THE ROLE OF THE STATIONARY PHASE IN GAS CHROMATOGRAPHY

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INTRODUCTION

The large number of stationary-phase liquids described in the literature and offered for sale by suppliers is bewildering to the gas chromatographer. The choice of a suitable stationary phase for a given separation of volatile compounds can be facilitated by a study of the balance of intermolecular forces between the solute and solvent. A qualitative prediction of the retention behaviour of a solute on a given phase can be made using the vapour pressure of the solute and the classification of liquids first proposed by EWELL, HARRISON AND BERG[1]. This has been discussed in some detail by KEULEMANS[2], AMBROSE AND AMBROSE[3] and HARDY AND POLLARD[4].

In gas chromatography, apart from the vapour pressure of the solute, the most important single factor governing the magnitude of the solute retention on a stationary phase is the nett electron donor–acceptor interaction between solute and solvent. The most common example of this is hydrogen-bond formation. The electron donor–acceptor properties of both solute and stationary-phase liquid can be used as a basis for a quantitative classification of phases and as a basis for a method of identification of unknown volatile compounds by gas chromatography.

MOLECULAR STRUCTURE AND INTERMOLECULAR INTERACTIONS

First we must consider briefly the structure of the molecules, their intermolecular interactions and the factors which govern the specific retention volume of a solute. Non-polar molecules in gas chromatography are confined mainly to gases and saturated hydrocarbons. Polar molecules contain one or more polar atoms or groups together with a neutral or non-polar part, usually saturated hydrocarbon chains. The polar part may be the strongly electronegative atoms F, O or N or electron attracting groups, *e.g.* $-NO_2$, $-C\equiv N$, $-CF_3$ or electron repelling groups such as $-NMe_2$, $-CH_3$, $-CMe_3$, $-OMe$. It is the type and number of polar groups in a molecule relative to the size of the neutral part which determines the polarity or distribution of electrons on the different parts of the molecule. If the molecule contains double bonds with their mobile π-electrons this allows electron attracting or repelling groups to transmit their effects further through the hydrocarbon part of the molecule than is possible with saturated hydrocarbon chains. Most polar molecules can act as electron donors or acceptors or both, given the required environment, but usually one effect is predominant. It is the concentration of electrons or the electron-cloud density which governs the potential donor or acceptor property of the molecule.

In compounds which form hydrogen bonds with electron donors the proton, because of its small size, can present a concentrated area of effective positive charge (low electron density) close to a donor molecule which has an area of high electron density. For example: chloroform has a low concentration of excess electrons spread over the three chlorine atoms leaving a concentrated deficit of electrons on the proton. The compound 1-nitropropane has an excess of electrons on the nitro group and a deficit of electrons on the α-methylene group while the remaining part of the alkyl chain is substantially neutral. The amino hydrogen atoms of primary and secondary amines act as acceptor sites while the nitrogen of tertiary amines act as donors. In aromatic compounds there can be an excess or deficit of π-electrons on each side of the ring depending on whether the substituent groups are electron repelling or attracting. In aromatic compounds both the π-electron atmosphere of the ring and the substituent groups can act as donor or acceptor sites. Steric hindrance may also play an important part in the availability of donor or acceptor sites. For example: the work of FITZGERALD[5,6] shows that 2,6-dimethylphenol has a larger retention volume than the close boiling 3-methylphenol on the donor-type phase diaminodiphenyl sulphone and 2,6-dimethylpyridine has a larger retention volume than the close boiling 3-methylpyridine on the acceptor-type phase tris-(2-cyanoethyl)-nitromethane.

THE FACTORS GOVERNING THE RETENTION VOLUME

Now let us consider the factors governing the retention volume. If ideal behaviour is assumed in the gas phase the partial pressure of a solute 1 above a solution in solvent 2 at temperature $T°$ K is given by:

$$p_1 = Py_1 = x_1\gamma_1 p°_1$$

where x_1 and y_1 are the mole fractions of solute 1 in the liquid and vapour respectively, P is the total pressure, $p°_1$ is the vapour pressure of solute 1 at T °K and γ_1 is the activity coefficient of component 1 (referred to the standard state of pure liquid solute at the same temperature and pressure). The partition coefficient K has been defined[3] as:

