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QUANTUM CHEMICAL PARAMETERS IN CORRELATION ANALYSIS OF GAS-LIQUID CHROMATOGRAPHIC RETENTION INDICES OF AMINES

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SUMMARY

For a structurally diverse set of primary, secondary and tertiary heterocyclic amines, correlations were found between the Kováts retention indices obtained on the methyl phenyl silicone phase OV-101 and quantum chemically calculated parameters. The total energy calculated by CNDO/2 molecular orbital method was chosen as a measure of a solute's ability to participate in dispersive interactions with the stationary phase. As a measure of the solute's ability to undergo polar interactions with the stationary phase, a parameter was proposed which reflects the highest local intermolecular dipole moment. It is defined as the largest difference in electron charges between two atoms. A two-parameter regression equation was derived which describes satisfactorily the retention of structurally different polar solutes on a relatively non-polar stationary phase. Some evidence is also provided that charge-transfer complexes are formed during the separations process.

INTRODUCTION

The distribution of a solute between a mobile and a stationary chromatographic phase is dependent on the forces existing between the solute molecules and those of each phase. Most generally, these forces are divided in two groups: (1) polar forces, arising from permanent or induced electric fields associated with both the solute and the molecules of the two phases; (2) non-polar, non-specific dispersion forces (London or Van der Waals forces). This classification of the forces has been employed in many theoretical and practical approaches to the description of retention data in molecular terms¹⁻⁶, although the individual authors differ as to their definition of polar interactions.

To find the structural descriptors best related to the ability of a particular solute to undergo both polar and dispersive interactions is an important step to understanding of the processes of chromatographic separations. Having established the quantitative relationships between the structural parameters and the retention data, one can get a deeper insight into the mechanism of a particular type of chromatographic separation. Conversely, some structural information may be obtained

from the retention data of solutes, which is of paramount importance for correlation analysis with other properties of solutes, *e.g.*, pharmacological activity^{7,8}.

THEORETICAL

For some time, correlation analysis has been applied in chromatography. Usually, however, liquid chromatographic retention data have been correlated with other experimental, free-energy-related parameters or substituent contributions⁹⁻¹². Since the successful application of topological indices¹³⁻¹⁵, correlation analysis has occasionally been used for the description of gas-liquid chromatographic (GLC) retention indices of more or less congeneric groups of compounds. A number of equations have been reported for such homologous or congeneric sets of solutes, relating GLC retention indices to such constitutive-additive properties of solutes as molecular connectivity, refractivity, polarizability, etc. The best correlations found were for the less polar stationary phases and the more congeneric sets of solutes. Intuitively, in this context, the term congeneric means possessing the same or similar ability to undergo one type of interaction with the chromatographic phase, *i.e.*, dispersive or polar interactions. Usually the compounds studied were congeneric in respect of their polarities, whereas their dispersive properties could vary significantly as was the case with homologous series.

In attempts to correlate retention data with structural parameters for more diverse sets of solutes the dipole moment has been considered as a quantitative measure of solute polarity, either determined experimentally or calculated quantum chemically. The two-parameter correlation equations describing GLC retention indices in terms of molar refractivity (dispersion parameter) and dipole moment (polarity parameter) have been of limited statistical value in the case of a non-congeneric set of substituted phenols⁵.

In 1983 Buydens *et al.*¹⁶ published the results of studies on the prediction of GLC retention indices for a group of solutes consisting of aliphatic ethers, esters, alcohols, ketones and aldehydes. To get meaningful structure-retention relationships, subsets of mono- and bifunctional derivatives were considered separately. Multiple regression analysis was carried out with the following quantum chemically (CNDO/2) calculated electronic parameters: magnitude of the total dipole moment; sum of the absolute values of the charges in a given molecule; sum of the corresponding charges on the atoms constituting the functional group and on the atoms in α position to this functional group. The last parameter, which may be considered as describing a local molecular polarity, was found to be much more important for solute retention than the total dipole moment.

In 1976 Karger *et al.*² and Scott¹ observed that compounds like dioxane or 1,4-dichlorobutane have an overall dipole moment of approximately zero (two dipoles in opposite directions cancel) but behave as polar solutes.

Bearing in mind all these observations, we came to the conclusion that the overall dipole moment does not properly reflect the ability of a solute to take part in polar interactions during the chromatographic process. In a search for a suitable submolecular measure of solute polarity, we turned our attention to the largest difference in atomic charges.

