$	mR_M	R_{M0}	b	$PC1/R_M$	mR_M	R_{M0}	b	$PC1/R_M$
$mR_{M(FM1)}$	1.00	**0.98**	**-0.96**	**-1.00**	**0.92**	**0.88**	**-0.85**	**-0.93**	0.75	0.48	-0.39	**0.85**
$R_{M0(FM1)}$	**0.98**	1.00	**-1.00**	**-0.97**	**0.89**	**0.85**	**-0.82**	**-0.90**	0.69	0.41	-0.32	**0.81**
$b_{(FM1)}$	**-0.96**	**-1.00**	1.00	**0.94**	**-0.86**	**-0.82**	**0.80**	**0.87**	-0.65	-0.37	0.28	-0.77
$PC1/R_{M(FM1)}$	**-1.00**	**-0.97**	**0.94**	1.00	**-0.92**	**-0.88**	**0.85**	**0.93**	-0.75	-0.49	0.41	**-0.85**
$mR_{M(FM2)}$	**0.92**	**0.89**	**-0.86**	**-0.92**	1.00	**0.99**	**-0.98**	**-1.00**	0.73	0.48	-0.40	**0.83**
$R_{M0(FM2)}$	**0.88**	**0.85**	**-0.82**	**-0.88**	**0.99**	1.00	**-1.00**	**-0.98**	0.76	0.53	-0.46	**0.84**
$b_{(FM2)}$	**-0.85**	**-0.82**	**0.80**	**0.85**	**-0.98**	**-1.00**	1.00	**0.97**	-0.76	-0.55	0.48	**-0.84**
$PC1/R_{M(FM2)}$	**-0.93**	**-0.90**	**0.87**	**0.93**	**-1.00**	**-0.98**	**0.97**	1.00	-0.72	-0.46	0.38	**-0.82**
$mR_{M(FM3)}$	0.75	0.69	-0.65	-0.75	0.73	0.76	-0.76	-0.72	1.00	**0.93**	**-0.89**	**0.98**
$R_{M0(FM3)}$	0.48	0.41	-0.37	-0.49	0.48	0.53	-0.55	-0.46	**0.93**	1.00	**-0.99**	**0.83**
$b_{(FM3)}$	-0.39	-0.32	0.28	0.41	-0.40	-0.46	0.48	0.38	**-0.89**	**-0.99**	1.00	-0.77
$PC1/R_{M(FM3)}$	**0.85**	**0.81**	-0.77	**-0.85**	**0.83**	**0.84**	**-0.84**	**-0.82**	**0.98**	**0.83**	-0.77	1.00
ALOGPs	**0.85**	**0.90**	**-0.91**	**-0.83**	**0.88**	**0.87**	**-0.86**	**-0.88**	0.61	0.33	-0.24	0.74
AC logP	**0.82**	**0.88**	**-0.89**	**-0.80**	**0.81**	**0.80**	-0.79	**-0.80**	0.62	0.35	-0.27	0.74
ALOGP	**0.87**	**0.91**	**-0.92**	**-0.86**	**0.88**	**0.87**	**-0.86**	**-0.87**	0.66	0.39	-0.31	0.77
MLOGP	0.75	0.72	-0.70	-0.76	**0.81**	**0.82**	**-0.82**	**-0.80**	0.73	0.54	-0.47	**0.80**
KOWWIN	0.72	**0.81**	**-0.84**	-0.70	0.71	0.71	-0.71	-0.70	0.51	0.26	-0.19	0.62
XLOGP2	**0.91**	**0.94**	**-0.94**	**-0.90**	**0.90**	**0.89**	**-0.88**	**-0.90**	0.70	0.44	-0.35	**0.82**
XLOGP3	**0.88**	**0.93**	**-0.94**	**-0.87**	**0.87**	**0.86**	**-0.84**	**-0.87**	0.65	0.38	-0.30	0.77
$LogP^C$	**0.90**	**0.90**	**-0.88**	**-0.89**	**0.91**	**0.91**	**-0.90**	**-0.91**	0.75	0.50	-0.42	**0.84**
$LogP^V$	**0.91**	**0.90**	**-0.88**	**-0.91**	**0.92**	**0.92**	**-0.91**	**-0.92**	0.76	0.52	-0.44	**0.86**
CLogP	**0.82**	**0.88**	**-0.91**	**-0.80**	**0.81**	**0.80**	-0.79	**-0.80**	0.58	0.31	-0.23	0.70
PSA	**0.90**	**0.82**	-0.77	**-0.91**	**0.92**	**0.90**	**-0.89**	**-0.92**	**0.81**	0.60	-0.52	**0.87**
MSA	0.69	0.76	-0.79	-0.67	0.55	0.53	-0.52	-0.55	0.42	0.13	-0.04	0.56
$Log\ D_{(pH=7.90)}$	**0.92**	**0.92**	**-0.91**	**-0.92**	**0.93**	**0.92**	**-0.91**	**-0.93**	0.74	0.48	-0.40	**0.85**
$Log\ D_{(pH=8.20)}$	**0.92**	**0.92**	**-0.91**	**-0.92**	**0.93**	**0.92**	**-0.91**	**-0.93**	0.74	0.48	-0.40	**0.85**
$Log\ D_{(pH=8.40)}$	**0.92**	**0.92**	**-0.91**	**-0.92**	**0.93**	**0.92**	**-0.91**	**-0.93**	0.74	0.48	-0.40	**0.85**
$Log\ D_{(pH=8.80)}$	**0.92**	**0.92**	**-0.91**	**-0.92**	**0.93**	**0.92**	**-0.91**	**-0.93**	0.74	0.48	-0.40	**0.85**
$Log\ D_{(pH=9.10)}$	**0.92**	**0.92**	**-0.91**	**-0.92**	**0.93**	**0.92**	**-0.91**	**-0.93**	0.74	0.48	-0.40	**0.85**

