PAPER CHROMATOGRAPHY AND CHEMICAL STRUCTURE
VII. THE SEPARATION OF *meta*- and *para*-DERIVATIVES OF BENZENE

S. MARCINKIEWICZ AND J. GREEN
Walton Oaks Experimental Station, Vitamins Ltd., Tadworth, Surrey (Great Britain)

(Received June 26th, 1962)

INTRODUCTION

In preceding papers[1–6] we have discussed several aspects of the relationship between chromatography and chemical structure. This communication deals with the chromatography of *m*- and *p*-derivatives of benzene. Little information concerning this problem appears to exist. It did not appear to be known whether all *m*- and *p*-isomers differ in R_F or whether only certain pairs do. Although the separation of a few isomeric pairs has been described, other pairs have been found to have identical R_F values. How much of this is due to system differences is unclear, as no detailed chromatographic study of *m*- and *p*-isomers has been made nor has the theoretical basis for any differences been investigated. This study attempts to answer the following questions. Can *m*- and *p*-isomers be separated in neutral, partition systems? Do they obey MARTIN's equation? What factors determine their separation, if any? The chromatography of *o*-derivatives was not considered in detail, although a few such compounds were examined. It is well established that *o*-derivatives often have different R_F values from their *m*- and *p*-isomers, but this arises from different causes, particularly those due to steric crowding, internal hydrogen bonding and polar interactions of the types we have already discussed[2,4].

PERPAR, TIŠLER AND VRBASKI[7] separated *m*- and *p*-nitroaniline in a reversed phase neutral system. TURI[8] was able to separate *m*- and *p*-aminophenol in an acid system, but not in a neutral one. This suggests that, under conditions in which salt formation is possible, separation of isomers with basic groups may be increased. The results of ETTEL, POSPISIL AND DEYL[9] seem to confirm this, for they were unable to separate the aminophenols in ammoniated systems. TURI[8] separated *m*- and *p*-phenylenediamine, however, even in a neutral system and EKMANN[10] also separated these isomers. ETTEL, POSPISIL AND DEYL[9] separated *m*- and *p*-nitrobenzoic acid, but used ammoniated solvents, which must markedly influence salt formation of acids. The *m*- and *p*-isomers of nitrophenol, however, could be separated even in neutral systems[9]. BATE-SMITH AND WESTALL[11], WAGNER[12] and GASPARIC[13] have all described the separation of *m*- and *p*-dihydroxybenzene in both neutral and acid systems. GESSNER AND SMITH[14] achieved a slight separation of *m*- and *p*-chlorophenol in two out of eight systems investigated, but the nature of the stationary phase in these two systems makes it uncertain whether purely partition conditions existed.

It is clear that a theoretical analysis of the separation of *m*- and *p*-isomers (in

common with the analysis of other constitutive effects) cannot be easily made in compounds where other complicating factors may affect chromatography. Thus the separation of acids, as shown by BARK AND GRAHAM[15] is complicated by adsorption, and so might the chromatography of strong bases be. Phenols such as the nitrophenols must be regarded with caution. Thus, *m*- and *p*-nitrophenol differ exceedingly in acid strength, and they may even exist in different tautomeric forms (see later). Considerations such as these seemed to provide a ready explanation for many of the literature separations, except for the case of resorcinol and quinol, which were clearly separable in several partition systems and could not be regarded as compounds in which other phenomena are present. (A possibility, in the case of resorcinol and quinol, was that tautomerism might exist. But many physical studies on these compounds have shown that tautomerism is virtually absent in them. A calculation according to our previously described method[5] showed that, in order to account for the observed separation of resorcinol and quinol on tautomeric grounds, the latter would have to exist as about 20–30 % of diketo form, obviously impossible in view of other physical data.) There appeared to be no way of deciding, *ab initio*, whether any given pair of *m*- and *p*-isomers would have different R_M values nor whether group addition to benzene compounds obeyed MARTIN's equation[15].

In this study, a series of *m*- and *p*-derivatives of benzene were chromatographed in several systems. The following general stipulations were observed.

1. The system must consist of two neutral phases.

2. The effects of adsorption must be shown to be non-existent.

3. If at all possible, each pair of isomers must be run in at least two systems, one of which should preferably be reversed phase. Before it can be safely concluded that two isomers have identical R_F values they should be shown to be inseparable in as many systems as possible. The systems must be such that (a) chromatography is near ideal (demonstrated by the use of substances of known R_F as standards) and (b) the R_F values are, as far as possible, in the range (0.2–0.8), most sensitive to small differences.

EXPERIMENTAL

Chromatography

Whatman No. 4 papers were used for all systems. Confirmatory runs were made on glass paper. The techniques used were, in general, similar to those described earlier[5]. The following systems were used: (A) trigol (triethylene glycol, 10 % w/v in chloroform)/2,2,4-trimethylpentane (iso-octane); (B) trigol (as above)/di-isopropyl ether; (C) (reversed phase) olive oil (10 % w/v in light petroleum)/85 % (v/v) aqueous ethanol; (D) (reversed phase) ethyl oleate (5 % w/v in light petroleum)/25 % (v/v) aqueous ethanol. Some other systems were used with certain compounds and are described in the text.

Visualisation

The aniline derivatives were visualised by exposing the papers to nitrous fumes for a few minutes, then spraying with an alkaline solution of β-naphthol. Other compounds were detected by incorporating a small amount of sodium fluorescein into the trigol phase (5 mg/100 ml $CHCl_3$ solution). After development these papers were sprayed with 5 % (w/v) sodium carbonate solution and then dried, when nearly all the compounds investigated appeared as quenching spots under ultra-violet light. As pre-

viously described[2,3], R_M values were calculated directly from the migration data and R_F values were derived from them. The R_M values of one or two compounds with specially low R_F values, such as phloroglucinol, were determined by the over-running technique, as described previously[5].

