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Commentary

Regarding the sources of data analyzed with quantitative structure–skin permeability relationship methods (commentary on 'Investigation of the mechanism of flux across human skin in vitro by quantitative structure–permeability relationships')

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Abstract

We investigated the sources of data used in recently published predictive models of skin permeability. It was found that skin permeability coefficients for 63 compounds are poorly documented. We hypothesized that these coefficients were calculated using the simple two variable, three parameter 'Potts and Guy' regression equation and hence were not derived from experimental measurements. We therefore examined the distribution of residuals of these reported coefficients compared with the Potts and Guy predictions. The residuals cannot be described by a normal distribution. A substantial (51%) number of residuals equaled 0.00. Further analysis demonstrated that 89% (56 out of 63) of the skin permeability coefficients can be explained as being calculated by the Potts and Guy equation using different documented octanol–water partition coefficients, and/or transcription errors. The results strongly suggest that these 63 skin permeability coefficients are calculated and not experimentally determined—a conclusion subsequently confirmed by one of the developers of the data set. Continued use of these data would lead to biased model selection, underestimation of experimental variability, and overestimation of model predictive ability.

Keywords: Data analysis; Quantitative structure–skin permeability relationship; Skin flux mechanism; Human skin; Skin absorption

1. Introduction

Chemicals in contact with skin permeate it with varying rapidity and efficiency. If the chemical is toxic, there is potential danger of local or systemic adverse effects. Because the enormous number of potentially hazardous chemicals prohibits experimental determination for all but a small percentage, there is a great need for an accurate means of predicting the transdermal penetration of chemicals.

The quantitative structure–permeability relationships (QSPR) method typically applies multi-variable stepwise regression of skin permeability data to any number of physical/chemical descriptors and holds the possibility for providing a means to predict skin permeability—an important component in the prediction of dermal penetration.

A previous commentary (Poda et al., 2001) noted anomalies in the statistical distribution of major molecular descriptors in a recently introduced (Kirchner et al., 1997) set of skin permeability coefficients, and questioned the appropriateness of using these data in the types of QSPRs exemplified by Cronin et al.'s (1999) approach. That analysis (Poda et al., 2001) led us to investigate the sources of the data set introduced by Kirchner et al. (1997) and subsequently used by Cronin et al. (1999). We hypothesized that these data were *calculated* using a simple regression equation and are not *experimental* data. We therefore examined the distribution of residuals of these reported data from the simple Potts and Guy (1992) equation. For comparison, we also examined the distribution of residuals of another data set (Flynn, 1990) from the Potts and Guy prediction.

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2. Background

A recent paper by Cronin et al. (1999) claims to have achieved excellent correlation (r^2=0.86) between the authors' QSPR model and skin permeability coefficients from 107 compounds (seven of 114 original coefficients were found to be outliers and were removed). The authors first analyzed 47 molecular descriptors of physical/chemical properties of the compounds, which yielded a QSPR with a correlation (r^2) of 0.70 for all 114 compounds. Following this analysis, Cronin et al. chose a simple first-order model with two independent variables and three parameters, first proposed by Potts and Guy (1992):

$$\log K_p = a \log P + b\,\mathrm{MW} + c \quad (1)$$

where K_p is the permeability coefficient, P is octanol–water partition coefficient, MW is molecular weight, and a, b and c are parameters to be fitted. (For K_p values reported herein, the units are (cm/h); logarithms are base 10.)

The original Potts and Guy (1992) regression was based on data published by Flynn (1990).

The Flynn data set is a collection of human skin permeability coefficients from an aqueous vehicle, derived primarily from in vitro measurements and collected from a variety of literature sources. The data are comprised of 97 measurements of 94 chemicals spanning a range of molecular weight (18–765) and log P (−2.25–5.49). Regression of Eq. (1) against these data yielded a correlation (r^2) of 0.67[1].

Cronin et al. (1999) have analyzed a larger data set of 114 compounds. Application of Eq. (1) to these data yielded an r^2 of 0.86 after removal of seven outliers—a noteworthy statistical improvement over the original Potts and Guy model. We address the question: what is the basis for this improvement?

3. Method of analysis

Cronin et al. (1999) report that ''Permeability coefficients (K_p) for the passage of 114 compounds *through human skin in vitro* were taken from Kirchner et al. (1997) . . . '' (italics added). The referenced paper (Kirchner et al., 1997) reports the sources of these data. Of the 114 chemicals, 51 were taken from the Flynn (1990) data base. The remaining 63 ''had *calculated* log K_p values that were obtained from the Occupational Safety and Health Association'' (sic) (OSHA) (Kirchner et al., 1997; italics added). The authors do not specify whether these were calculated from experimental data or by some other means. An examination of the source of the 'OSHA data' cited by both Kirchner et al. and Cronin et al.—''Wittaker et al. (1993)'' (sic)—reveals this to be a one paragraph abstract (Wittaker et al., 1993) with no mention of specific skin permeability coefficients.