$$K = \frac{x_1}{y_1} \cdot \frac{n_s}{n_m} = \frac{P}{\gamma^\infty_1 p°_1} \cdot \frac{n_s}{n_m} = \frac{n_s RT}{\gamma^\infty_1 p°_1}$$

where n_s and n_m are the moles per ml in the stationary phase and mobile phase respectively and γ^∞ refers to infinite dilution. The specific retention volume per gram of stationary phase V_g is given by[3]:

$$V_g = \frac{K\ 273}{\rho_L\ T}$$

where ρ_L is the density of the stationary phase at the column temperature T. Then:

$$V_g = \frac{273\ R}{M_L} \cdot \frac{1}{\gamma^\infty_1 p°_1}$$

where M_L is the molecular weight of the stationary-phase liquid. The absolute retention volume V is given by:

$$V = \frac{NRT}{\gamma^{\infty}{}_1 p^{\circ}{}_1}$$

where N is the number of moles of stationary phase on the column and T is the temperature at which the volumetric flowrate is expressed.

For ideal solutions $\gamma^{\infty} = 1$ and V_g is inversely proportional to the vapour pressure of the compound. However, in gas chromatography there is usually a large molecular size difference between the volatile component and the stationary phase so, even when these are both normal paraffins, there is a small negative deviation from Raoult's law due to entropy effects. This is of the form[7]:

$$\log \gamma^{\infty} = D(n_1 - n_2)^2$$

where n_1 and n_2 are the number of carbon atoms in the solute and solvent and D is a constant having a negative value.

Many volatile compounds and stationary phases form non-ideal solutions so that V_g is controlled by both p° and γ^{∞} and for a given p°, V_g is controlled by the value of γ^{∞}.

The value of γ^{∞} is determined by the balance of intermolecular forces between the molecules of solute 1 and stationary-phase liquid 2. For solutions where excess entropy effects are small we have:

$$\log \gamma^{\infty} = k\,(E_{11} + E_{22} - 2E_{12})$$

where E is the energy of interaction between the molecules. High values of E may be due to dipole, induced dipole and electron donor–acceptor interactions. Let us consider three circumstances.

(1) E_{11} *predominant.* Here the solute has a high positive value of $\log \gamma$ and therefore a small retention volume relative to a compound of the same boiling point which forms an ideal solution. For example: associated compounds such as nitromethane and methanol on the non-polar phase Apiezon L. (This is equivalent to saying that at the high temperature and low concentration in the liquid phase these compounds are much less associated than in the pure liquid state and would therefore have a much higher "effective" p°.)

(2) E_{22} *predominant.* The solute has a high positive value of $\log \gamma$ and therefore a smaller retention volume than a compound of the same boiling point which forms an ideal solution, *e.g.* saturated hydrocarbons on a glycerol phase.

(3) E_{12} *predominant.* Here electron donor–acceptor interaction between the solute and solvent leads to a negative value of $\log \gamma$ and thus to long retention times, *e.g.* 1,1,2-trichloroethane on Reoplex 400 or primary amines on polyethylene glycol.

PIEROTTI *et al.*[8] have studied and correlated the value of γ^{∞} for a wide range of solutes and solvents.

The effects of these intermolecular interactions are often larger at lower temperatures.

CLASSIFICATION OF STATIONARY PHASES

An approximate measure of the electron donor or acceptor properties of a stationary phase can be obtained from the ratio of the retention volumes of two compounds of about the same boiling point, one an acceptor and one a donor. Table I shows examples of this method based on retention data measured at 125° C on a number of

TABLE I

ACCEPTOR/DONOR RETENTION RATIOS ON PHASES

Phase*	Ratio of retention volumes (125°C)				
	$\frac{CHCl_3}{CCl_4}$	$\frac{CHCl_2\text{-}CH_2Cl}{Dioxane}$	$\frac{Cyclohexanol}{Cyclohexanone}$	$\frac{Pyrrole}{Pyridine}$	$\frac{Aniline}{NMe_2\text{-}aniline}$
REO	1.66	1.32	1.17	—	2.52
XF 1150	1.65	1.64	—	2.14	1.79
DGS	1.10	1.26	—	1.73	2.88
APL	0.665	1.74	0.981	0.676	0.48
m-Bis	0.890	1.32	—	0.862	0.70
ZONYL	0.925	0.595	—	0.435	0.91
FCP	0.874	0.572	0.622	0.700	0.705
QF 1	0.840	0.986	—	0.714	0.706
AROCLOR	0.636	0.720	—	0.705	0.507
TNB	1.25	0.568	0.490	—	1.01