RESULTS

Tables I and II give the R_F and R_M values of several *m*- and *p*-derivatives of benzene, and also of some 1,3,5-trisubstituted derivatives (no other trisubstituted isomers were studied as, in all other structural isomers, *ortho*-effects must be present). If system A is considered first, it is seen that only some of the pairs of *m*- and *p*-isomers differ in R_M. The results show that, in order for there to be a difference in the R_M values of the two

TABLE I

R_F AND R_M VALUES OF *meta*- AND *para*-SUBSTITUTED BENZENE DERIVATIVES IN SYSTEM A (TRIGOL/ISO-OCTANE)

Compound	*Isomer*	R_F	R_M
Dinitrobenzene	*m*	0.35	+0.275
	p	0.35	+0.275
Nitroanisole	*m*	0.715	−0.398
	p	0.555	−0.097
Nitrotoluene	*m*	0.84	−0.721
	p	0.81	−0.620
Nitrobenzyl bromide	*m*	0.64	−0.252
	p	0.59	−0.161
Nitro-N-dimethylaniline	*m*	0.785	−0.569
	p	0.335	+0.207
Nitrophenyl acetate	*m*	0.50	0
	p	0.50	0
Nitrophenyl benzyl ether	*m*	0.705	−0.377
	p	0.53	−0.051
Chloronitrobenzene	*m*	0.81	−0.620
	p	0.795	−0.585
Cyanonitrobenzene	*m*	0.205	+0.591
	p	0.205	+0.591
Related compounds			
Nitrobenzene		0.68	−0.332
sym.-Trinitrobenzene		0.11	+0.909

isomers, at least one substituent must be an *ortho/para*-directing group; that is, activating with respect to these positions in the nucleus (INGOLD's + M effect[17]). Thus the two isomers of dinitrobenzene, cyanonitrobenzene, and nitrophenyl acetate, which contain only *meta*-directing groups, had identical R_M values. In marked contrast to the nitrophenyl esters, the benzyl *ethers* of *m*- and *p*-nitrophenol were easily separable in this system (the ether group being *ortho/para*-directing). If the isomers had different R_M values, the *meta*-isomer always migrated faster, irrespective of the type of substituents present. Isomers that showed no chromatographic difference in System A were then rigorously examined in several other systems, but in none of these was separation effected. Thus, the two dinitrobenzenes and the two cyanonitrobenzenes

TABLE II

R_F AND R_M VALUES OF *meta*- AND *para*-SUBSTITUTED BENZENE DERIVATIVES AND SOME *sym*-TRISUBSTITUTED DERIVATIVES IN SYSTEM B (TRIGOL/DI-ISOPROPYL ETHER) AND CALCULATED R_M VALUES

Compound	*Isomer*	R_F	R_M	*Calc.* R_M	R_M — *Calc.* R_M
Dinitrobenzene	*m*	0.81	—0.629		
	p	0.81	—0.629		
Nitrophenol	*m*	0.64	—0.244	+0.182	—0.426
	p	0.525	—0.041	+0.182	—0.223
Nitroaniline	*m*	0.55	—0.081	+0.182	—0.263
	p	0.29	+0.393	+0.182	+0.216
Dihydroxybenzene	*m*	0.100	+0.954	+0.993	—0.039
	p	0.085	+1.031	+0.993	+0.038
Phenylenediamine	*m*	0.044	+1.342	+0.993	+0.349
	p	0.027	+1.556	+0.993	+0.563
Aminophenol	*m*	0.069	+1.300	+0.993	+0.137
	p	0.0475	+1.342	+0.993	+0.207
Chloroaniline	*m*	0.725	—0.420	—0.585	+0.165
	p	0.705	—0.377	—0.585	+0.208
Chlorophenol	*m*	0.795	—0.585		
	p	0.795	—0.585		
Cresol	*m*	0.795	—0.585		
	p	0.795	—0.585		
Cyanophenol	*m*	0.46	+0.068	+0.036	+0.032
	p	0.46	+0.068	+0.036	+0.032
Cyanonitrobenzene	*m*	0.86	—0.775		
	p	0.86	—0.775		
Nitrobenzaldehyde	*m*	0.78	—0.553		
	p	0.78	—0.553		
Hydroxybenzaldehyde	*m*	0.47	+0.057	+0.258	—0.201
	p	0.35	+0.267	+0.258	+0.009
Hydroxybenzoic acid methyl ester	*m*	0.68	—0.319		
	p	0.65	—0.276		
Toluidine	*m*	0.795	—0.585	—0.585	0
	p	0.795	—0.585	—0.585	0
Hydroxybenzyl alcohol	*m*	0.10	+0.954		
	p	0.085	+1.031		
Amino-N-dimethylaniline	*m*	0.635	+0.237		
	p	0.485	—0.009		
N,N-Tetramethylaniline	*m*	>0.95			
	p	>0.95			
Nitro-N-dimethylaniline	*m*	>0.95			
	p	0.78	—0.553		
sym-Trisubstituted derivatives and others					
p-Ethylphenol		0.86	—0.796		
p-Ethylaniline		0.86	—0.796	—0.796	0
3,5-Dinitroaniline		0.069	+1.130	+0.784	+0.346
3,5-Dinitrophenol		0.14	+0.785	+0.784	0
3,5-Dichloroaniline		0.76	—0.500	—0.750	+0.250
3,5-Xylenol		0.85	—0.747	—0.750	+0.003
Phenol		0.725	—0.420		
Aniline		0.725	—0.420		
Phloroglucinol*		0.004	+2.350	+2.406	—0.06

* Over-running technique used.

had identical R_F values in reversed phase System C (0.57 and 0.59 respectively). The two dinitrobenzenes, furthermore, had identical R_F values in the following systems:

0.30 in dimethylformamide/cyclohexane
0.24 in dimethylformamide/iso-octane
0.67 in formamide/cyclohexane–hexane (2 : 1)
0.18 in formamide/iso-octane

As a result of these examinations it was concluded (and the conclusion is an important one, as much of the later work hangs on it) that *m*- and *p*-dinitrobenzene are chromatographically indistinguishable in neutral partition systems. Nitrobenzene and *sym*-trinitrobenzene were then chromatographed and $\Delta R_M(NO_2)$ was calculated as + 0.615. As will be seen from Table I, these four nitrobenzenes (in which steric effects are absent) all obey MARTIN's equation with respect to the additive nature of the nitro group. This means that, whatever the chromatographic effects of interaction of the NO_2 group with the ring, they can be considered identical in these four compounds.

In System B (Table II), many more derivatives could be chromatographed. Once again it can be seen that, whatever the nature of the substituents, if separation occurred the *m*-isomer always migrated faster. Furthermore, as in System A, only when at least one substituent was *ortho/para*-directing did the isomers differ in R_M. However, the converse was not true: some *m*- and *p*-derivatives did not separate, even if one or sometimes both groups were *ortho/para*-directing.

