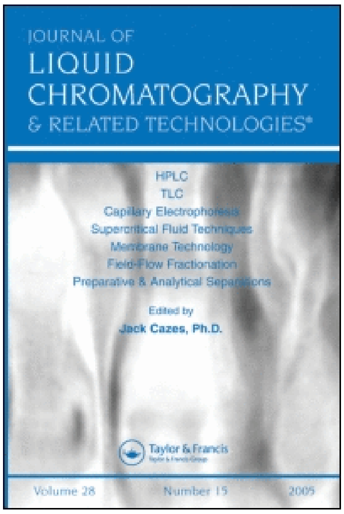


Journal of Liquid Chromatography & Related Technologies
Publication details, including instructions for authors and subscription information:
http://www.informaworld.com/smpp/title~content=t713597273

Investigation of Structure-Property Relationships for Imidazoquinolone Derivatives. I. Relation Between Partition Coefficient and Chromatographic Retention

Zs. Budvári-Bárány[a]; Gy. Szász[a]; K. Takács-Novák[a]; J. Vámos[a]; J. Kökösi[a]; M. Józan[a]
[a] Institute of Pharmaceutical Chemistry Semmelweis Medical University Budapest, Hungary

To cite this Article Budvári-Bárány, Zs. , Szász, Gy. , Takács-Novák, K. , Vámos, J. , Kökösi, J. and Józan, M.(1990) 'Investigation of Structure-Property Relationships for Imidazoquinolone Derivatives. I. Relation Between Partition Coefficient and Chromatographic Retention', Journal of Liquid Chromatography & Related Technologies, 13: 8, 1485 — 1497

To link to this Article: DOI: 10.1080/01483919008048971

URL: http://dx.doi.org/10.1080/01483919008048971

JOURNAL OF LIQUID CHROMATOGRAPHY, 13(8), 1485-1497 (1990)

INVESTIGATION OF STRUCTURE-PROPERTY RELATIONSHIPS FOR IMIDAZOQUINOLONE DERIVATIVES. I. RELATION BETWEEN PARTITION COEFFICIENT AND CHROMATOGRAPHIC RETENTION

ZS. BUDVÁRI-BÁRÁNY, GY. SZÁSZ,
K. TAKÁCS-NOVÁK, J. VÁMOS,
J. KÖKÖSI, AND M. JÓZAN
Institute of Pharmaceutical Chemistry
Semmelweis Medical University
Budapest, Hungary

ABSTRACT

New imidazoquinazolone derivatives as potential pharmacons, prepared in our laboratory, have been studied by HPLC and TLC methods. As a part of a structure-properties research study (SPR) this paper deals with the relationship between the chemical structure of the compounds and their chromatographic retention. A good correlation has been found between log k' or R_M and the concentration of the organic modifier in mobile phase, as well as log k' or R_M and log P. On the basis of the measured retention data, log P prediction was made for compounds with very poor water-solubility.

INTRODUCTION

The 1-octanol/water partition coefficient (P) is an important parameter for QSAR studies. This was shown more than 25 years ago by Hansch, et al. (1). The relationship between log P and the biological activity of drugs has been exhibited by hundreds of papers. Consequently, the partition coefficient keeps its position in the armament of drug design in spite of the fact, that the shake flask-method generally used for its determination has a number of disadventages. The method can provide rather inaccurate results for compounds with poor water (or 1-octanol) solubility and requires that the solute be ultrapure.

The drawbacks of the shake flask method led us to initiate research aimed at finding a more adventageous solution for the determination of P or to substitute the P-values by another structurally related parameter.

Considering the well known relation between the chromatographic retention and the partition coefficient, it is quite reasonable that first of all the different chromatographic methods be considered for the determination (prediction) of the partition coefficient.