Considering these compounds, the inter-correlation with some of the computed lipophilicity indices (XLOGP2, XLOGP3, $LogP^C$, $LogP^V$, CLogP) and also with log D values was significantly improved for the experimental parameters obtained using the first mobile phases FM1 and FM2 respectively.

EXPERIMENTAL

Chemicals and reagents

All chemicals were of analytical reagent grade. The new polyheterocyclic Schiff bases and Mannich bases (Fig. 1) evaluated in the present study were synthesized in our laboratory (Department of Organic Chemistry, Iuliu Haţieganu University of Medicine and Pharmacy, Cluj-Napoca, Roumania).[26] Analytical grade methanol, 1-butanol and 1-octanol were purchased from Merck (Darmstadt, Germany).

Chromatographic procedure

All the chromatographic measurements were carried out on HPTLC LiChrospher® silica gel 60 RP-18 WF_{254s} plates purchased from Merck (Darmstadt, Germany). Standard solutions of the investigated compounds (1 mg mL^{-1}) were prepared in dimethylsulfoxide (DMSO, Schiff bases) or chloroform (Mannich bases) and 2 μL were applied in duplicate in all cases by means of a Linomat 5 TLC applicator (Camag, Switzerland) at 15 mm from bottom edge of the plates. Chromatography was performed in a normal developing chamber saturated for 15min at room temperature (≈20^0C). The chromatographic retention was measured using phosphate buffer (pH=7.10) as aqueous component and methanol (FM1) respectively methanol containing 10% of different upper alcohol (1-butanol and 1-octanol) (FM2 and FM3 respectively) as the hydrophobic additive of the mobile phase. Different proportions of aqueous-organic component (from 50% to 90% organic component, with an increment of 10%) were investigated according to the lipophilic character of the studied compounds. Also, the following pH values for the working mobile phases (organic component : phosphate buffer v/v) were obtained: 7.94 (for 50:50, v/v); 8.17 (for 60:40, v/v); 8.42 (for 70:30, v/v); 8.75 (for 80:20, v/v) and 9.13 (for 90:10, v/v) respectively. The developing distance was 8 cm in all cases. The spots were detected after elution under UV light at λ = 254 nm. An average value for the retention factor (R_F) was determined using two identical spots in all cases.