CALCULATIONS AND GROUP ADDITIVITY

It is axiomatic that, if a given pair of *m*- and *p*-isomers have different R_M values, in at least one of them MARTIN's equation for group additivity cannot be obeyed. It is not excluded that this may be so for both isomers. Since the two dinitrobenzenes were shown to have identical R_M values in every system examined, it was useful to take this pair as a starting point. Table III gives the calculations of a series of group ΔR_M

TABLE III

CALCULATED ΔR_M PARAMETERS IN SYSTEM B (TRIGOL/DI-ISOPROPYL ETHER)

Group	*Calculation*	ΔR_M
NO_2	R_M(trinitrobenzene) — R_M(dinitrobenzene)	+ 0.602
Benzene	R_M(dinitrobenzene) — 2 × $\Delta R_M(NO_2)$	— 1.833
OH	R_M(phenol) — R_M(benzene)	+ 1.413
NH_2	R_M(aniline) — R_M(benzene)	+ 1.413
CN	R_M(cyanonitrobenzene) — R_M(benzene) — $\Delta R_M(NO_2)$	+ 0.456
CHO	R_M(nitrobenzaldehyde) — R_M(benzene) — $\Delta R_M(NO_2)$	+ 0.678
Cl	R_M(chlorophenol) — R_M(phenol)	— 0.165
CH_3	R_M(cresol) — R_M(phenol)	— 0.165
C_2H_5	R_M(*p*-ethylphenol) — R_M(phenol)	— 0.376

parameters, obtained only from compounds whose *m*- and *p*-isomers had identical R_M values: that is no assumptions were made as to which compound obeyed MARTIN's equation. (In each case the substitution of the benzene ring by a group X replaces one

aromatic hydrogen atom, whose ΔR_M value is unknown. We therefore include the latter in the former, and $\Delta R_M(X)$ includes the $\Delta R_M(H)$ value. Since all the calculations are made in the same way, no error is incurred.) $\Delta R_M(NO_2)$ was found to be constant in System B. From this, it was possible to calculate the hypothetical R_M value of benzene itself in System B, and hence all the other group ΔR_M parameters as shown in Table III. Using these group parameters, it was possible to calculate what would be the theoretical R_M values of many of the compounds of Table II, if MARTIN's equation was obeyed. These are shown in Table II where they can be directly compared with experimental values.

Several points emerge from these calculations:

1. With the exception of the nitrophenols and aminophenols, most phenol derivatives obeyed MARTIN's equation reasonably. The R_M value of *p*-hydroxybenzaldehyde was in agreement with theory, but that of the *m*-isomer was not.

2. There was a considerably discrepancy in the experimental and calculated R_M values for all the substituted anilines, both di- and tri-substituted.

3. *sym*-Trisubstituted phenol derivatives have R_M values in excellent agreement with calculated values, even when derived from a *m*-substituted phenol whose R_M value is in gross disagreement with theory (*cf.* *sym*-3,5-dinitrophenol and *m*-nitrophenol).

4. $\Delta R_M(NH_2)$ and $\Delta R_M(OH)$ are identical in this system, and $\Delta R_M(CH_3)$ is identical in both *p*-toluidine and *p*-cresol. Differences in the mesomeric moments of the latter molecules must have little effect on $\Delta R_M(CH_3)$.

Study of the R_M values of the aniline derivatives reveals that—in confirmation of a trend already observed—the *m*-substituted isomer usually shows a greater discrepancy than the *p*-isomer; and, indeed, the difference between the calculated and observed R_M values for *p*-nitroaniline, *p*-aminophenol and *p*-chloroaniline is approximately constant, and the difference is approximately twice as large for *p*-phenylenediamine. This suggests that $\Delta R_M(NH_2)$ could, with advantage, be calculated from the latter compound, as follows:

$$\Delta R_M(NH_2) = \frac{R_M(p\text{-phenylenediamine}) - R_M(\text{benzene})}{2} = +1.694$$

If this value is now used to re-calculate the R_M values of substituted anilines (Table IV), aniline and the toluidines show a constant discrepancy, but all the other *p*-substituted anilines fall into line, as do the *sym*-trisubstituted compounds. The *m*-compounds, on the other hand, cannot be correlated so, and clearly they do not obey MARTIN's equation.

Dimethylaniline derivatives also do not obey the additivity principle and a constant additive value for $\Delta R_M[N(CH_3)_2]$ cannot be obtained from the data of Table II, as the following calculations show.

$$\begin{aligned}\Delta R_M[N(CH_3)_2] &= R_M(p\text{-aminodimethylaniline}) - R_M(\text{benzene}) - \Delta R_M(NH_2)\\ &= +0.411\end{aligned}$$

If this value is now used to calculate the R_M value of *p*-nitro-N-dimethylaniline, a value of + 0.182 is found, in pronounced disagreement with the observed R_M value of this compound, — 0.553.

TABLE IV

EXPERIMENTAL AND CALCULATED R_M VALUES OF ANILINES IN SYSTEM B

Compound	*Experimental* R_M	*Calculated* R_M	*Difference*
Aniline	—0.420	—0.139	—0.281
p-Toluidine	—0.585	—0.304	—0.281
m-Toluidine	—0.585	—0.304	—0.281
p-Ethylaniline	—0.796	—0.515	—0.281
p-Nitroaniline	+0.398	+0.463	—0.065
m-Nitroaniline	—0.081	+0.463	—0.544
3,5-Dinitroaniline	+1.130	+1.065	+0.065
p-Aminophenol	+1.300	+1.274	+0.026
m-Aminophenol	+1.130	+1.274	—0.144
p-Chloroaniline	—0.377	—0.304	—0.073
m-Chloroaniline	—0.420	—0.304	—0.116
3,5-Dichloroaniline	—0.500	—0.469	—0.031
m-Phenylenediamine	+1.342	+1.556	—0.214

EFFECTS OF SYSTEM CHANGE ON SEPARATION OF *meta*- AND *para*-ISOMERS

A possibility to be considered at this stage was that the separation of certain *m*- and *p*-isomers could have been due to adsorption effects of the paper. In accordance with a practice we have used previously[3,5], this was tested by chromatographing key compounds in the same partition system as used above, but supported on glass paper. No difference in R_M was found for any compound and this confirms our previous conclusions that R_M values of most compounds (except acids) in systems using an impregnated stationary phase are unaffected by the support. As a further check on the adsorption problem, several compounds were chromatographed in reversed phase system C — since if any adsorption were present it must surely affect the R_M values in reversed phase and direct phase systems in the opposite direction. Although not many simple benzene derivatives run in this system, Table V gives the R_F values of those that do.