Thus, 63 log K_p values from Kirchner et al.'s data set of 114, also used by Cronin et al., are poorly documented. Some of the co-authors of the Wittaker abstract were affiliated with the Syracuse Research Corporation (SRC). The SRC has developed a dermal absorption estimation program (DermWin, http://esc.syrres.com/interkow/Dermal.htm), which the web site claims is based on a method developed by the US Environmental Protection Agency (US EPA, 1992). The EPA method for calculation of log K_P is a trivial modification of the original Potts and Guy equation (−2.72 for c, Eq. (1), instead of −2.74). We therefore applied the Potts and Guy equation, using the EPA's value of c, to the 63 compounds in question and examined the residuals. For comparison, we performed identical analyses on the Flynn (1990) data base of 97 permeability measurements—that is, we examined the residual differences between the Flynn data base and the Potts and Guy predictions.

4. Results and discussion

Fig. 1a shows a comparison between the log K_p values reported in Kirchner et al. (1997) for the 63 compounds ('OSHA data') and the predicted log K_p values using Eq. (1), with log P values taken from Kirchner et al. (1997; log K_{ow} from their Table 1) and MWs listed by Cronin et al. (1999; M_r from their Table 1). Kirchner et al. do not list molecular weights). A striking feature of Fig. 1a is an apparent lack of departure of a large proportion of these reported K_p values from the regression.

To quantify the nature of the log K_p values for the 63 compounds added by Kirchner et al. (1997) and used by Cronin et al. (1999), we examined the distribution of residuals from the Potts and Guy regression model. The most obvious characteristic of these values is the large proportion (51%) of data with a residual of exactly 0.00 from the regression line (Fig. 1b). If the log K_p values in this data set are in fact measured, and not calculated by a regression line, we can reasonably assume that the residual values should follow a continuous distribution around some mean value near zero. As the observed residuals appear closer to a normal distribution than any other continuous density curve known to us, subsequent calculations assume normality.

First, we attempted to specify the normal distribution that best describes the large proportion of residuals at or near zero. A normal density with a mean of 0.00 and standard deviation 0.014 correctly characterizes the large proportion (62%) of values with residuals within the range [−0.04, 0.04] from the observed data (Fig. 1b). Under this particular normal density, however, the overall distribution

[1]Potts and Guy (1992) reported a value of −6.3 for c, using units (cm/s) for K_p. For units (cm/h), the value is −2.74. Potts and Guy report 0.71 for a and −0.0061 for b.