To parametrize atomic charge differences in a given molecule we applied stan-

standard quantum chemical calculations, using the commercially available standard computer programs. Thus, at present one need not be a professional quantum chemist to generate structural data of interest for correlation analysis. To calculate molecular parameters from orbitals, the now classical Roothaan method is commonly applied which involves the complete neglect of differential overlap (CNDO/2 method). This semiempirical approximate molecular orbital method, in which only the valence electrons are considered explicitly, is recognized as satisfactorily reflecting the electronic properties of solutes.

First we chose for correlation analysis a relatively non-polar GLC stationary phase, OV-101. The set of solutes consisted of primary, secondary and tertiary, saturated and unsaturated alkyl and heterocyclic amines. Thus, the solutes were not congeneric in respect of their dispersive and polar properties, but on the other hand the nitrogen lone electron pair present in each compound made feasible some additional interactions with the stationary phase, *e.g.*, charge-transfer interactions.

EXPERIMENTAL

Chromatography

Gas chromatographic studies were carried out using a Pye Unicam Series 104 apparatus with a flame ionization detector. A coil-shaped Pyrex glass column (1 m \times 2 mm I.D.) was silanized with a 5% solution of dimethylchlorosilane (DMCS) in toluene. The column was next washed with pure toluene and heated in the chromatograph oven at 300°C. The stationary phase, 5% methyl phenyl silicone (OV-101, Applied Science) on Chromosorb W HP (80–100 mesh), was packed into the column. The carrier gas (argon) flow-rate was 30 cm³/min.

The solutes under study were injected into the column either as pure liquids or as solutions.

The detector temperature was fixed at 250°C. Several oven temperatures (precision \pm 0.1°C) were applied in order to study the temperature dependence of the retention times: the respective plots were extrapolated to 130°C. The retention times were measured with an electronic timer with a mean standard deviation of 0.1 sec. For any given amine or *n*-alkane standard, the difference between the retention time of the solute and that of a non-retained gas, methane was calculated. Each measurement of retention time was done in triplicate and the results were averaged. The Kováts retention indices calculated are given in Table I.

Quantum chemical calculations

The calculations were done by the CNDO/2 molecular orbital method. The program^{17,18} was adapted for the RIAD system computer. Standard values of the bond lengths and angles were assumed¹⁹. The quantum chemical parameters used subsequently in correlation analysis are given in Table I. These are: (1) total energy, E_T , assumed by us as a bulk measure of the solute's ability to participate in non-specific interactions with a stationary phase; (2) energy of the highest occupied molecular orbital, E_{HOMO} , the parameter related to the ability of the solute to form charge-transfer complexes; (3) submolecular polarity parameter, A , introduced by us as a measure of the solute's ability to take part in polar solute-stationary phase interactions.