* The phases are identified under Fig. 1.

phases using the apparatus and methods described by BROWN[9]. A high value of the ratio indicates a donor-type phase while a low value indicates an acceptor-type. The selectivity of a given phase for a particular separation has been determined by BAYER[10] who uses the ratio of retentions of two homologous series corrected to identical boiling point as a selectivity coefficient. A similar approach has been used by HUEBNER[11] who used the retention ratio of methanol to alkane to determine the "polarity index" of surface active agents.

The use of the V_g of only two compounds, donor and acceptor, to determine the properties of a stationary phases does not give much information about the relative polarity of the phase and does not permit one to distinguish readily between a non-polar phase and one which has both donor and acceptor properties. If, however, three test compounds are used, one non-polar, one an acceptor and one a donor it is possible to determine the relative polarity of the phase as well as its donor or acceptor potential. If V_n, V_a and V_d are the retention volumes of the non-polar, the acceptor and donor compounds on the phase we can calculate the "retention fractions".

$$F_n = V_n/(V_n + V_a + V_d)$$

and similarly F_a and F_d and these can be plotted on a triangular graph. The retention volumes of *n*-decane, 1,1,2-trichloroethane and dioxane were measured on a number of phases at 125° and the retention fractions calculated to give the classification of phases shown in Fig. 1. The non-polar phases are close to the *n*-decane apex and have high values of F_n and the polar phases have low values of F_n. The acceptor-type

phases are towards the dioxane apex and have a high value of F_d and a low value of F_a, while the donor-type phases are towards the trichloroethane apex and have a high value of F_a and a low value of F_d.