Chromatographic lipophilicity parameters

Common lipophilicity estimators R_M ($R_M = \log (1/R_F - 1)$)[27] and extrapolated R_{M0} values ($R_M = R_{M0} + bC$, where b represents the slope directly related to the specific surface area of the stationary phase while C represents the volume fraction of the organic solvent in the mobile phase composition),[28] were derived from the retention factors R_F. In addition, the new proposed lipophilicity parameters mR_M (arithmetic mean of R_M values for mobile phases containing diferent proportion aqueous-organic component)[29, 30] and scores $PC1/R_M$ (new indices corresponding to the first principal component obtained by applying the Principal Component Analysis (PCA) on the retention data R_M)[23, 31] were investigated in order to evaluate the chromatographic behavior, chromatographic interaction mechanism and (dis)similarity of discussed new polyheterocyclic Schiff bases and Mannich bases from the lipophilicity point of view.

Computed lipophilicity indices

Nowadays, there is a large number of computer software able to calculate different lipophilicity descriptors based on various algorithms. Some of the computed lipophilicity indices (based on electrotopological-state descriptors (ALOGPs), group contributions descriptors (AC logP and miLogP), fragmental methods and reductionist approaches (KOWWIN), atom type and correction factors descriptors (XLOGP2, XLOGP3)) have been obtained using the Virtual Computational Chemistry Laboratory website [Virtual Computation Chemistry Laboratory, http://www.vcclab.org]. After previous molecule drawing and geometry optimization the Chem3D Ultra 8.0 [http://www.cambridgesoft.com] software based on different fragmental and atomistic methods was used in order to calculate three log P values (CLogP, $LogP^C$ – Crippen's LogP and $LogP^V$ – Viswanadhan's logP). As the electronic structure of the investigated bases may implies even ionic forms, distribution coefficients at working pH (five log D values) and two surface area descriptors (PSA-Polar Surface Area and MSA - Molecular Surface Area) were calculated according to the theoretical computations using Marvin program by Chemaxon*, Calculator Plugin and Chemical Terms* available as free internet module Marvin Sketch 5.3.2 (http://www.chemaxon.com/marvin/sketch/index.php).

CONCLUSIONS

Different indices of lipophilicity for new potentially anti-inflammatory 1,2,4-triazole derivatives (Schiff bases and Mannich bases) were determined for the first time by RP-TLC using phosphate buffer: methanol respectively phosphate buffer: methanol with 10% of different upper alcohol (1-butanol and 1-octanol) as the hydrophobic additive of the mobile phase. The obtained parameters show that the quality of experimental-computed lipophilicity parameters correlations are weakly dependent on the presence of upper alcohol additive, 1-butanol having a weak positive effect on the chromatographic lipopilicity determination.

Highly significant correlations between different experimental indices of lipophilicity and various computed log P values obtained in case of Mannich bases suggest that Schiff bases are susceptible for dissociation and their lipophilicity determination can be affected in the considered experimental conditions (as is revealed by computation log D values).

Acknowledgments: This work was possible with the financial support offered by Roumanian Ministry of Education, Research, Youth and Sport through research project PN-II-ID-PCE-2011-3-0366.