TABLE V

R_F VALUES OF *meta*- AND *para*-ISOMERS IN REVERSED PHASE SYSTEM C (OLIVE OIL/85% ETHANOL)

Compound	*m-Isomer*	*p-Isomer*
Dinitrobenzene	0.57	0.57
Chloronitrobenzene	0.67	0.67
Cyanonitrobenzene	0.59	0.59
Nitroanisole	0.70	0.74
Nitro-N-dimethylaniline	0.56	0.64
N,N-Tetramethylphenylenediamine	0.71	0.76
Nitrophenyl acetate	0.78	0.78

In confirmation of the results in other systems, the nitrobenzenes, cyanobenzenes and nitrophenyl acetates were still inseparable. The nitroanisoles, nitrodimethylanilines and tetramethylphenylenediamines (all containing *ortho/para*-directing substituents)

had different R_F values, and as expected, the *p*-isomers of the latter compounds ran *faster* than the *m*-isomers; this again suggests that any difference between the R_F values of *m*- and *p*-isomers is unlikely to be due to adsorption effects.

HALOPHENOLS

The isomeric chlorophenols (although they each contain two *ortho/para*-directing substituents) could not be separated in any of our systems. GESSNER AND SMITH[14], using a formamide/hexane system, found the R_F values of the *m*- and *p*-isomers to be 0.7 and 0.6 respectively, but the separation appeared to depend on the method of impregnation. We compared the two compounds in their system and obtained R_F values of 0.67 and 0.63, but the *p*-chlorophenol spot was diffuse and difficult to locate exactly. When the system was used on glass paper instead of Whatman No. 4 paper, both substances migrated just behind the front and did not separate. With hexane as a solvent, R_F values might be especially sensitive to the amount of moisture in the paper; or, alternatively, adsorption might just begin to play a part when the mobile phase is completely non-polar. In any case, any difference between these two compounds must be considered as marginal and perhaps doubtful except in special systems.

The apparent chromatographic identity of *m*- and *p*-chlorophenol was of especial interest as it indicates that the phenolic ionisation strength has (as we have already suggested[2]) little affect on R_M. It also indicates that the inductive effect of the halogen group (which must produce a different mesomeric displacement in the aromatic ring, depending on whether it is *meta* or *para* to the OH group) cannot be directly and simply related to R_M differences. In order to study this question in more detail all twelve mono-halophenols were chromatographed in reversed phase System D (neither of the trigol systems were suitable for all twelve compounds). The results are given in Table VI (together with the cresols, for comparison) and show that *all three* isomers of each

TABLE VI

R_F AND R_M VALUES OF HALOPHENOLS IN SYSTEM D (ETHYL OLEATE/25% AQUEOUS ETHANOL)

Compound	R_F	R_M
o-, *m*-, *p*-Fluorophenol	0.89	−0.906
o-, *m*-, *p*-Chlorophenol	0.725	−0.417
o-, *m*-, *p*-Bromophenol	0.54	−0.070
o-, *m*-, *p*-Iodophenol	0.385	+0.204
o-Cresol	0.78	−0.547
m-, *p*-Cresol	0.85	−0.767

halophenol have identical R_F values in this system. There is thus not only no separation of *m*- and *p*-isomers, but there is no *ortho*-effect in these compounds. The significance of these results is discussed further below.

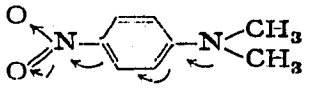
DISCUSSION

Analysis of the problem of *m*- and *p*-isomers revolves round four paramount questions. First, why do some pairs of isomers have different R_M values, while others have the

same R_M value? (Although it is true that non-separation in any finite number of systems cannot, theoretically, be used as an argument for universal non-separation in all other systems, there is sufficient distinction between the two classes of compounds to comprise a valid effect. As shown later, our interpretation of the phenomenon does not in any case exclude the eventual separation of certain of these identical pairs.) Secondly, can the *degree* of separation be correlated with molecular interactions? Thirdly, why, with certain exceptions, do *p*-substituted and *sym*-trisubstituted compounds obey MARTIN's equation, whereas *m*-substituted compounds often do not? Finally, why do *m*-compounds run faster than *p*-compounds (in direct phase), irrespective of the substituents?