TABLE I
KOVÁTS RETENTION INDICES NORMALIZED TO 130°C AND STRUCTURAL PARAMETERS OF THE AMINES STUDIED

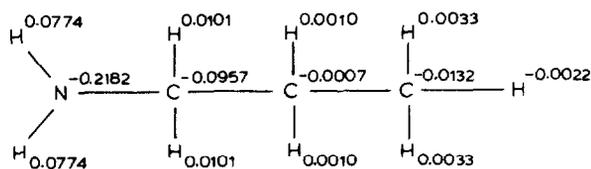
No.	Solute	Retention indices* (I_{ov-101})	Temperature of column** (°C)	Total energy, E_T (a.u.)***	Polarity parameter, Δ (electrons)	Energy of HOMO (a.u.)	Dipole moment, μ (D)	Molar refractivity (MR)
1	Allylamine	463	100-140	-38.100	0.3101	-0.4964	1.7229	18.926
2	<i>n</i> -Butylamine	553	100-150	-48.597	0.3110	-0.4843	1.8984	24.044
3	<i>sec</i> -Butylamine	471	100-150	-48.596	0.3413	-0.4847	1.8975	24.044
4	<i>tert</i> -Butylamine	501	100-140	-48.593	0.3600	-0.4895	1.8971	24.044
5	<i>n</i> -Pentylamine	635	110-150	-57.281	0.3113	-0.4767	1.9067	28.688
6	<i>n</i> -Propylamine	466	100-140	-39.917	0.3139	-0.4936	1.8930	19.400
7	Isopentylamine	615	110-150	-57.272	0.3145	-0.4793	1.8896	28.688
8	Isopropylamine	469	100-140	-39.921	0.3439	-0.5048	1.8317	19.400
9	Diallylamine	660	110-180	-62.519	0.2833	-0.4686	1.7733	32.572
10	Di- <i>n</i> -propylamine	694	100-150	-65.964	0.2917	-0.4655	1.8426	33.520
11	Diethylamine	527	100-140	-48.602	0.2897	-0.4767	1.8613	24.323
12	Methyl- <i>n</i> -pentylamine	706	110-180	-65.963	0.2786	-0.4626	1.8929	33.520
13	Methyl- <i>n</i> -hexylamine	871	110-190	-74.583	0.2786	-0.4590	1.9108	38.164
14	Methyl- <i>n</i> -butylamine	630	110-180	-57.279	0.2786	-0.4670	1.8793	28.876
15	Di- <i>n</i> -butylamine	943	130-210	-83.242	0.2875	-0.4595	1.8501	42.808
16	Pyrazine	696	110-180	-54.621	0.1723	-0.4564	0.0048	21.482
17	Pyridine	692	110-200	-50.866	0.2338	-0.4707	2.1019	23.890
18	β -Picoline	841	110-200	-59.554	0.2198	-0.4652	2.1261	28.508
19	3-Chloropyridine	890	110-210	-66.356	0.2570	-0.4757	2.1976	26.349
20	Chloropyrazine	895	110-210	-70.038	0.3024	-0.4658	2.1565	28.757
21	2-Chloropyridine	870	150-210	-66.245	0.3120	-0.4737	3.6424	26.349
22	4-Cyanopyridine	955	150-220	-68.643	0.2471	-0.4809	0.9618	28.205

* Retention index value extrapolated 130°C.

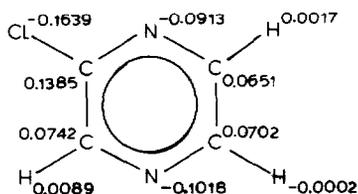
** Temperature range over which a linear dependence of retention data on temperature was found.

*** a.u. = Atomic units.

The parameter, Δ , represents the largest difference in atomic charges. To determine it, electron densities on particular atoms in the molecule are calculated and the two atoms which have the highest electron excess and deficiency respectively yield Δ (see Fig. 1 for illustration).



$$\Delta = 0.0957 - [-0.2182] = 0.3139$$



$$\Delta = 0.1385 - [-0.1639] = 0.3024$$

Fig. 1. Examples of electron excess charge densities and polarity parameter, Δ .

For the sake of comparison the overall dipole moment, μ , was calculated for each solute by the CNDO/2-MO method. The numerical data are given in Table I along with the molar refractivities, MR, calculated by summing individual bond refractivities according to Vogel *et al.*²⁰.

RESULTS AND DISCUSSION

The multiple regression analysis carried on with the data in Table I has yielded several statistically valid equations. The "best" is eqn. 1

$$I_{OV-101} = (301.88 \pm 306.70) - (11.66 \pm 2.74)E_T - (1016.80 \pm 752.66)\Delta$$

$$n = 22, s = 67.45, R = 0.93 \quad (1)$$

relating the Kováts retention index normalized to 130°C, I_{OV-101} , to the quantum chemically calculated total energy, E_T , and polarity parameter, Δ , where n is the number of compounds considered, s the standard deviation from the regression equation and R the multiple correlation coefficient; the 95% confidence limits calculated by the t-test are indicated. The equation is significant at the $p < 0.0001$ significance level.

The one-parameter equation relating I_{OV-101} to E_T is also significant at

$p < 0.0001$, but the correlation coefficient is much lower, $R = 0.89$. The equation relating I_{OV-101} to Δ is of lower statistical significance: $p < 0.06$; $R = 0.51$. However, in eqn. 1 the term Δ ($p < 0.006$) and E_T ($p < 0.0001$) are of high statistical significance.

The statistical analysis for the whole set of compounds studied confirms the predominance of non-specific interactions in the retention of amines on the relatively non-polar stationary phase OV-101. On the other hand, polar interactions, as characterized by the submolecular polarity parameter, Δ , are also of importance. A negative sign of the coefficient for Δ in eqn. 1 indicates that, in the case of two solutes having the same E_T but different Δ , the more polar one would be less strongly retained on the non-polar OV-101 phase.