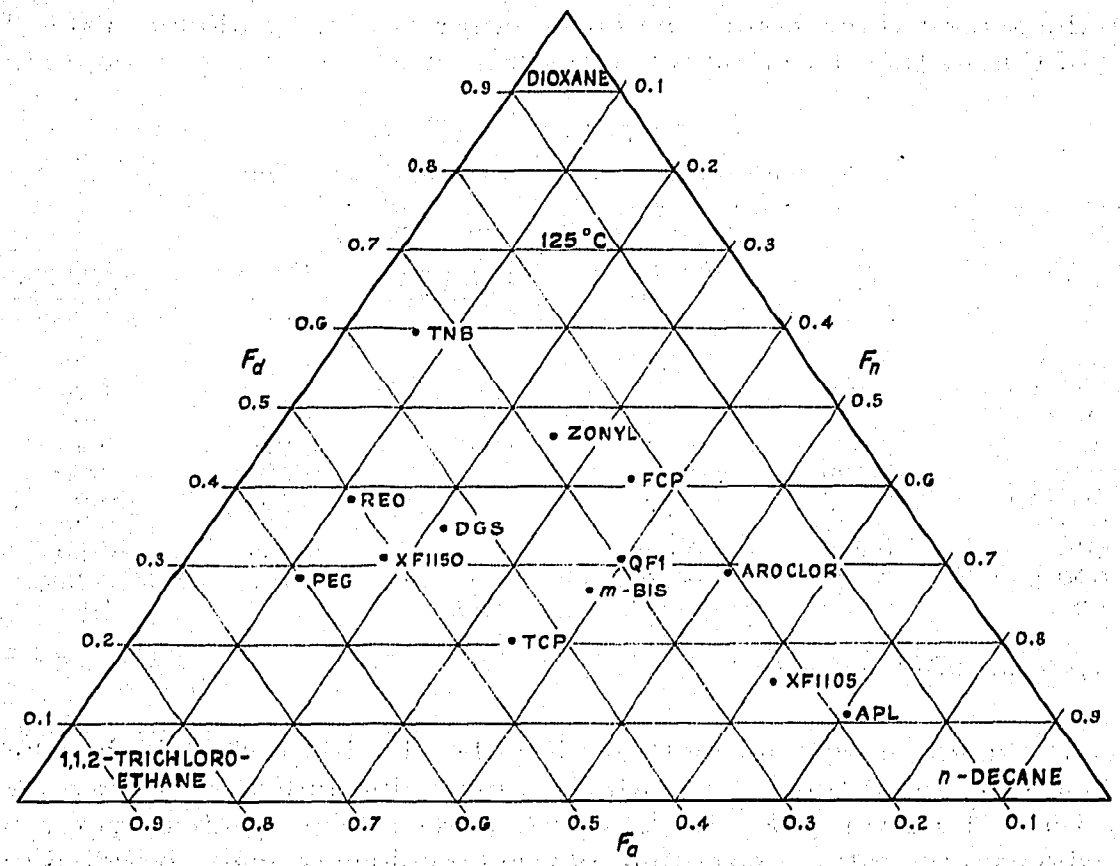

Fig. 1. Classification of stationary phases. F_n, F_a, F_d are retention fractions (see text) calculated from retention data measured at 125° by the author. The phases are: APL = Apiezon L (Metropolitan, Vickers); XF1105, XF1150 = cyanoethylated silicones (General Electric) with 5% and 50% cyanoethyl groups; AROCLOR = Aroclor 1262 (Monsanto) chlorinated diphenyl; QF1 = fluorinated silicone (Applied Science Labs.); *m*-Bis = *m*-bis-(*m*-phenoxyphenoxy)-benzene (Eastman); TCP = tricresyl phosphate (Albright and Wilson); Zonyl = Zonyl E7 (Du Pont) pyromellitic perfluoro ester; DGS = diethylene glycol succinate (Research Specialities Co.); PEG = polyethylene glycol 1500 (Carbide and Carbon); REO = Reoplex 400 (Geigy); TNB = 1,3,5-trinitrobenzene; FCP = diester of tetrachlorophthalic acid and 1-H,1-H,5-H-octafluoro-1-pentanol.

Similarly, using the retention data of McNAIR[12], TENNEY[13] and BROWN[9] for *n*-hexane, ethanol and 2-butanone at 100° C we have the classification of the phases shown in Fig. 2. The position on the triangular graph for a given phase is determined by the choice of the three test compounds and these can be varied to suit a particular problem. The "functional group retention ratio" to be described below can be used with advantage instead of the retention volume of a single compound in the classification of phases by this method.

The number of donor-type phases available to the gas chromatographer is adequate, but there are few strong acceptor-type phases suitable for use over a range of temperature. Many strong acceptor compounds have a rigid molecular structure which gives them melting points close to a temperature where they have an appreciable vapour pressure. The compound di-*n*-butyl tetrachlorophthalate is a useful

weak acceptor-type phase (Fig. 2) but it has sufficient alkyl content to give appreciable retention values for aliphatic hydrocarbons. If this compound could be modified by replacing the butyl groups by more strongly electron withdrawing non-hydrocarbon