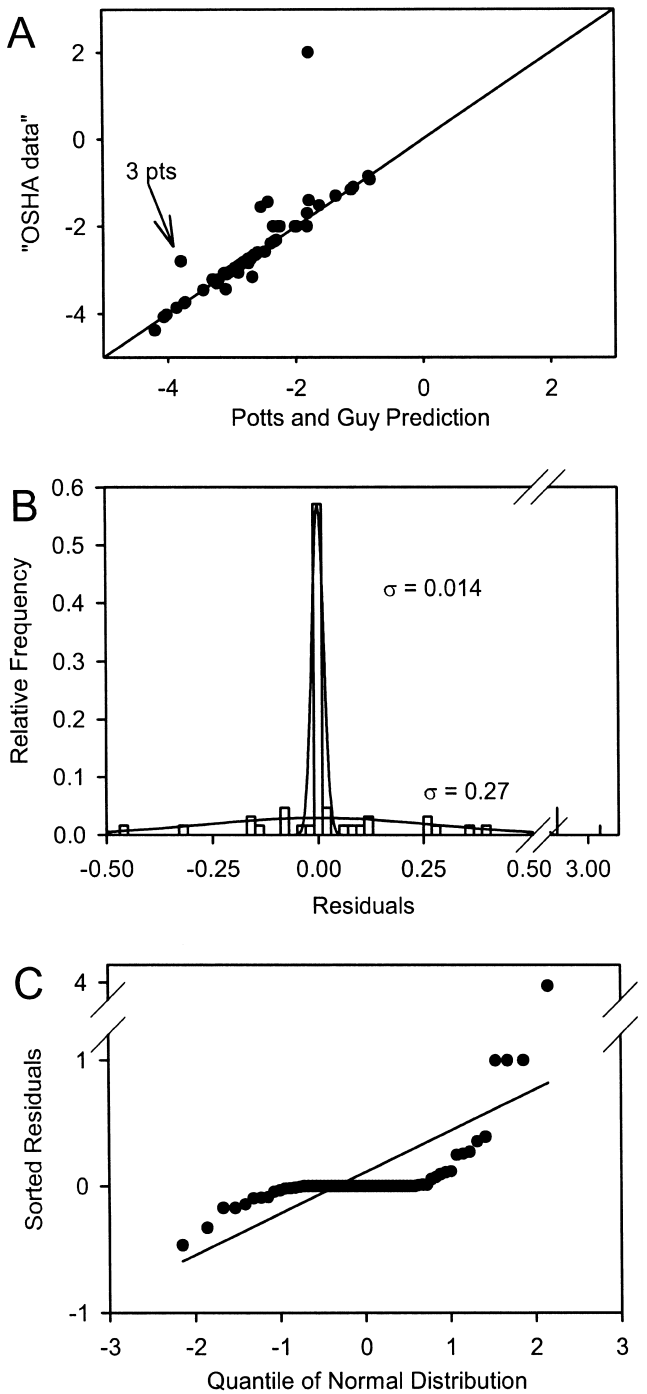

Fig. 1. Distribution of residuals of 'OSHA data' and Potts and Guy prediction. (a) The log of the permeability coefficients ($\log K_p$) of 63 compounds reported by Kirchner et al. (1997) ('OSHA data') compared with $\log K_p$ values calculated with the Potts and Guy (1992) equation. K_p units are cm/h. Line of identity is also shown. (b) Distribution of residuals. Relative frequency of residuals is shown (bars) along with continuous normal distributions with standard deviations (σ) as indicated. (c) Q–Q plots of residuals. Sorted residuals of 'OSHA data' and Potts and Guy prediction (y-axis) versus quantiles of a standard normal distribution. Solid line is the linear regression.

of observed residuals would be extremely unlikely. For instance, the probability of being outside the range [−0.04, 0.04], would be essentially zero for the given normal density. This strongly contradicts the observed distribution of residuals, where 38% fall outside this range.

Using a different approach, a normal density with mean 0.00 and standard deviation 0.27 correctly predicts that 94 percent of the observed residuals will fall within 0.5 of the mean (i.e., between [−0.5, 0.5]), but drastically underestimates the proportion of observed residuals exactly equal to 0.00 within some roundoff error (51% observed versus 6% expected).

The lack of correspondence between the residuals and any normal distribution is verified through a Q–Q plot (Fig. 1c). A quantity that follows any normal distribution would result in a plot that approximates a straight line—i.e., the quantiles of the two distributions agree. Significant nonlinearity in the tails of the Q–Q plot demonstrates the deviation from normality.

Potential explanations for these results can be summarized as either: (1) the residuals follow some non-normal continuous density which better predicts the observed spread and the large proportion of fitted values being identical to the observed $\log K_p$; or (2) the large proportion of residual values near zero reflects some unusual characteristic of these data where the regression line perfectly explains a subset of chemical compounds with virtually no error, and the distribution of residuals is essentially discrete. We find the first explanation highly unlikely as the deviation from normality (as seen in Fig. 1c) is almost entirely due to the spike of near-zero residuals. The second explanation also seems highly implausible if the $\log K_p$ values are measured rather than calculated.

For comparison, we performed a similar analysis on the original Flynn (1990) data set of 97 reported $\log K_p$ values (Fig. 2a). That is, we compared the $\log K_p$ values reported by Flynn (1990) with predicted values from the Potts and Guy (1992) equation, using octanol–water partition coefficients and molecular weights listed by Flynn (1990).

In contrast to the 'OSHA data', the observed residuals from the Flynn data appear to follow a normal distribution (Fig. 2b). Using a normal distribution with a mean of −0.02 and S.D. of 0.73 which we calculated from the residuals, we would expect that 11% of the data would fall within ±0.1 of the mean. A similar proportion (19%) of the observed residuals fall within that range. Within the range of ±1.0 of the mean, 18% of a randomly chosen data set are expected to fall, while 20% are observed. Thus, while there is a somewhat greater concentration of residuals near zero than expected, overall the distribution of residuals from the Flynn data set appear to be normally distributed, in contrast to the 'OSHA data'. The correspondence between these residuals and a normal distribution is verified by the Q–Q plot (Fig. 2c) which approximately follows a straight line throughout the entire range of residuals.