The thin-layer chromatographic R_M value has been found to correlate with log P by several authors. In a majority of the early papers, reversed-phase TLC was used (2-15) and paraffin oil or silicon oil served as stationary phase. It was shown in our laboratory for a set of pyridopyrimidine derivatives (n=18) that R_M values obtained with reversed-phased TLC using a RP-di-C_1 stationary phase were well correlated with log P (16).

Due to its known advantages over other chromatographic methods, HPLC seems to be the "method of choice" for QSAR studies when the prediction, determination or substitution of log P values is wanted. Though no "breakthrough" can be observed as concerns the spreading of QSAR-application of HPLC.

In 1974 Haggerty and Murill (17) suggested the utility of RPHPLC for the determination of partititon coefficients. Since then it has been shown by several authors that 1-octanol/water partition coefficient can be predicted from retentions on various types of RP HPLC columns (18-25). As hydrophobic stationary phases, the chemically bonded phases, mainly silanes (ODS, etc.) are used usually. Although the residual silanol groups may result in some hydrophylic places of the hydrophobic surface (17), the correlation coefficients obtained without any presilylation of the surface are in most cases accurate enough for use in QSAR studies (25).

While the HPLC method has been shown quite useful for neutral and acidic compounds, some difficulties could be observed with basic compounds (26). In spite of this and similar other reports, it was shown that the retention of basic compounds in RPHPLC systems can be used to predict their 1-octanol/water partition coefficients (27). The deviations occuring in the correlation between lipophilicity measured by HPLC and the shake flask method have been interpreted by Sabatka, et al. (28).

There have been very few studies of the validity of the log P - log k' relationship for heterocyclic compounds. In our previous work, a good correlation was found between log P and log k' for a series of pyridopyrimidine derivatives (29). Similarly, we found suitable R_M values determined on RP-di-C_1 stationary phase for the prediction of log P for rutecarpine derivatives (30). In the present work we are dealing with another structural group of nitrogen bridged heterocycles. A series of new imidazoquinazolones synthetized recently in our laboratory were investigated for log P - log k' relationship, including an answer to the question whether presilylation of the ODS-column materials is necessary or not in case of the studied and other similar compounds. Since these compounds represent very weak basicity (for pK_a values see Table 1) and having no interacting polar group other than the basic nitrogen, N-2, the presilylation of the ODS stationary phase did not seem necessary. In addition to the HPLC study, TLC search was also made for a log P - R_M correlation.

EXPERIMENTAL

Materials

All the model substances (Table I, compounds 1-10) were synthesized in our laboratory and the method will be published elsewhere. The identification and quality control of the substances were performed via melting point determination and HPLC chromatography. The chemicals and solvents were of analitycal grade and Li-Chrosorb for HPLC (Merck) quality.

Chromatography

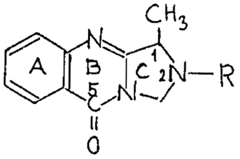
The HPLC apparatus consisted of a Waters Model 501 (Millipore, USA) solvent delivery system, a variable wavelength detector (Model OE 308, Labor MIM, Budapest, Hungary) and a recorder, type OH-814/1 (Radelkis, Hungary). The reversed phase LiChrosorb (5-RP-18) C_{18} column measured 250x4.6 mm, and was prepacked with material with a particle size of 5 μm (BST, Hungary).

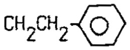
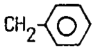
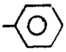
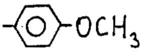
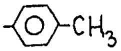
20 ul of sample solution (0.1 mg/ml eluent) was injected. Methanol-water in different ratios as mobile phase was applied. The flow rate was 0.7 ml/min. All experiments were run under isocratic conditions at 25 °C. The log k' values were calculated as the mean of the retention values obtained from six injections.

TLC was carried out by using RP-di-C_1 factory made (Kieselgel 60 F_{254} silanisiert, Merck) plates as stationary phase; previously the plates were reactivated by heating at 160 °C for 60 min. The sample solutions were applied to a Nanomat Autoliner. The chromatograms were developed in a "Desaga" normal chamber with acetone-water of different concentration as mobile phase.

