

## Note

### Adsorption of monosubstituted phenols on Sephadex® G-15

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(First received February 24th, 1978; revised manuscript received April 17th, 1978)

Sephadex® dextran gels exhibit various kinds of selectivities of which a high affinity for certain aromatic compounds was first pointed out by Gelotte<sup>1</sup>. Determann and Walter<sup>2</sup> suggested that the ether groupings on the crosslinks between the dextran chains are responsible for the adsorption of aromatic solutes, and Brook and Housley<sup>3</sup>, and later Brook and Munday<sup>4</sup>, suggested that the affinity of the gel for phenols is due to an interaction between the phenolic hydroxyl hydrogen and the crosslink. They reported that the adsorption by Sephadex dextran gels of a series of mono-substituted phenols could be correlated by the Hammett equation except in the case of halogen-substituted phenols which are more strongly adsorbed than would be predicted from this equation. Brook and Munday interpreted this as indicative of a different mechanism for adsorption of halophenols.

This paper re-examines this question and it is proposed that all the investigated monosubstituted phenols including the halophenols are adsorbed onto the gel by the same mechanism.

#### *The Hammett equation*

For 3- and 4-substituted benzene derivatives a plot of the logarithm of the equilibrium constant  $K$  for one reaction against  $\log K$  for a second reaction is usually linear. Hammett<sup>5</sup> formalized this linear free-energy relation (LFER) as in eqn. 1

$$\log \frac{K}{K_0} = \rho \cdot \log \frac{K'}{K'_0} \quad (1)$$

where  $K_0$  refers to the unsubstituted benzene derivative and  $\rho$  is a proportionality constant. The ionization of substituted benzoic acids ( $K'$ ) was chosen as a standard and  $\log (K'/K'_0)$  was defined as the substituent constant  $\sigma$ . Hence eqn. 1 becomes

$$\log \frac{K}{K_0} = \rho \cdot \sigma \quad (2)$$

where  $\rho$  is a constant, specific for the reaction involved.

For the adsorption of monosubstituted phenols by Sephadex gels, eqn. 2 can be written as

$$\log \frac{K_d(Y-ArOH)}{K_d(ArOH)} = \rho \cdot \sigma_Y \quad (3)$$

However, since Y itself contributes to the adsorption onto the gel, a correction for this adsorption should be made, and hence eqn. 3 is modified to

$$\log \frac{K_d(\text{Y-ArOH})}{K_d(\text{ArOH}) \cdot K_d(\text{Y})} = \rho \cdot \sigma_Y \quad (4)$$

where  $K_d(\text{Y})$  denotes the contribution from the substituent group Y and is calculated as the ratio of  $K_d$  values for Ar-Y and benzene.

#### MATERIALS AND METHODS

Sephadex® G-15 (Batch No. 2014) and Dextran® 500 ( $\bar{M}_w = 450,000$ ) were obtained from Pharmacia, Uppsala, Sweden. All of the chemicals were of sufficient purity to give single quasi-symmetrical elution peaks. The solutions were made up in water purified in a Milli-Q 4 Housing System (Millipore, Bedford, Mass., U.S.A.).

The solutes were eluted through columns ( $45 \text{ cm} \times 0.8 \text{ cm}^2$ ) with a linear flow-rate of *ca.* 3 cm/h at  $35 \pm 0.2^\circ$ . The column was pre-washed and eluted with Millipore water. The test solutes were loaded on to the column in a volume of 0.3 ml (0.8% of column volume) and their concentration never exceeded 1 g/l. The eluent concentrations were recorded continuously by a differential refractometer (Model R403, Waters Assoc., Milford, Mass., U.S.A.) and samples of the eluate were collected in tubes at 2-h intervals in a closed box containing air saturated with water vapour.

The dynamic distribution coefficient ( $K_d$ ) is defined<sup>5</sup> as

$$K_d = \frac{V_e - V_o}{V_i}$$

where  $V_e$  and  $V_o$  are the peak elution volumes of the test solute and a void-volume indicator (Dextran 500) respectively.  $V_i$ , the internal water volume of the gel column, was calculated from the peak elution volume of  $^2\text{H}_2\text{O}$ , which however gives an over-estimate due to isotope exchange<sup>7</sup> with labile hydrogen atoms (hydroxyl) on the gel. The exchange reaction should thus retard the movement of deuterium of the deuterated water in the column to about the same extent as tritium of tritiated water ( $\text{H}^3\text{HO}$ )<sup>8</sup>. The true internal water volume can be obtained as the peak elution volume of  $\text{H}_2 \text{ } ^{18}\text{O}$  (ref. 9) and the ratio

$$\varphi = \frac{V_{\text{H}^3\text{HO}} - V_o}{V_{\text{H}_2 \text{ } ^{18}\text{O}} - V_o}$$

can be determined. For G-15 the ratio is 1.075 (ref. 10) and hence

$$K_d = \varphi \cdot \frac{V_e - V_o}{V_{\text{H}_2 \text{ } ^{18}\text{O}} - V_o} = 1.075 \cdot \frac{V_e - V_o}{V_{\text{H}_2 \text{ } ^{18}\text{O}} - V_o}$$