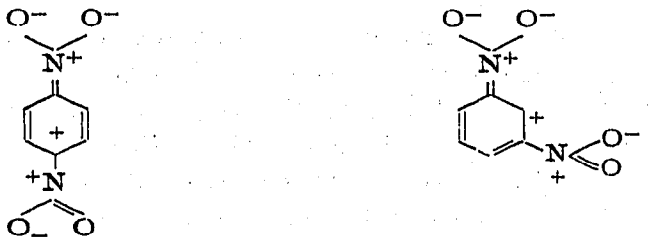
The only previous suggestion that appears to have been made to account for the separation of a pair of *m*- and *p*-isomers has been by FRANC AND JOKL[18]. They considered that MARTIN's equation was, in fact, not obeyed and substituted for it an equation which included a parameter derived from the molecular dipole moment of the compound under study. As we have already shown[2], in alkylbenzenes where the deviation from MARTIN's equation arises simply from polarization in the alkyl group, which must be directly related to the moments of such compounds, such a correction is useful. However, FRANC AND JOKL used it to correlate the R_F values of di- and polyhydroxybenzenes, particularly to explain the separation of quinol and resorcinol. Their view that vectorial dipole moments affect R_F values cannot be supported, for both practical and theoretical reasons. FRANC AND JOKL calculated the R_F values of quinol and resorcinol by assuming they had dipole moments of zero and 1.6 D respectively. In fact, this is an error. Quinol, because it contains two angular groups *para* to each other, has a vectorial dipole moment whose magnitude is dependent on the direction of the O–H bonds[19]. Its value has been variously determined as 2.47 D[20] and 1.40 D[21]. In either case, it is not appreciably different from the dipole moment of resorcinol (1.6 D) and the calculations of FRANC AND JOKL cannot be justified. It seems unlikely, indeed, that the vectorial dipole moment of a compound should affect its partition coefficient. DIKSTEIN[22] has shown that the cohesion energy of a solute enters the solubility parameter and has an important effect in determining R_F, and KETELAAR[23] shows that the Keesom energy of a molecule (that part of the cohesion energy affected by group polarizations) is influenced only by the individual group moments and not by the total molecular dipole, since the former are operative only over small distances of the order of atomic radii. As a consequence, the cohesion energies of molecules such as *o*-, *m*- and *p*-dichlorobenzene (and hence their boiling points, solubilities in regular solutions, etc.) are almost identical, although their molecular dipole moments are quite different. INGOLD[17], considering chemical reactions, remarks that solvation energy differences must be nearly independent of those parts of the molecule that do not change in the reaction, solvation forces being highly localized on solute molecules. The results in Tables I and II offer unequivocal evidence that the dipole moment of a compound cannot affect its R_F value appreciably. The moment of *m*-dinitrobenzene[24] is 4.07 D, and that of *p*-dinitrobenzene is close to zero (both NO_2 groups lying in the plane of the ring)[25]: this difference is perhaps the largest possible between two isomers, and yet these compounds have identical R_F values. The fact that $\Delta R_M(NO_2)$ is additive for *sym*-trinitrobenzene confirms that dipole effects do not directly influence R_M, and much similar evidence can be adduced from Tables I and II. The cyanobenzenes are exactly analogous to the nitrobenzenes: the *m*-compound

must have a large moment and *p*-dicyanobenzene has a moment of zero, as the CN group is linear. Table V shows that both *m*-nitro-N-dimethylaniline and *m*-N,N-tetramethylphenylenediamine run more slowly (reversed phase system) than their respective *p*-isomers. Since the directions of the interaction moments in the two *p*-isomers are quite different, the nitro group reinforcing while the dimethylamino group opposes the mesomeric moment of the other dimethylamino group (I), one would certainly not expect both *m*-isomers to run faster, if the vectorial dipole moment was of importance. In fact, the reinforcing interaction moment of the nitro and dimethylamino groups *para* to each other is as much as + 1.86 D[26], without apparent influence on R_M.



(I)

The separation of *m*- and *p*-isomers cannot be a simple function, either, of the electronic interactions of substituents with the aromatic ring. Such interactions must occur, and indeed they must markedly influence the resonance structures of *m*- and *p*-derivatives, but (as we have concluded previously as a result of our study of alkylated benzenes and ethers[2]) charge separation *by itself* does not seem to affect R_M unless there is a secondary effect on intramolecular or intermolecular hydrogen bonding. Thus the resonance forms of *m*- and *p*-dinitrobenzene involve contributions from markedly different charged structures (II)



(II)

and similar examples are to be found in Tables I and II. Furthermore, the variations in R_M cannot be simply correlated with the inductive effects of substituents. If the electronegativity or electropositivity of substituents were important it would certainly not be possible for compounds as different as the chlorophenols, cyanophenols and di- and trinitrobenzenes all to obey the additivity principle (see Table II), whereas *m*-nitrotoluene which has a different R_F value from *p*-nitrotoluene, clearly does not. The best illustration of this is in the study of the twelve halophenols. DIKSTEIN's thermodynamic derivation[22] of MARTIN's postulate has shown that the partition coefficient of a substance is partly determined by its molar volume. In Fig. 1, we have plotted the R_M values of the four *p*-halophenols against the molar volumes of the various *p*-substituents. Included in the plot is the R_M value of a hypothetical "*p*-cresol" obtained from the R_M value of *p*-cresol, corrected for the hyperconjugation effect of the CH_3 group attached to the aromatic ring, as follows:

System D in this series is identical with System 1 described previously[2] and the hyperconjugation effect of the CH_3 group can be corrected for by considering the

three α-hydrogen atoms of *p*-cresol to be δ-hydrogen atoms. In System D, ΔR_M-(α-hydrogen) is + 0.014 and ΔR_M(δ-hydrogen) is + 0.096 and the correction is therefore 3 × 0.082 or + 0.246. Thus, if *p*-cresol contained no electronic effects, its R_M value would be not — 0.767, but — 0.521, and this is the value included in the plot of Fig. 1. The plot is closely linear, although *p*-fluorophenol runs slightly faster than

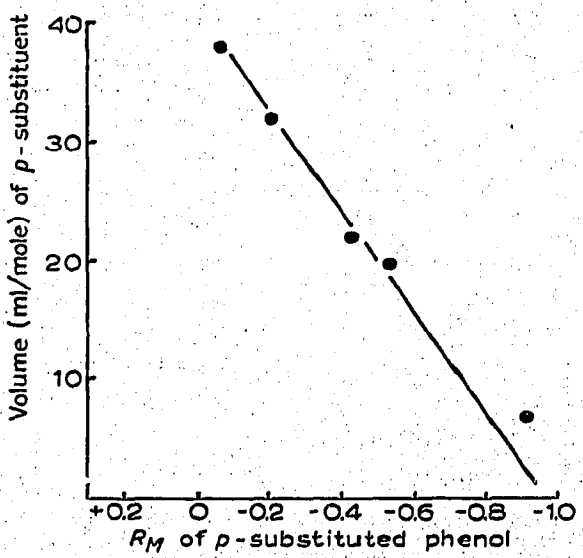
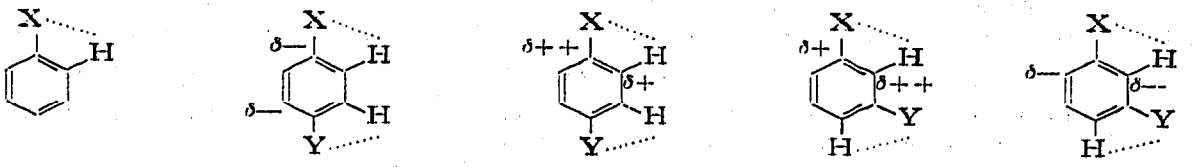