Table 1

Structure, pK_a and log P values for examined compounds



Number of compounds	R	pK_a	log P
1	CH_3	2.75	0.849
2	C_2H_5	2.87	1.015
3	n-C_3H_7	3.04	1.395
4	n-C_4H_9	3.06	1.604
5	CH_2CH_2OH	2.37	0.462
[a] 6			
[a] 7			
[a] 8			
[a] 9			
[a] 10			

[a] Due to very poor water solubility, experimental data with proper accuracy were not available.

Saturation time was 30 min. The spots were assigned under UV light, λ = 254 nm and the R_M values were calculated from the mean of the R_F values from six different developments.

Partition coefficient

For the determination of log P values, the shake-flask method was used. Britton - Robinson buffer solution (pH = 5.0) served as aqueous phase. Considering their pK values, the examined compounds exist at pH = 5 mainly in nonionised form. The two phases were mutually saturated by shaking 1 hour and separating by centrifugation (20 min; 730 g) before experiment. The compounds were dissolved in the buffer solution (2×10^{-4} - 5×10^{-3} M) and the aqueous phase was equilibrated with 1-octanol at three volume ratios (w/o = 10:1, 10:2, 10:5) by 1 hour shaking. After centrifugation the decrease of the concentration in the aqueous phase was measured spectrophotometrically at λ =300 nm (Specord UV-VIS, Zeiss, Jena).

The logarithm of the apparent partition coeffiecient at three different volume ratios were calculated from six parallel measurements and then averaged (n = 18). To obtain the true partititon coefficients (see Table I) the following correction was use $\log P = \log P_{app} + \log (1 + 10^{pK_a-5.0})$. The standard deviation of the data is less than +/-0.04.

RESULTS AND DISCUSSION

The log k' data of the compounds measured at different methanol concentrations can be seen in Table II. The methanol concentration ranged between 60 and 80 percent; at methanol concentrations less than 60 percent the retention times were too long and shape of the peaks were nonideal. The good correlation between log k' and MeOH concentration of the mobile phase shows no

Table II

log k' values and correlation between log k' and methanol concentration of the mobil-phase

Number of compounds	MeOH : H_2O					r
	80:20	75:25	70:30	65:35	60:40	
1	- 0.319	- 0.288	- 0.194	- 0.174	0.057	- 0.927
2	- 0.244	- 0.218	- 0.041	0.041	0.250	- 0.974
3	0.0086	0.057	0.206	0.321	0.636	- 0.958
4	0.172	0.255	0.456	0.580	0.913	- 0.975
5	- 0.538	- 0.538	- 0.394	- 0.328	- 0.149	- 0.961
6	0.274	0.347	0.573	0.731	1.126	- 0.969
7	0.204	0.268	0.497	0.687	1.028	- 0.976
8	0.259	0.339	0.543	0.741	1.071	- 0.978
9	0.230	0.294	0.520	0.706	1.066	- 0.973
10	0.494	0.543	0.809	0.998	1.402	- 0.967

specific interaction between the solutes and the components of the mobile phase with the change in methanol content. This also shows that the silanophil effect has no influence on the results. This assumption seems to be confirmed by the very weak basicity of the compounds. As expected, a gradual increase of retention values can be observed for the N-alkyl homologues (compds 1-4, see Table II). A radical decrease in retention is observed for the hydroxy derivative (compound 5). An interesting phenomenon occurs with the compounds having N-phenyl substitutent. The retention of the N-phenethyl derivative (compd 6) is somewhat larger than that of the N-phenyl derivative (compd 8) in spite of the precence of the CH_2-CH_2 group in the former compound. This may be interpreted by the electron attracting effect of the phenyl ring attached directly to the N2 atom, decreasing the polarity of it and weakening the desorptive effect of the mobile phase. As a further consequence, the retention of the benzyl derivative (compd 7) is smaller than that of the phenyl one. The methylene group between the aromatic ring and N2 does not increase affinity for the column as much as expected. The hydrophobicity of the p-CH_3 group seems to be much greater than that of the joining methylene group(s) between the N2 and the phenyl group (compare the log k' of compds 10 and 6,7).