#### RESULTS AND DISCUSSION

Log  $K_d$  values for benzene and nine monosubstituted benzene derivatives as

TABLE I

## DISTRIBUTION COEFFICIENTS AND GROUP CONTRIBUTIONS FOR BENZENE AND MONOSUBSTITUTED BENZENE DERIVATIVES

No.	Compound	$\log K_d$	Group (Y)	Group contribution Y (Y)
1	Benzene	0.4837	$\emptyset$	0.4837
2	Toluene	0.5998	CH <sub>3</sub>	0.1160
3	Phenol	0.6061	OH	0.1224
4	Anisole	0.5798	OCH <sub>3</sub>	0.0960
5	Benzaldehyde	0.4702	CHO	-0.0136
6	Acetophenone	0.4953	COCH <sub>3</sub>	0.0116
7	Fluorobenzene	0.5311	F	0.0473
8	Chlorobenzene	0.7737	Cl	0.2900
9	Bromobenzene	0.8901	Br	0.4063
10	Iodobenzene	1.0637	I	0.5800

well as contributions from the substituent group Y are reported in Table I. The contribution of Y is calculated as

$$Y(Y) = \log K_d(\text{ArY}) - \log K_d(\text{benzene}) = \log K_d(Y)$$

Table II gives the  $\log K_d$  values of 18 monosubstituted phenols. Log  $K_d$  values were

TABLE II

DISTRIBUTION COEFFICIENTS AND  $\sigma_Y$  VALUES OF MONOSUBSTITUTED PHENOLS  
 $\sigma$  values were taken from ref. 11, if not stated otherwise.

No.	Phenol	$\log K_d$		$\Delta$ (exptl. - calc.)	$\sigma_Y$
		Exptl.	Calc.		
21	3-CH <sub>3</sub>	0.7179	0.7221	-0.0042	-0.07
22	4-CH <sub>3</sub>	0.7087	0.7221	-0.0134	-0.17
24	3-OH	0.7624	0.7285	0.0339	0.12*
25	4-OH	0.6753	0.7285	-0.0532	-0.36
27	3-OCH <sub>3</sub>	0.7398	0.7021	0.0377	0.12
28	4-OCH <sub>3</sub>	0.6647	0.7021	-0.0374	-0.27
30	3-CHO	0.6676	0.5925	0.0751	0.36
31	4-CHO	0.6779	0.5925	0.0854	0.43**
33	3-COCH <sub>3</sub>	0.6864	0.6177	0.0687	0.31
34	4-COCH <sub>3</sub>	0.7006	0.6177	0.0829	0.44***
36	3-F	0.7197	0.6534	0.0663	0.34
37	4-F	0.6756	0.6534	0.0222	0.06
39	3-Cl	0.9812	0.8961	0.0851	0.37
40	4-Cl	0.9553	0.8961	0.0592	0.23
42	3-Br	1.1013	1.0124	0.0889	0.39
43	4-Br	1.0761	1.0124	0.0637	0.23
45	3-I	1.2703	1.1861	0.0842	0.35
46	4-I	1.2486	1.1861	0.0625	0.28

\* D. H. McDaniel and H. C. Brown, *J. Org. Chem.*, 23 (1958) 420.\*\* A. A. Humffray, J. J. Ryan, J. P. Warren and Y. H. Yung, *Chem. Commun.*, (1965) 610.\*\*\* O. Exner, *Collect. Czech. Chem. Commun.*, 31 (1966) 65.

also calculated as the sum of the contributions from the benzene ring ( $\emptyset$ ) and the two substituents.

$$\log K_d(\text{calc.}) \left\{ \begin{array}{c} \text{OH} \\ | \\ \emptyset - \text{Y} \end{array} \right\} = Y(\emptyset) + Y(\text{OH}) + Y(\text{Y})$$

The difference between the experimentally found  $\log K_d$  value and the calculated value is denoted  $\Delta(\text{exptl.} - \text{calc.})$ .

$$\Delta(\text{exptl.} - \text{calc.}) = \log K_d(\text{Y-ArOH}) - [Y(\emptyset) + Y(\text{OH}) + Y(\text{Y})]$$

$$\Delta(\text{exptl.} - \text{calc.}) = \log \frac{K_d(\text{Y-ArOH})}{K_d(\emptyset) \cdot K_d(\text{OH}) \cdot K_d(\text{Y})} = \log \frac{K_d(\text{Y-ArOH})}{K_d(\text{ArOH}) \cdot K_d(\text{Y})}$$

Eqn. 4 can now be written as

$$\log \frac{K_d(\text{Y-ArOH})}{K_d(\text{ArOH}) \cdot K_d(\text{Y})} = \Delta(\text{exptl.} - \text{calc.}) = \rho \cdot \sigma_Y \quad (5)$$