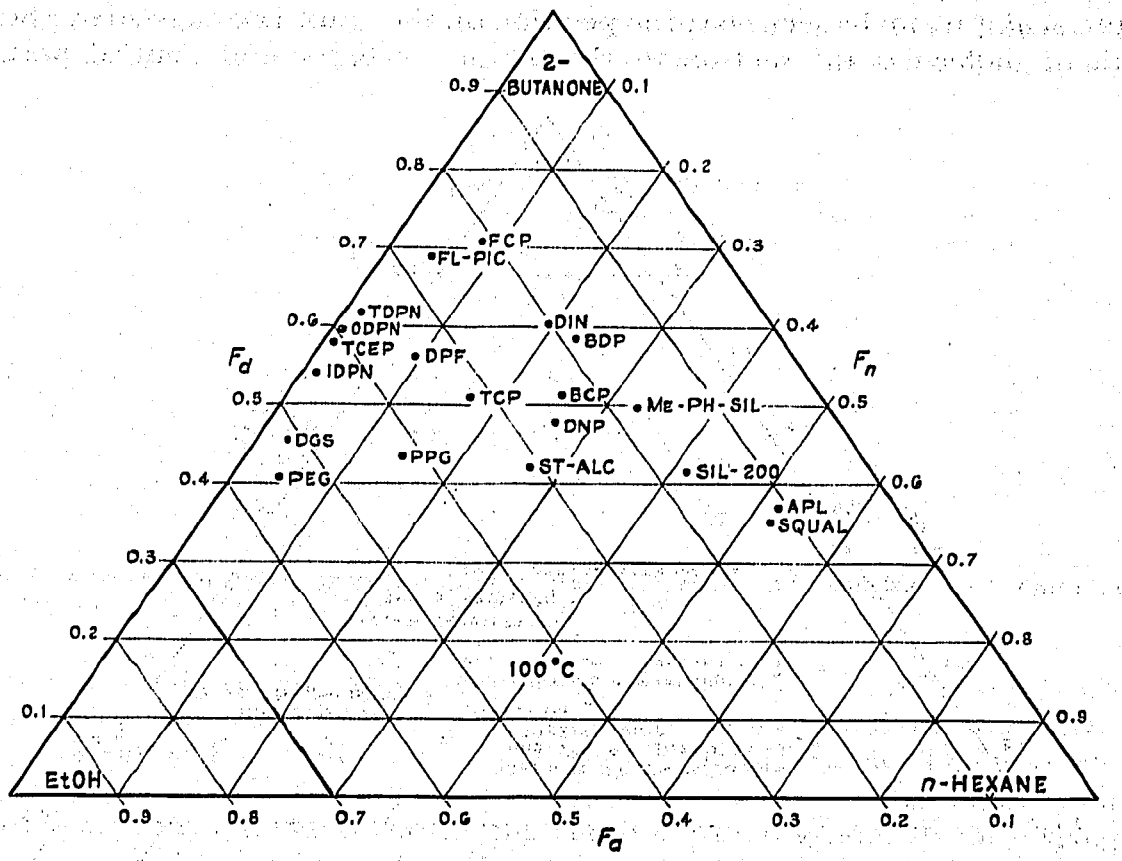

Fig. 2. Classification of stationary phases. F_n, F_a, F_d are retention fractions (see text) calculated from the retention data at 100° C and 67° C of BROWN[9], MCNAIR[12], TENNEY[13], and RAUPP[14]. The phases are: APL = Apiezon L; SQUAL = squalane; Sil-200 = Silicone DC-200; Me-Ph-Sil = methylphenyl silicone (General Electric 81705); St-alc = stearyl alcohol; DNP = dinonyl phthalate; BCP = di-*n*-butyl tetrachlorophthalate; BDP = benzyldiphenyl, DIN = di-*n*-octyl ester of 4,4-dinitrodiphenic acid; TCP = tricresyl phosphate; PEG = polyethylene glycol; PPG = polypropylene glycol; DGS = diethylene glycol succinate; DPF = diphenyl formamide; IDPN = iminodipropionitrile; ODPN = oxydipropionitrile; TDPN = thiodipropionitrile; TCEP = 1,2,3-tris-(2-cyanoethyl)-propane; FL-PIC = fluorene picrate; FCP = diester of tetrachlorophthalic acid and 1-H, 1-H, 5-H-octafluoro-1-pentanol.

groups a more polar and more strongly acceptor-type phase should be obtained. The diester of tetrachlorophthalic acid with 1-H,1-H,5-H-octafluoro-1-pentanol was made and on testing proved to be a much stronger acceptor than the butyl ester as can be seen from Fig. 2. This stable fluoroester is a liquid at room temperature and has a boiling point of 180° C/0.6 mm Hg.

IDENTIFICATION OF ORGANIC COMPOUNDS BY GAS CHROMATOGRAPHY

If we reverse the procedure just described for the classification of phases and run a number of compounds on three selected phases, one neutral, one an acceptor and one a donor and plot the three "affinity fractions" analogous to the "retention fractions"

on a triangular diagram we have a method for studying the donor–acceptor properties of the volatile compounds. In an earlier communication[15] the author proposed this method for the identification of compounds by gas chromatography. From Fig. 1 of the earlier communication and from Fig. 3 of this paper which is based on data of HORNING *et al.*[16] it can be seen that the position on the graph is governed by the relative strength of molecular interaction of the donor, acceptor and neutral parts of the

