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## Letter to the Editor

### Comment on the proposed $R_Q$ transformation method for optimizing mobile phase composition in high-performance liquid chromatography

Sir,

Toon and Rowland have suggested recently<sup>1,2</sup> a simple graphical method for the rapid selection of an optimal mobile phase composition for the high-performance liquid chromatographic (HPLC) analysis of a multi-component mixture. If correct, their treatment has exciting implications not only in the design of, for example, optimal gradient elution systems, but also in the developing use of HPLC for providing solute physico-chemical data. However, a number of points arise from their treatment that require comment.

(1) Taking the suggested<sup>1,2</sup> transformation term  $R_Q$  as

$$R_Q = \log(1 - R_O/R_T) \quad (1)$$

where  $R_O$  and  $R_T$  are non-retained and retained solutes respectively, it follows that, as

$$R_Q = \log\left(\frac{R_T - R_O}{R_O} \cdot \frac{R_O}{R_T}\right) \quad (2)$$

then

$$R_Q = \log\left(\frac{k}{k+1}\right) \quad (3)$$

where  $k$  is the capacity factor, equal to  $(R_T/R_O) - 1$ . These deductions show that  $R_Q$  is always negative, which is at variance with Figs. 1, 3, 5 and 8 in ref. 1.

(2) Fig. 8 in ref. 1 is based on the data of Crechiolo and Hill<sup>3</sup> for thiamphenicol determined in an octadecylsilane HPLC system using aqueous methanol eluents, and purports to show how a found curvilinear relationship between  $\log k$  and the percentage of organic modifier in the eluent can be linearized using the  $R_Q$  transformation. Assuming eqn. 1 to be correct, then Toon and Rowland<sup>1,2</sup> are suggesting a plot of  $\log[\log(k/1+k)]$  versus percentage composition as the normalization step. Least-squares regression of the data of Crechiolo and Hill<sup>3</sup> (Table I), according to  $\log(k/1+k)$  versus percentage composition, gives a correlation coefficient ( $r$ ) of 0.994 whereas using  $\log[\log(k/1+k)]$ ,  $r = 0.955$  [the sign of  $\log(k/1+k)$  has been reversed to accomplish regression].

(3) It therefore appears that the authors intend  $R_Q$  to be equal to  $k/1+k$ , which we shall call  $R_Q^*$ .

(4)  $R_Q^*$  cannot be 1.0 or greater, which means that the ordinate intercepts of  $R_Q^*$

TABLE I

HPLC RETENTION DATA FOR THIAMPHENICOL AND CHLORAMPHENICOL DETERMINED IN AN ODS/METHANOL-WATER SYSTEM (ESTIMATED FROM THE PAPER BY CRECHIOLO AND HILL<sup>3</sup>)

Methanol in mobile phase (%)	$k$	
	Thiamphenicol	Chloramphenicol
10	17	
15	12	
20	6.2	19
25	4.5	13
30	3.2	10
35	2.7	7.2
40	2.2	5.5
45	2.1	4.5
50	1.8	3.5

versus percentage mobile phase composition also should not be equal to or greater than 1.0.

(5) Let us now attempt to use  $R_Q^*$  for linearization of retention data with respect to mobile phase composition. To do so we have simply returned to the paper of Crechiolo and Hill<sup>3</sup> using, this time, data for chloroamphenicol that were published in the same figure as for thiamphenicol. Fig. 1 shows that the  $R_Q^*$  transformation does not linearize the retention data, with regressions of  $\log R_Q^*$  versus percentage composition giving  $r = 0.991$ , whereas simply plotting  $\log k$  versus percentage composition gives  $r = 0.996$ .