Also close correlation can be seen between R_M values and the concentration of the organic modifier (acetone) of the mobile phase (Table III). The concentration of acetone ranged through the experiments from 25 to 65 percent with 5 percent increase. (Table III contains only the data in 10 percent ranges.) The chromatographic behaviour of the compounds related to the chemical structure shows the same tendencies as were shown in the HPLC experiments.

We have made correlation analysis between log k', R_M (see Table II and III) and log P data for compounds 1-5. The most

Table III

R_M values and correlation between R_M and acetone concentration of the mobil-phase

Number of compounds	$(CH_3)_2CO : H_2O$					
	25:75	30:70	40:60	50:50	60:40	r
1	0.191	0.132	- 0.003	- 0.228	- 0.352	0.995
2	0.357	0.271	0.113	- 0.112	- 0.250	0.996
3	0.739	0.619	0.419	0.125	- 0.070	0.998
4	1.073	0.943	0.663	0.317	0.060	0.999
5	0.020	- 0.008	- 0.132	- 0.395	- 0.541	0.990
6	1.723	1.404	0.985	0.489	0.156	0.996
7	1.566	1.227	0.852	0.431	0.122	0.995
[a] 8	1.548	1.398	0.928	0.469	0.152	0.997
9	1.522	1.325	0.866	0.424	0.093	0.998
[a] 10	1.689	1.684	1.129	0.598	0.237	0.994

[a] tailing spots

Table IV

Log P values predicted by HPLC and TLC retention

Number of compound	$\log P_{pred}$	
	log k'	R_M
6	1.803	1.804
7	1.699	1.739
8	1.779	1.797
9	1.733	1.684
10	2.158	1.959

approporiate peak (spot) shapes, retention values as well as correlation coefficients were found at the mobile phase composition of $MeOH:H_2O$ 80:20 for HPLC and $acetone:H_2O$ 60:40 for TLC. The retention values gained in these latter systems correlated well with the corresponding log P values:

$$\log P = 1.611 \cdot \log k'_{80\%} + 1.362 \qquad (1)$$

$$n = 5 \quad r = 0.997 \quad s = +/-0.04 \quad F = 502$$

$$\log P = 1.912 \cdot R_{M60\%} + 1.506 \qquad (2)$$

$$n = 5 \quad r = 0.999 \quad s = +/-0.02 \quad F = 184$$

Using equations 1 and 2 the log P data for five compounds (No 6-10, Table I) were predicted. Due to the very poor water-solubility the direct experimental determination of log P was not available for these compounds. The predicted data can be seen in Table IV. It can be seen that the predicted log P values of the two series agree quite well in spite of the fact, that the prediction was done with the use of retention parameters (R_M, log k') deteremined in RP-chromatographic systems with different stationary phases.

The good correlation together with the low standard deviation of the predicted values allow to conclude, that HPLC and TLC provide accurate methods of log P prediction for SPR as well as QSAR studies. The authenticity of prediction was supported through correlation with another structural property as isotropic surface area. This is included in another publication (31).

The authors are grateful to I. Kovács-Derzsi for her excellent technical assistance.