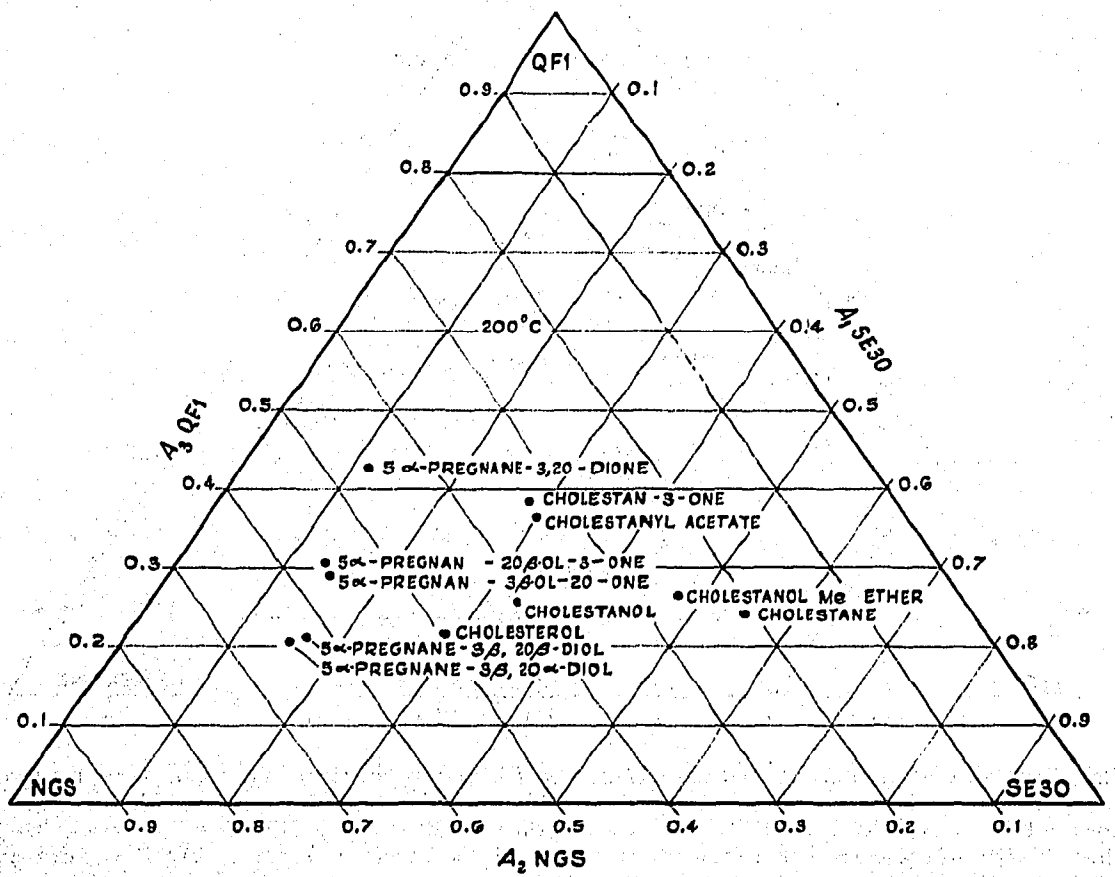

Fig. 3. Identification of steroids. A_1, A_2, A_3 affinity fractions (see BROWN[15]) from the data of HORNING *et al.*[16]. Phases: SE 30 = silicone; NGS = neopentyl glycol succinate; QF 1 = fluorinated silicone.

molecules with the stationary-phase liquids, the position of a given compound is fixed for 3 given phases and the position of a member of a homologous series depends on the length of the hydrocarbon chain.

The value of this method of identifying volatile compounds by gas chromatography would be improved considerably if the effect of hydrocarbon chain length on the retention volume could be eliminated and we could study the behaviour of the functional groups alone. This can be done by taking the ratio or log of the ratio of V_g of a compound RX containing the functional group X to the V_g of a suitable homomorph hydrocarbon, *e.g.* V_g *n*-hexanol/V_g *n*-hexane or nitrobenzene/benzene. For a homologous series of compounds containing the same functional group we should expect this to be a constant at a given temperature as the graphs of log V_g against carbon number for two homologous series are often almost parallel straight lines. Values of the "functional group retention ratio", V_g RX/V_g RH for a number of

different homologous series on various phases are shown in Table II. These were calculated from the data of BROWN[9], MCNAIR[12] and RAUPP[14] using extrapolation of the plot of V_g RH against carbon number to obtain values for V_g of the two lower hydrocarbon homomorphs. It can be seen that the functional group retention ratios are constant to within the accuracy of the experimental data. The choice of homomorph hydrocarbon can be made to suit the problem; for example, the homomorph