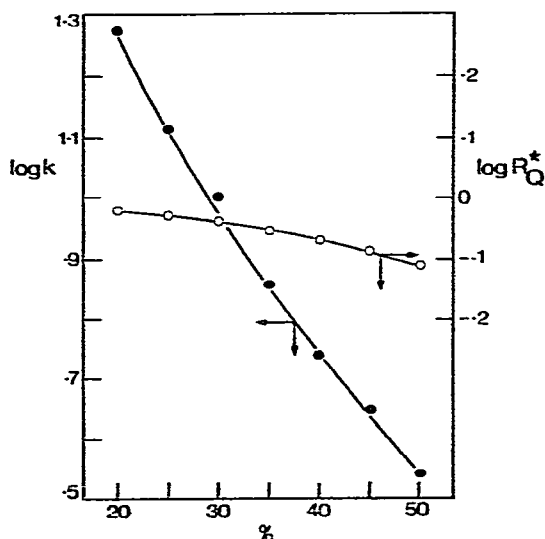


Fig. 1. Plots of  $\log k$  and  $\log R_Q^*$  for chloramphenicol against the percentage by volume of methanol in the eluent using the data of Crechiolo and Hill<sup>3</sup> (Table I)...

TABLE II

GROUP EXTRA-THERMODYNAMIC TERMS (ASSUMING  $k_i = 10.0$ ) OBTAINED FROM CAPACITY FACTOR DATA USING EQNS. 4 AND 5

$k_j$	$\tau$ (eqn. 4)	$\tau_Q^*$ (eqn. 5)
10.1	0.004	0.0004
10.2	0.008	0.0008
10.5	0.021	0.0019
11.0	0.04	0.0036
15.0	0.17	0.013
20.0	0.30	0.020
100	1.00	0.037
500	1.70	0.041
1000	2.0	0.041

(6) Structural effects in liquid chromatography are well described using simple extra-thermodynamic terms such as<sup>4</sup>

$$\tau = \log k_j - \log k_i \quad (4)$$

where  $j$  and  $i$  are solutes differing by a structurally discrete function. Toon and Rowland have claimed<sup>2</sup> that  $R_Q^*$  values obtained by extrapolation to zero percentage of organic modifier are useful in describing drug physico-chemical characters. One would assume that if this were so, a group extra-thermodynamic term could be obtained from  $R_Q^*$ , *i.e.*,

$$\tau_Q^* = \log (R_Q^*/R_{Q_i}^*) \quad (5)$$

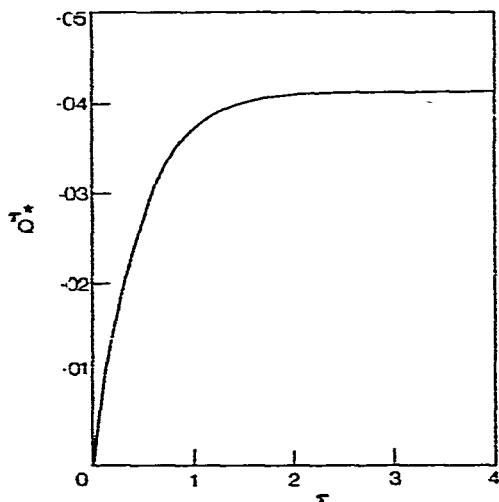


Fig. 2. Relationship between extra-thermodynamic group terms  $\tau$  and  $\tau_Q^*$ , where  $\tau$  is obtained from  $\log(k_j/k_i)$  and  $\tau_Q^*$  from  $\log(R_Q^*/R_{Q_i}^*)$  using a  $k_i$  value of 10 and  $k_j$  values from 10.1 to 1000 (Table II).

Table II and Fig. 2 shows that  $\tau_Q^*$  is quasi-linear with respect to  $\tau$  only between values of 0 and approximately 0.25. This arises because with  $k_f$  values between 100 and infinity the range in  $\tau_Q^*$  is only 0.037 to approximately 0.042 (Fig. 2).

In conclusion, neither  $R_Q^*$  nor  $R_Q$  can be described as being appropriate for the linearization of HPLC retention data with respect to mobile phase composition, nor can extrapolated intercept values be of any general use for describing structural characteristics. Other transformations based on log-log plots<sup>5</sup>, log  $k$  versus organic modifier volume fraction<sup>6</sup> or log  $k$  versus organic modifier thermodynamic activity<sup>7</sup> seem more useful.

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