Fig. 1. Relationship between R_M values of *p*-substituted halophenols and molar volume of the halogen atom.

predicted. (The molar volume of *p*-fluorophenol might in fact be rather smaller than calculated. There are some grounds for believing that the C–F bond in aromatic compounds is somewhat shorter than in alkyl fluorides because of the exceptional electronegativity of the fluorine atom[27].) Since the inductive effect of the "corrected" CH_3 group can be considered as zero (or at any rate slightly positive) it is clear that the negative inductive effects of the halogen atoms can play little part in affecting R_M since the only determinant in this series would appear to be molar volume. This is reinforced by the further observation that, although the mesomeric polarizations in *o*-, *m*- and *p*-halophenols differ, the three isomers, in each case, have identical R_F values. It can be concluded, therefore, that the separation of *m*- and *p*-isomers cannot be a simple function of either the direction or magnitude of the electronic polarizations in these molecules, although it is clear that some factor must exist that is related to the electronic effects of groups attached to the ring. Molar volume differences, although they partly determine absolute R_M values (*cf.* DIKSTEIN[22] and above), cannot account for the difference between *m*- and *p*-isomers and, especially, they cannot be used to explain why some pairs separate and some do not.

In preceding parts[2,4–6], we have shown that many deviations from the strict additivity of groups required by MARTIN's equation can be explained in terms of intra- and intermolecular hydrogen bonding and the competition between them. In a study of alkylated phenols and aromatic ethers[2] it was suggested that electronic polarizations operated by affecting the strength of C–H bonds sufficiently to influence solvent-solute interactions. We believe that this is the factor that also determines the separation of *m*- and *p*-benzene derivatives. The energy of a hydrogen bond is usually

considered to be mainly electrostatic in origin, and is highest when the bond joins two strongly electronegative elements, such as O, N or F. In substituted aromatic compounds it is possible for hydrogen bonding to occur as shown (III).



(a) (b) (c) (d) (e)

(III)

The energy of the various C-H···X bonds depicted in (III) can be considered to be a function of (1) the distance C-H···X, (2) the electronegativity of X (or if X is a polyatomic group, of the atom nearest to the hydrogen atom and included in the bond) and (3) the electronic state of the C atom included in the bond. The formation of such an internal hydrogen bond, if it occurs, must have a chromatographic effect, for it will compete with the formation of intermolecular hydrogen bonds between solute and solvents. The energy of an intramolecular C–H···X bond of this sort must normally be low if the carbon atom is uncharged or bears a fractional positive charge. However, under conditions in which the carbon atom bears a fractional negative charge, the energy of the bond may be considerably increased. In a monosubstituted benzene (IIIa), any effect of this kind is automatically included in ΔR_M for the group X. In a *p*-disubstituted compound, two conditions can be distinguished. If Y is a group that is *ortho/para*-directing, that is it repels electrons into the ring by either an inductive or mesomeric mechanism, a fractional negative charge is induced on the *para*-C atom (IIIb), but the *meta*-C atom is hardly affected[17,28] and so the C–H···X bond is relatively unaffected. In this case the chromatographic effect will be similar to that in a monosubstituted derivative and will be included in the parameter $\Delta R_M(X)$. Deviation from MARTIN's equation will thus be minimal. If Y is electron-withdrawing, there is a general de-activation of the ring (IIIc) and since the *meta*-C atom bears a fractional positive charge under these conditions, the C–H···X bond is too weak to compete with the intermolecular hydrogen bond. *Para*-compounds can thus be expected to obey MARTIN's equation, unless other effects are present. Consider now a *m*-disubstituted benzene. If group Y is electron-withdrawing (IIId), the C atom involved in the C–H···X bond is relatively unaffected, as above. If, however, Y is *ortho/para*-directing (IIIe), then the C atom involved in the C–H···X bond bears a fractional negative charge whose strength will depend on the total inductive and mesomeric displacements produced by Y. This will, according to our hypothesis, increase the strength of the intramolecular hydrogen bond, thus affecting the partition coefficient. This hypothesis seems to us to be the only one capable of accounting for the chromatography of *m*- and *p*-isomers. It explains why one *ortho/para*-directing group is necessary for separation and the fact that *m*-derivatives (if separation occurs) always run faster than *p*-derivatives in direct phase systems.

The hypothesis is supported by the possibility of demonstrating a relationship between the magnitude of the charge induced on a *para*-carbon atom by a substituent group or atom and the difference between the R_M values of the *m*- and *p*-isomer.

Table VII lists the differences in R_M between *m*- and *p*-substituted nitrobenzenes (calculated from the data of Table I) and Hammett's *para*-constants for the various substituents[20]. Those constants are the best measurement of the fractional charge induced on an aromatic C atom by a *para*-substituent, due to the sum total of reso-

TABLE VII

DIFFERENCES BETWEEN R_M VALUES OF *meta*- AND *para*-SUBSTITUTED NITROBENZENES (DATA FROM TABLE I) AND HAMMETT'S SUBSTITUENT CONSTANTS

Substituent in nitrobenzene	R_M(meta)— R_M(para)	σ(para)
NO_2	0	+0.788
OCH_3	—0.301	—0.268
CH_2Br	—0.091	+0.184*
$N(CH_3)_2$	—0.776	—0.600
$OCH_2C_6H_5$	—0.326	—0.415
Cl	—0.035	+0.227
CN	0	+0.628
CH_3	—0.101	—0.170

* Figure for CH_2Cl.

nance and polar effects of the substituent on the aromatic ring. (Since the nitro group in aromatic systems obeys MARTIN's equation, by studying substituted nitrobenzenes, we eliminate the difficulty of studying variations due to both substituents together.) Fig. 2 shows the plot of these R_M differences against Hammett's constants, which lie on a smooth curve.