REFERENCES

1. D. Sunil, A. M. Isloor, P. Shetty, B. Chandraakantha and K. Satyamoorthy, *Med. Chem. Res.*, **2011**, *20*, 1024-1032.
2. D. Sunil, A. M. Isloor, P. Shetty, P.G Nayak and K. S. R. Pai, *Arabian J. Chem.*, **2013**, *6*, 25-33.
3. Y. L. N. Murty, B. Govindh, B. S. Diwakar, K. Nagalakshmi and K. V. R. Rao, *Med. Chem. Res.*, **2012**, *21*, 3104-3110.
4. T. Plech, M. Wujec, M. Majewska, U. Kosikowska and A. Malm, *Med. Chem. Res.*, **2013**, *22*, 2531-2537.
5. S. M. Sondhi, A. Surbhi, R. Reshma, K. Nikhil and R. Partha, *Med. Chem. Res.*, **2011**, *21*, 3620-3628.
6. Nithinchandra, B. Kalluraya, S. Aamir and A. R. Shabaraya, *Eur. J. Med. Chem.,* **2012**, *54*, 597-604
7. B. Testa, P. Crivori, M. Reist and P. A. Carrupt, *Perspect. Drug Discov. Des*., **2000**, *17*, 179-211.
8. K. Valkó, *J. Chromatogr*. A, **2004**, *1037*, 299-310.
9. C. Giaginis and A. Tsantili-Kakoulidou, *J. Liq. Chromatogr. Relat. Technol*. **2008**, *31*, 79-96.
10. N. Perisić-Janjić, G. Y. Vastag, J. Tomić and S. Petrović, *J. Planar Chromatogr*., **2007**, *20*, 353-359.
11. A. Hawryl, E. Kumierz, P. Pisarczyk, M. Wujec and M. Waksmundzka-Hajnos, *Acta Chromatogr.*, **2012**, *24*, 271–290.
12. J. M. Pallicer, C. Calvet, A. Port, M. Rosés, C. Ràfols and E. Bosch, *J. Chromatogr. A*, **2012**, *1240*, 113- 122.
13. C. Sarbu, R. D. Naşcu-Briciu, D. Casoni, A. Kot-Wasik, A. Wasik and J. Namiésnik, *J. Chromatogr. A*, **2012**, *1266*, 53-60.
14. E. Caiali, D. Casoni, P. Ionita, V. David and C. Sârbu, *J. Liquid Chromatogr. Rel. Technol*., **2014**, *7*, 2287-2301.
15. A. Medvedovici, V. David, V. David and C. Georgita, *J. Liq. Chromatogr. Rel. Technol*. **2007**, *30*, 199-213.
16. S. Udrescu, A. Medvedovici and V. David, *J. Sep. Sci*. **2008**, 31, 2939-2945.
17. S. Udrescu, I. D. Sora, V. David and A. Medvedovici, *J. Liq. Chromatogr. Rel. Technol*. **2010**, *33*, 133-149
18. S. Udrescu, I. D. Sora, F. Albu, V. David and A. Medvedovici, *J. Pharm. Biomed. Anal*. **2011**, *54*, 1163-1172.
19. A. Medvedovici, S. Udrescu, F. Albu, F. Tache and V. David, *Bioanalysis* **2011**, *3*, 1935-1947.
20. V. David, T. Galaon, E. Caiali and A. Medvedovici, *J. Sep. Sci*. **2009**, *32*, 3099-3106.

21. R. Kaliszan, *Chem. Rev*. **2007**, *107*, 3212-3246.
22. M. Rosés, E. Bosch, C. Ràfols and E. Fuguet, *Adv. Chromatogr*., **2012**, *50*, 377-414.
23. D. Casoni, A. Kot-Wasik, J. Namiésnik and C. Sârbu, *J. Chromatogr. A*, **2009**, *1216*, 2456-2465.
24. D. Casoni and C. Sârbu, *J. Sep. Sci.*, **2012**, *35*, 915-921.
25. P.-A. Carrupt, B. Testa and P. Gaillard, in "Reviews in Computational Chemistry", Vol. *11*, Wiley-VCH, New York, 1997, pp. 241-315.
26. A. Toma, D. Hapău, M. Naghi, L. Vlase, C. Mogoşan and V. Zaharia, *Studia UBB Chemia*, **2013**, *LVIII, 2*, 93-104.
27. E. C. Bate-Smith and R. G. Westall, *Biochim. Biophys. Acta*, **1950**, *4*, 427-440.
28. E. Soczewinski and C. A. Wachtmeister, *J. Chromatogr*., **1962**, *7*, 311-320.
29. D. Casoni and C. Sârbu, *Chromatographia*, **2009**, *70*, 1277-1282.
30. R. D. Briciu, A. Kot-Wasik, A.Wasik, J. Namiésnik and C. Sârbu, *J. Chromatogr. A*, **2010**, *1217*, 3702-3706.
31. R. D. Briciu, A. Kot-Wasik, J. Namiesnik, and C. Sârbu, *J. Sep. Sci*., **2009**, *32*, 2066-2074.