TABLE II

FUNCTIONAL GROUP RETENTION-RATIOS

*Phase**	*DIN*	*APL*	*FL-PIC*	*DNP*	*SQUAL*	*ADOL-40*		
Temp.(C°)	*100*	*100*	*100*	*100*	*100*	*153*	*153*	*153*
Group	$>C=O$	$>C=O$	*-OH*	*-OH*	*-OH*	*-OH*	*-Br*	*-Cl*
Carbon No. 3	—	—	87	8.9	3.4	15.0	11.0	6.25
4	—	—	81	9.8	3.8	15.6	11.0	6.32
5	11.3	2.97	84	9.5	3.8	15.6	10.8	6.28
6	11.3	3.04	86	8.9	3.9	15.5	10.4	6.15
7	11.1	2.92	—	8.6	4.0	14.9	—	—
8	10.7	2.98	—	—	3.9	15.0	—	—

* These data were calculated from the data of BROWN[9], MCNAIR[12] and RAUPP[14]. The phases are identified under Fig. 2. Adol-40 is 9,10-octadecen-1,12-diol.

for 2-pentanone could be *n*-pentane or *n*-propane if the functional group is taken as $-CO-CH_3$, or for the study of the acceptor interactions of chloroform with donor phases the homomorph could be taken as fluorotrichloromethane. The best choice is probably the hydrocarbon having the same carbon skeleton or the same number of carbon atoms as the compound containing the functional group. In measuring these ratios for compounds containing functional groups which interact strongly with the stationary phase the retention volumes of the homomorph hydrocarbons may be too small to be measured with sufficient accuracy and it is recommended that these be determined from a log V_g against carbon number graph for the higher members of the series.

The functional group retention ratio for a few aromatic compounds has been determined by BORER[17] using benzene as the homomorph. A similar approach has been employed by CLAYTON[18] with steroids. EVANS AND SMITH[19] have used *n*-nonane as a universal homomorph and KOVATS[20] a more complicated function of retention volumes which he named the Retention Index and which EVANS AND SMITH[19] state is, "100 times the carbon number of a hypothetical hydrocarbon having the same retention as the unknown". SWOBODA[21] goes one step further and uses the difference between KOVATS' Index for a compound and for its homomorph. Another method for the identification of unknown compounds from retention data has been proposed recently by MERRITT AND WALSH[22] using the ratio of retention volumes of the unknown on two carefully chosen phases.

Let us return to the functional group retention ratio. The value of this ratio for a given homologous series is a measure of the interaction of the functional group with the stationary phase. In fact $-RT$ ln RX/RH is the nett free energy change ΔG on the transfer of one mole of a compound RX from the vapour at one atmosphere

in the mobile phase to solution at a low given concentration in the stationary phase plus that for removal of one mole of the homomorph from the stationary phase to the mobile phase under the same conditions. Values of the functional group retention ratio or ΔG, measured on one or more stationary phases, are very useful for the identification of unknown compounds. This easily measured thermodynamic quantity which is a quantitative measure of functional group to phase interaction can be divided into its enthalpy and entropy terms by measuring its temperature dependence.

By using the values of the functional group retention ratio on three carefully selected phases it is possible to plot the group retention fractions derived from them on a triangular graph which shows only one point for each class of compounds. Fig. 4 is such a graph based on the data of BROWN[9], MCNAIR[12] and RAUPP[14]. This method can be employed for the identification of unknown compounds provided that a method is available for transforming the unknown compound into its unknown