It would follow that, providing a fractional negative charge is induced on the *para*-C atom of a *meta*-isomer, any factor that decreases the C–H···X distance will

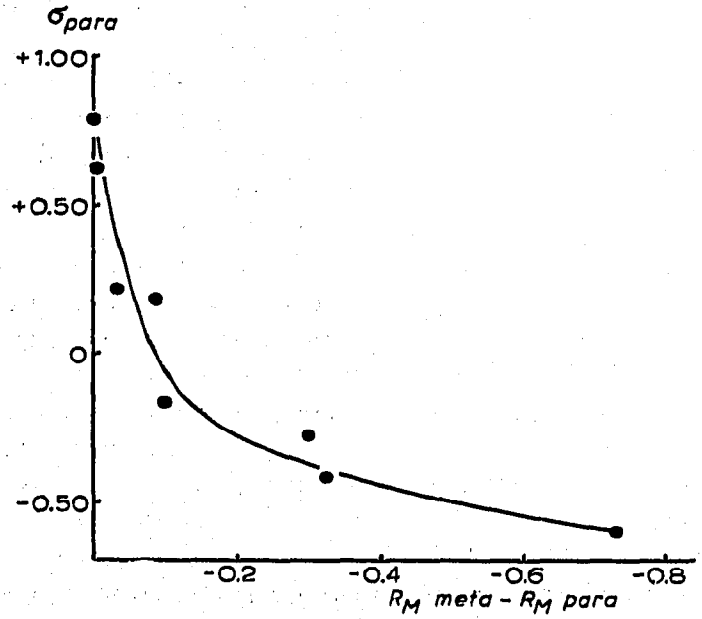
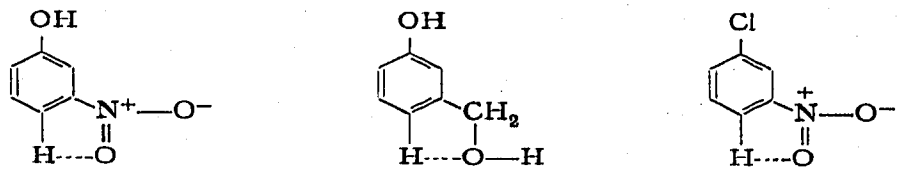


Fig. 2. Relationship between Hammett's σ(*para*) constants and R_M(*meta*) — R_M(*para*).

operate in the same direction as the induced charge and still further increase the strength of the intramolecular hydrogen bond and hence the separation of isomers. This is supported by the results in Tables I and II, which show that the largest differences between isomers occur with compounds such as the nitrophenols, nitroanilines, nitrodimethylanilines and hydroxybenzaldehydes. In the *m*-isomers of these compounds, the C–H···X distance is decreased and bonding facilitated by the possibility of 5-membered ring formation (IVa) and (IVb).



(a) (b) (c)

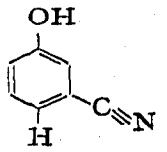
(IV)

The effect of this phenomenon can be observed by comparing the various chloro-substituted derivatives in Tables I and II. Thus, although quinol and resorcinol have different R_M values, *m*- and *p*-chlorophenol do not, because the C–H···Cl hydrogen bond is much weaker than the C–H···O bond. (The marginal separation of chlorophenols in the formamide/hexane system may be partly due to the fact that intermolecular hydrogen bonding between the aromatic C atom and hexane is even weaker.) Although the Cl atom is itself an *ortho/para*-directing group and might be expected to induce a charge as in (V), this must be very weak, as the Cl atom is reluctant to take part in the implied increase in covalency which is shown in the fully mesomeric structure (VI).

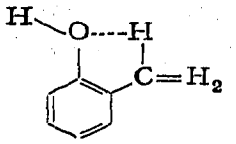
(V) (VI)

The electron-repelling effect of halogen atoms attached to the benzene ring by mesomeric interaction of the lone pair is in fact opposed by the inductive effect of the halogen, which is electronegative. This is why, in aromatic substitution, halogen substituents are uniquely *ortho/para*-directing but deactivating to the ring. Similar conditions must occur in the ground states and lead to uncertainty about the strength of the negative charge induced in the *para*-position by halogens. The general weakness of the effect of Cl is indeed substantiated by the fact that $\sigma(para)$ for this group is positive, whereas for nearly all other *ortho/para*-directing groups it is negative[30]. Unlike *m*- and *p*-chlorophenol, however, *m*- and *p*-chloronitrobenzene do show a slight difference in R_M value. This, we believe, may be due to a slight effect on internal hydrogen bonding due to the possibility of 5-membered ring formation in the *m*-nitro compound as shown in (IVc); the *m*- and *p*-isomers of chloroaniline also show a very slight R_M difference in System B: this we attribute to the powerful electron-repelling

effect of the NH_2 group, compared to the OH group[17], which may strengthen the C–H···Cl bond sufficiently to begin to compete with intermolecular hydrogen bonding. Cyanophenols have identical R_M values, however, since the —C≡N group is linear and any C—H···N bond must be very weak indeed (VII), because of the distance involved.



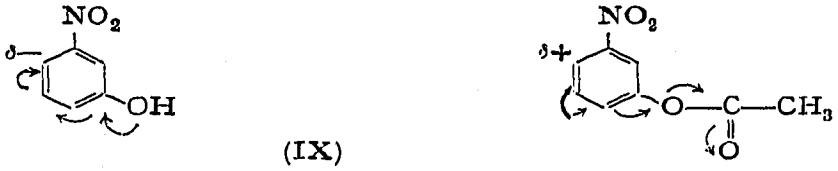
(VII)



(VIII)

(BAKER AND CARUTHERS[30] found no evidence of intramolecular hydrogen bond formation in *o*-hydroxybenzonitrile and attributed this to a similar reason. They state that the distance between the nitrogen atom of the nitrile group and the oxygen atom in this compound is more than 3 Å, too great for normal hydrogen bonding.) The absence of an *ortho*-effect in the *o*-halophenols (Table VI) may also be ascribed to the absence of hydrogen bonding between the two *ortho*-groups. The clear separation of *o*- and *p*-cresol in System D shows that an *ortho*-effect, if it is present, can certainly be observed in this system. Hydrogen bonding is known to be possible in *o*-cresol (VIII) but probably does not occur in *o*-chlorophenol (*cf.* BAKER[31]). This result confirms our earlier opinion[2] that the *ortho*-effect in chromatography is unlikely to be purely spatial in character.

The hypothesis also accounts for the fact that although *m*- and *p*-nitrophenol and their corresponding benzyl ethers show a large separation (Table I), *m*- and *p*-nitrophenyl esters have identical R_M values. This can be ascribed to the fact that the carbonyl group of the acetoxy group in (IX) competes for the lone pair of electrons on the oxygen atom and converts the + M effect of OH and OR into a — M effect[17].