The polarity parameter, Δ , proposed here characterizes the ability of solutes to participate in polar interactions much better than does the overall molecular dipole moment, μ . Replacing Δ in eqn. 1 by μ is not valid statistically; the significance level for μ is $p < 0.50$. This observation supports the assumption that interactions of the dipole-dipole type between the stationary phase and the solute concern the local dipoles in the molecules. Thus, the use of the overall molecular dipole moment as a measure of solute polarity may be misleading. The same may be true in the case of other types of intermolecular interactions, *e.g.*, drug-receptor interactions.

Eqn. 1, although highly significant, describes about 87% ($R^2 \times 100\%$) of the retention index changes in structural terms; more than 10% of the changes remains unexplained. This may be the result of errors in the determination of retention indices, especially upon extrapolation of experimental data. Another cause of the deviation of eqn. 1 from the ideal is the more or less approximate nature of the quantum chemical data, especially as the actual molecular conformations, bond lengths and angles are not known precisely.

The main source of error, however, is probably the approximate nature of the polarity measure. The parameter Δ is a better measure of the polar interactions of the solutes with the stationary phase than is the overall dipole moment. On the other hand, the polar interactions considered are actually the sum of contributions from several local intramolecular dipoles in closest contact with the phase; Δ is the best approximation of these interactions for the largest of the dipoles.

One can now ask whether any improvement of eqn. 1 is possible with the data given in Table I. In fact, eqn. 2 gives better statistics than eqn. 1 but its validity may be questioned:

$$I_{OV-101} = (-3076.91 \pm 1822.57) - (16.59 \pm 3.34)E_T - (1988.44 \pm 766.38)\Delta - (7098.45 \pm 3798.55)E_{HOMO}$$

$$n = 22, s = 50.26, R = 0.96 \quad (2)$$

All the parameters used in eqn. 2 are significant, at least at the $p < 0.0005$ level, but there is quite a strong intercorrelation between some regression parameters: the correlation between E_T and E_{HOMO} is $R = 0.77$. For comparison, the intercorrelation between E_T and Δ is only $R = 0.30$, which means that only 10% of the information provided by one of the parameters is contained in the other one.

To examine further the significance of E_{HOMO} for retention we excluded compounds 1, 6, 8 and 15 in Table I from the regression analysis, in order to decrease

the intercorrelation between E_T and E_{HOMO} from 0.77 to 0.53. The equation derived for the remaining eighteen solutes has the form:

$$I_{\text{OV-101}} = -(3143.57 \pm 2284.40) - (17.38 \pm 4.15)E_T - (1919.95 \pm 875.68)\Delta - (7106.91 \pm 4856.36)E_{\text{HOMO}}$$

$$n = 18, s = 55.05, R = 0.95 \quad (3)$$

The term E_{HOMO} in eqn. 3 is significant at the $p < 0.004$ level.

Thus, we can say that there is some evidence that E_{HOMO} is meaningful for quantitative description of the retention of amines on the non-polar phase OV-101. This, in turn, would suggest that in the process of chromatographic separation the charge-transfer complexes are formed between a donor solute molecule and the stationary phase acting as an electron acceptor.

It seemed interesting to compare the total energy, E_T , and molar refractivity, MR, as measures of the ability of the molecules to take part in non-specific, non-polar interactions. Molar refractivity has often been used in correlation analysis of retention data. The total energy calculated by the CNDO/2 method has been found to correlate with Kováts indices for a homologous series of esters²¹. One may expect that, in the case of closely congeneric sets of solutes, the two parameters MR and E_T will be strongly intercorrelated. Thus, both of them can serve as a good quantitative measure of the abilities of the compounds to participate in dispersive interactions. In the case of a more diverse class of substances the total energy seems to be the more reliable parameter. Some evidence in support of this assumption may be gained from the data given below. If the subseries of primary, secondary and tertiary (heterocyclic) amines are analysed separately, the correlations between the retention indices and MR are similar to those between retention indices and E_T , but only for the primary and secondary amines (Table II). For heterocyclic amines differing significantly in structure, the total energy, E_T , is much better correlated with retention than molecular refractivity.