REFERENCES

1. Hansch, C.L., Fujita, T., J. Amer. Chem. Soc. <u>85</u>, 2817, 1963.
2. Boyce, C.B., Millborrow, B.V., Nature (London) <u>208</u>, 637, 1965.
3. Biagi, G.L., Barbaro, A.M., Gamba, M.F., Guerra, M.C., J. Chromatogr., <u>41</u>, 371, 1969.
4. Biagi, G.L., Barbaro, A.M., Guerra, M.C., Gamba, M.F., ibid., <u>44</u>, 195, 1969.
5. Biagi, G.L., Guerra, M.C., Barbaro, A.M., Gamba, M.F., J. Med. Chem., <u>13</u>, 511, 1970.
6. Biagi, G.L., Barbaro, A.M., Guerra, M.C., Gamba, M.F., Advan. Chem. Series, <u>61</u>, 1973.
7. Biagi, G.L., Barbaro, A.M., Guerra, M.C., Forti, G.C., Fracasso, M.E., J. Med. Chem., <u>17</u>, 28, 1974.
8. Biagi, G.L., Gondolfi, O., Guerra, M.C., Barbaro, A.M., Cantella-Forti, G., ibid., <u>18</u>, 868, 1975.

9. Biagi, G.L., Barbaro, A.M., Gondolfi, O., Guerra, M.C., Cantella-Forti, G., ibid., 18, 873, 1975.

10. Seydel, J.K., Schaper, K.J., Wempe, E., Cordes, H.P., J. Med.. Chem., 19, 483, 1976.

11. Seydel, J.K., Schaper, K.J., Chem. Struktur und biologische Äktivität von Wirkstoffen, Verlag Chemie, Weinheim-New York, 1979.

12. Seydel, J.K., Ahrens, H., Losert, W., J. Med. Chem., 18, 234, 1975.

13. Tomlinson, E., J. Chromatogr., 113, 1, 1975.

14. Carlson, R.M., Carlson, R.E., Kopperman, H.L., J.Chromatogr., 107, 219, 1975.

15. Waisser, K., Spalenka, E., in Knoll, J. (Ed.): Chemical Structure-Biological Activity Relationships Quantitative Approaches, Akademiai Kiado, Budapest, 1980.

16. Papp, O., Valkó, K., Szász, Gy., Hermecz, I., Vámos, J., Hankó, K., Halász-Ignáth, Zs., J.Chromatogr., 252, 67, 1982.

17. Haggerty, W.J., Murill, E.A., Res. Dev., 25, 30, 1972.

18. McCall, J.M., Carlson, R.M., Hulshoff, A., Perrin, J.H., J. Chromatogr., 129, 263, 1976.

19. Hammers, W.E., Meurs, G.J., ibid., 247, 1, 1982.

20. Hafkenscheid, T.L., Tomlinson, E., Int. J. Pharm., 16, 225, 1983.

21. ibid., 292, 305, 1984.

22. Thus, J.L., Kraak, J.C., J. Chromatogr., 320, 271, 1985.

23. el Tayar, N., van de Waterbeemd, H., Testa, B., J. Chromatogr., 320, 293, 1985.

24. ibid., 320, 293, 1985.

25. Kaliszan, R., ibid., 220, 71, 1981.

26. Unger, S.H., Chiang, G.H., J. Med. Chem., 24, 262, 1981.

27. Kraak, J.C., van Rooij, H.H., Thus, J.L.G., J. Chromatogr., 352, 445, 1986.

28. Sabatka, J.J., Minick, D.J., Shumaker, T.K., Hodgson, G.L., Brent, D.A., ibid., 384, 349, 1987.

29. Shalaby, A., Budvári-Bárány, Zs., Szász, Gy., Hermecz, I., Acta Pharm. Hung., 55, 249, 1985.

30. Vámos, J., Kökösi, J., Szász, Gy., Hankó-Novák, K., Józan, M., Kapolka, P., Acta Pharm. Hung., 58, 247, 1988.

31. Takács-Novák, K., Józan, M., Nagy, P., Őrfi, L., Dunn III, W.J., Szász, Gy., Investigations of structure-property relationships for imidazoquinazolone derivatives. Part 2. Partition coefficients and solubility. J. Comput.-Aided Molec. Des., submitted for publication.