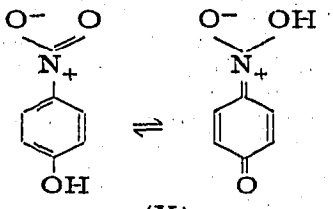


(IX)

It is also possible to account for the fact that *sym*-trisubstituted benzenes obey the group additivity principle even when the corresponding disubstituted derivatives do not. The best example is 3,5-dinitrophenol, which obeys MARTIN's equation with respect to all three groups exactly although *m*-nitrophenol shows an exceptionally large deviation (Table II). In the former molecule, the negative charge induced on the *para*-C atom by the OH group is likely to be neutralized by the unusually strong electron-withdrawing powers of the two *m*-substituted NO_2 groups, which deactivate all positions of the ring. Similar arguments apply to 3,5-dinitroaniline and 3,5-dichloroaniline.

The *p*-substituted anilines obey the group additivity principle less strictly than the other compounds investigated. It is not clear why this is so, but a possible reason is that some compound formation may take place between anilines and hydroxylic

solvents. The presence of even small amounts of compounds of the type, $RNH_3^+OH^-$, would, of course, affect the experimental R_M values considerably. Nitroanilines contain a potentially acidic group and may exist partly as solvated zwitterions, which would behave anomalously. Apart from amino compounds, the only major exception



(X)

to the additivity rule among *p*-substituted benzenes is *p*-nitrophenol. It is difficult to see why this should be so unless, like *p*-nitrosophenol, it too can exist as a tautomeric mixture (X). There is some evidence for this[32].

SUMMARY

The separation of *m*- and *p*-disubstituted benzene derivatives has been studied in four systems. It is shown that the R_M values of these compounds cannot be correlated with the existence of charge separations or with the vectorial dipole moment. The *p*-isomers generally obey the group additivity principle, whereas *m*-isomers, when they differ, depart from this principle. The separation of *m*- and *p*-isomers is explained in terms of a competition between intra- and intermolecular hydrogen bonding. It is shown how in *m*-isomers, the presence of an *ortho/para*-directing group can affect hydrogen bonding by inducing a fractional negative charge on the *para*-carbon atom. Benzene derivatives containing two *meta*-directing groups appear to be inseparable by partition chromatography, and this is discussed. Certain compounds, such as substituted anilines, have anomalous R_M values and may contain, in hydroxylic solvents, small amounts of compounds of the type, $RNH_3^+OH^-$. The chromatography of halophenols shows no influence of the inductive effect of the halogens; they separate according to their molar volume. There was no *ortho*-effect in halophenols in the reversed phase system studied, confirming that this effect is not spatial but mainly polar in origin.

REFERENCES

1 J. GREEN AND S. MARCINKIEWICZ, *J. Chromatog.*, 10 (1963) 35.
2 S. MARCINKIEWICZ, J. GREEN AND D. MCHALE, *J. Chromatog.*, 10 (1963) 42.
3 J. GREEN, S. MARCINKIEWICZ AND D. MCHALE, *J. Chromatog.*, 10 (1963) 158.
4 S. MARCINKIEWICZ AND J. GREEN, *J. Chromatog.*, 10 (1963) 184.
5 J. GREEN AND S. MARCINKIEWICZ, *J. Chromatog.*, 10 (1963) 354.
6 S. MARCINKIEWICZ AND J. GREEN, *J. Chromatog.*, 10 (1963) 366.
7 M. PERPAR, M. TIŠLER AND Ž. VRBASKI, *Mikrochim. Acta*, (1959) 64.
8 C. I. TURI, *Rend. Ist. Super. Sanita*, 21 (1958) 748.
9 V. ETTEL, J. POSPISIL AND Z. DEYL, *Collection Czech. Chem. Commun.*, 24 (1959) 234.
10 B. EKMANN, *Acta Chem. Scand.*, 2 (1948) 383.
11 E. C. BATE-SMITH AND R. G. WESTALL, *Biochim. Biophys. Acta*, 4 (1950) 427.
12 G. WAGNER, *Arch. Pharm.*, 286 (1953) 269.
13 J. GASPARIC, *Motorová Paliva*, 1 (1951) 1.
14 T. GESSNER AND J. N. SMITH, *Biochem. J.*, 75 (1960) 172.
15 L. S. BARK AND J. T. GRAHAM, *Analyst*, 85 (1960) 907.

[16] A. J. P. MARTIN, *Biochem. Soc. Symp. (Cambridge, Engl.)*, 3 (1949) 4.
[17] C. K. INGOLD, *Structure and Mechanism in Organic Chemistry*, Bell, London, 1953.
[18] J. FRANC AND J. JOKL, *Collection Czech. Chem. Commun.*, 21 (1956) 161.
[19] L. E. SUTTON, *Determination of Organic Structures by Physical Methods*, Academic Press, New York, 1955, p. 373.
[20] O. HASSEL AND E. NAESHAGEN, *Z. Physik. Chem.*, B6 (1930) 441.
[21] J. J. LANDER AND W. J. SVIRBELY, *J. Am. Chem. Soc.*, 67 (1945) 322.
[22] S. DIKSTEIN, *J. Chromatog.*, 2 (1959) 204.
[23] J. A. A. KETELAAR, *Chemical Constitution*, Elsevier, Amsterdam, 1953.
[24] K. HIGASI, *Bull. Inst. Phys. Chem. Res. (Tokyo)*, 20 (1941) 218.
[25] H. O. JENKINS, *J. Chem. Soc.*, (1936) 862.
[26] R. J. B. MARSDEN AND L. E. SUTTON, *J. Chem. Soc.*, (1936) 599.
[27] N. J. LEONARD AND L. E. SUTTON, *J. Am. Chem. Soc.*, 70 (1948) 1564.
[28] G. W. WHELAND, *Resonance in Organic Chemistry*, Wiley & Sons, New York, 1955.
[29] H. H. JAFFÉ, *Chem. Rev.*, 53 (1953) 191.
[30] W. BAKER AND G. N. CARUTHERS, *J. Chem. Soc.*, (1937) 479.
[31] W. BAKER, *J. Chem. Soc.*, (1934) 1684.
[32] N. V. SIDGWICK, *Organic Chemistry of Nitrogen*, Clarendon, Oxford, 1949.