TABLE II

CORRELATION COEFFICIENTS OF LINEAR EQUATIONS RELATING KOVÁTS INDICES, $I_{\text{OV-101}}$, TO MOLAR REFRACTIVITY, MR, AND TOTAL ENERGY, E_T

<i>Amines</i>	<i>Correlation with molar refractivity (MR)</i>	<i>Correlation with total energy, E_T</i>
Primary, nos. 1-8 Table I	0.90	0.90
Secondary, nos. 9-15 Table I	0.98	0.98
Tertiary (heterocyclic), nos. 16-22 Table I	0.85	0.94

We would stress that the correlation analysis of Kováts indices with the CNDO/2 data provides evidence to support the importance in retention of the molecular interactions, which generally can be separated into non-specific, dispersive interactions and rather complex polar interactions. To parametrize dispersive interactions, the quantum chemically calculated total energy may be applied. E_T has little or no chemical meaning. It is likely, however, that it reflects the non-specific properties of the solutes indirectly, and that other more chemically significant parameters are contained within it.

The submolecular polarity parameter proposed here, defined as largest difference in atomic charges in the molecule, seems to be more reliable for characterizing a molecule's ability to participate in polar interactions than is the overall dipole moment. The use of both E_T and Δ as molecular descriptors allows the prediction of retention data for test solutes covering a range of polarities.

This study is a preliminary one in the sense that the primary focus has been on the elucidation of non-empirical descriptors of retention, especially as regards molecular polarity. Application of quantum chemical parameters in correlation analysis of retention data allows the prediction of chromatographic behaviour (at least semiquantitatively) for solutes of diverse structures. For polar solutes chromatographed on a phase of low polarity, such an analysis has been successfully performed. In the case of polar stationary phases, the hydrogen bonding with the solute probably affects the solute's electronic structure and has to be considered when calculating quantum chemical parameters.

REFERENCES

- 1 R. P. W. Scott, *J. Chromatogr.*, 122 (1976) 35.
- 2 B. L. Karger, L. R. Snyder and C. Eon, *J. Chromatogr.*, 125 (1976) 71.
- 3 Cs. Horváth, W. Melander and I. Molnar, *J. Chromatogr.*, 125 (1976) 129.
- 4 M. Gassiot-Matas and G. Firpo-Pamies, *J. Chromatogr.*, 187 (1980) 1.
- 5 R. Kaliszan and H. D. Höltje, *J. Chromatogr.*, 234 (1982) 303.
- 6 H. Lamparczyk and A. Radecki, *Chromatographia*, 18 (1984) 615.
- 7 R. Kaliszan, *J. Chromatogr.*, 220 (1981) 71.
- 8 R. Kaliszan, *J. Chromatogr. Sci.*, 22 (1984) 362.
- 9 E. Tomlinson, *J. Chromatogr.*, 113 (1975) 1.
- 10 B. K. Chen and Cs. Horváth, *J. Chromatogr.*, 171 (1979) 5.
- 11 G. Dahlmann, H. J. K. Köser and H. H. Oelert, *Chromatographia*, 12 (1979) 665.
- 12 K. Jinno and K. Kawasaki, *Chromatographia*, 18 (1974) 90.
- 13 R. Kaliszan, *Chromatographia*, 10 (1977) 529.
- 14 R. Kaliszan and H. Lamparczyk, *J. Chromatogr. Sci.*, 16 (1978) 246.
- 15 D. Bonchev, Ov. Mekenjan, G. Protic and N. Trinajstic, *J. Chromatogr.*, 176 (1979) 149.
- 16 L. Buydens, D. L. Massart and P. Geerlings, *Anal. Chem.*, 55 (1983) 738.
- 17 J. A. Pople and G. A. Segal, *J. Chem. Phys.*, 44 (1966) 3289.
- 18 P. A. Dobosh, *Quantum Chemistry Program Exchange*, 11 (1969) 141.
- 19 *Tables of Interatomic Distances and Configurations in Molecules and Ions, Suppl. 1956–1959*, The Chemical Society, London, 1965.
- 20 A. J. Vogel, W. T. Cresswell and J. Leicester, *J. Phys. Chem.*, 58 (1954) 174.
- 21 F. Saura-Calixto, A. Garcia-Raso and M. A. Raso, *J. Chromatogr. Sci.*, 22 (1984) 22.