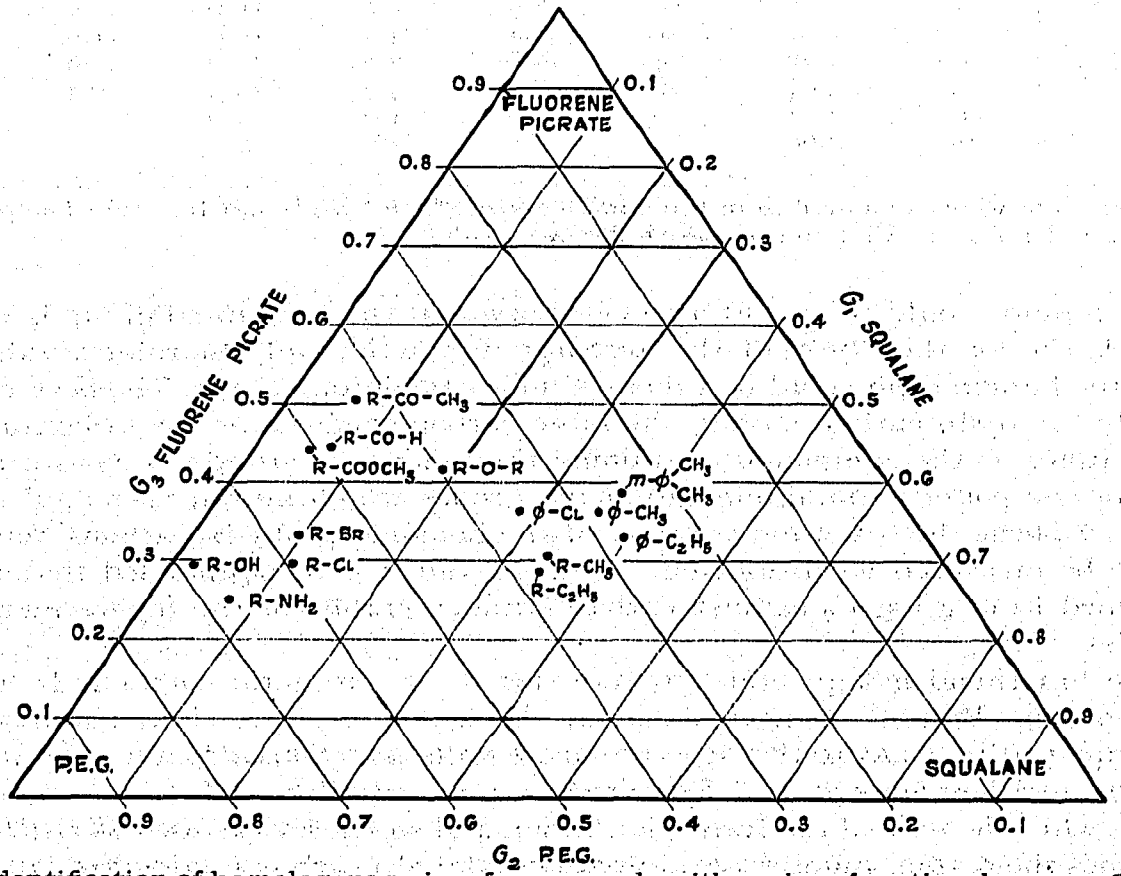

Fig. 4. Identification of homologous series of compounds with various functional groups. G_1, G_2, G_3 group retention fractions (see text) calculated from the data at 100°C of BROWN[9], MCNAIR[12] and RAUPP[14]; R = alkyl group; φ = phenyl group; PEG = polyethylene glycol.

homomorph so that this can also be gas chromatographed on the same three phases. This degradation has been done by micro vapour-phase hydrogenation for a number of oxygen, nitrogen and sulphur compounds by workers at the Bartlesville, U.S. Bureau of Mines Research Laboratories[23,24].

When the donor–acceptor interaction between a polar solute and a polar phase is not too high we find that the free energy change $\Delta G = -RT \ln RX/RH$ is a constant

for a given homologous series on a given phase over a range of column temperatures. However, if the specific donor–acceptor interaction is large there is a difference in orientational freedom on the phase between the polar compound and its homomorph leading to a change in entropy and this gives a variation of ΔG with temperature. For very strong interactions this may lead to variation in entropy and thus in ΔG with the length of the hydrocarbon part of the polar molecule.

SUMMARY

The molecular structure of volatile solutes and stationary-phase solvents, their molecular interactions, and the factors governing the magnitude of the retention volumes are briefly discussed.

A method is described for the classification of stationary phases into types according to their polarity and behaviour as electron donors or acceptors by measuring the retention volumes of three selected compounds on each phase.

A similar method is described for the identification of unknown compounds from their retention data on three carefully selected phases. This method uses the "functional group retention ratio", a quantitative measure of the interaction of the functional group in an homologous series of polar compounds with the stationary-phase liquids, to identify the functional group of the unknown compound.

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