Fig. 1 shows  $\log K_d$  ratios and substituent  $\sigma$  values in a plot of eqn. 3. The straight line was calculated by linear regression and the standard deviation  $s$  and the correlation coefficient  $r$  were calculated as outlined by Jaffé<sup>11</sup>. The linear regression equation is

$$\log \frac{K_d(\text{Y-ArOH})}{K_d(\text{ArOH})} = 0.3308\sigma_Y + 0.1708,$$

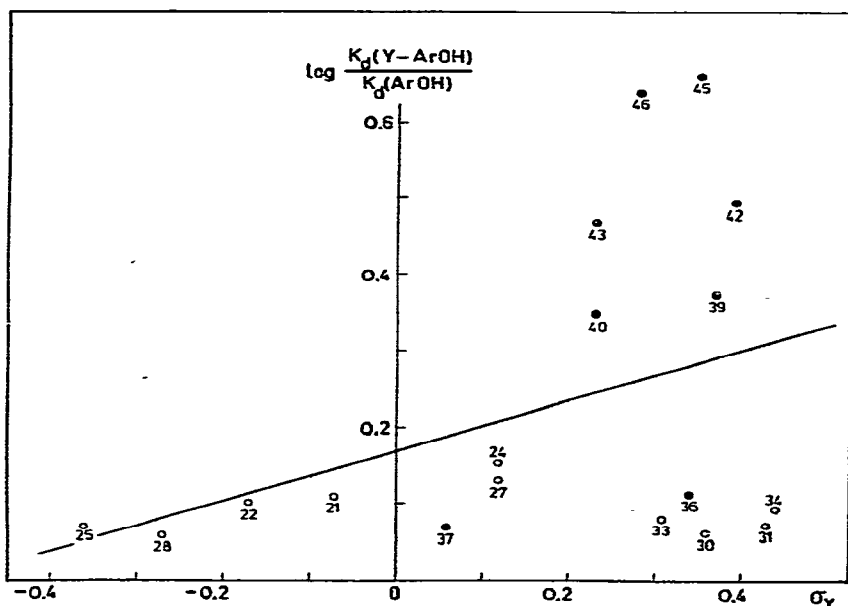


Fig. 1. Plot of data for monosubstituted phenols according to eqn. 3. The numbers refer to the listing in Table II. For simplicity the halophenols have been denoted by filled circles.

with  $n = 18$ ,  $s = 0.200$  and  $r = 0.386$ . With this plot, which is that used by Brook and Munday<sup>3</sup>, there is no indication of any correlation. If, however, as Brook and Munday pointed out, the data for the halogen derivatives are excluded, there is a fair degree of linearity among the remaining solutes. However, this is almost certainly due to the similar magnitudes of their Y-group contributions.

In Fig. 2 the  $\log K_d$  values are correlated with substituent  $\sigma$  values as in eqn. 5, *i.e.* correction is made for the contribution of the isolated Y group. The linear regression equation thus becomes

$$\Delta(\text{exptl.} - \text{calc.}) = 0.1801 \cdot \sigma_Y + 0.0132$$

where  $n = 18$ ,  $s = 0.005$  and  $r = 0.993$ . The correlation is now good, and this suggests the same mechanism of adsorption to the gel for all the investigated mono-substituted phenols, although no definite conclusion about the mechanism itself can be drawn as yet.

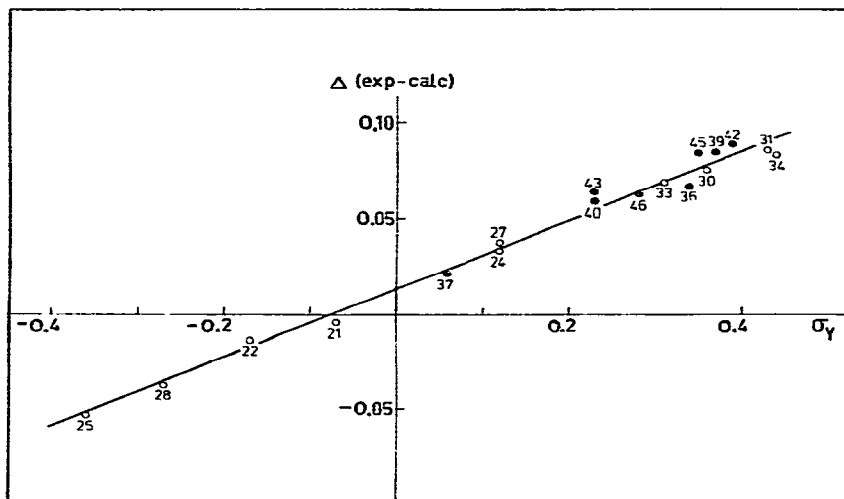


Fig. 2. Plot of the same data as in Fig. 1 but on the basis of eqn. 5. The notation is as in Fig. 1.

#### ACKNOWLEDGEMENTS

I wish to thank Dr. N. V. B. Marsden for invaluable discussions. This work was supported by the Swedish Natural Science Research Council (Grant K 2944) and Uppsala University.

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