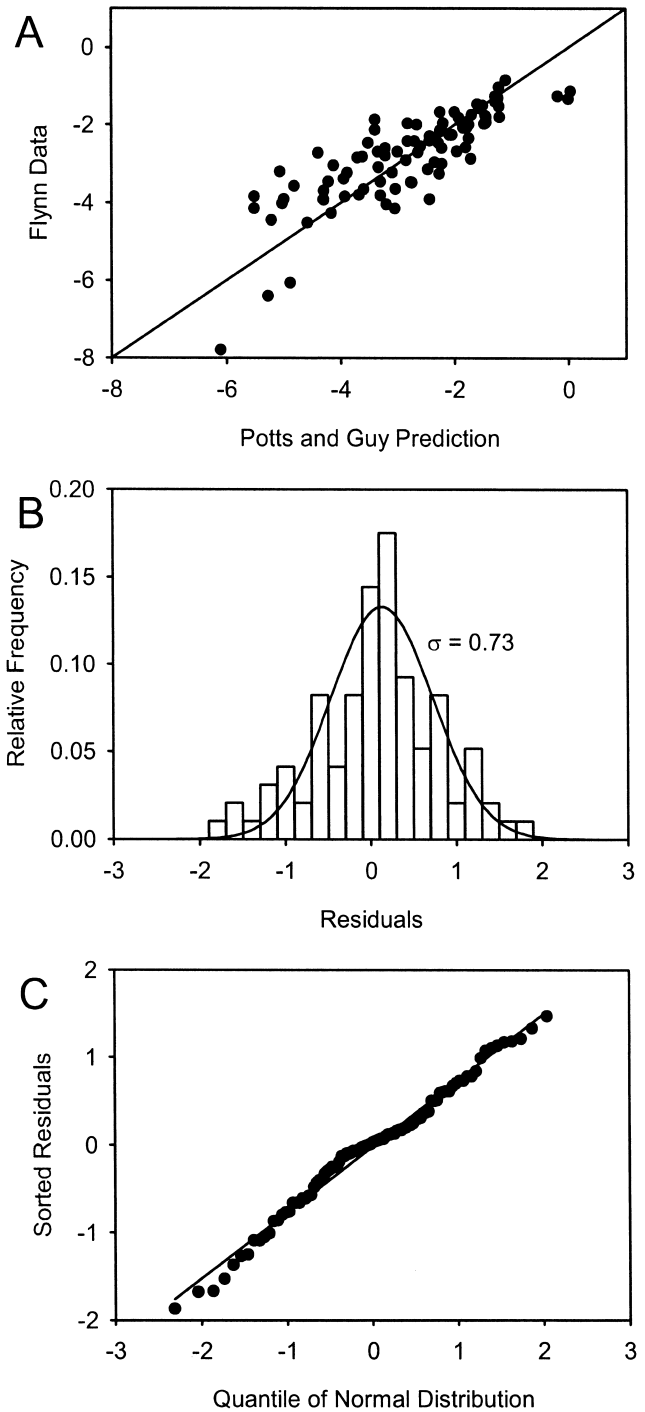

Fig. 2. Distribution of residuals of Flynn data and Potts and Guy prediction. (a) The log of the permeability coefficients ($\log K_p$) of 119 measurements reported by Flynn (1990) compared with $\log K_p$ values calculated with the Potts and Guy (1992) equation. K_p units are cm/h. Line of identity is also shown. (b) Distribution of residuals. Relative frequency of residuals is shown (bars) along with continuous normal distribution with standard deviation (σ) as indicated. (c) Q–Q plots of residuals. Sorted residuals of Flynn data and Potts and Guy prediction (y-axis) versus quantiles of normal distribution. Solid line is the linear regression.

Based on this analysis, a likely explanation for the origin of the 'OSHA data' is that they were calculated from the Potts and Guy equation. If this is the case, then what is the explanation for the significant number of data points that do not lie very close to the regression line?

We examined all compounds (16 in number) with reported $\log K_p$ values having residual magnitudes >0.1 compared with the Potts and Guy predictions. These are listed in Table 1. Table 1 also includes $\log P$ values reported by Kirchner et al. (1997), by Cronin et al. (1999), and $\log P$ values that were calculated using the SRC's web site (http://esc.syrres.com/interkow/kowdemo.htm).

Using these three sources for $\log P$, the predicted $\log K_p$ values from the Potts and Guy equation were calculated and listed in Table 1. Residual differences between reported $\log K_p$ values and calculated $\log K_p$ values for all three cases are also listed. Results may be summarized as follows.

The reported $\log K_p$ value for one compound, propylene dichloride ($+2.00$) differs only in sign from the Potts and Guy prediction (-2.00) using the $\log P$ value reported by Cronin et al. Three reported $\log K_p$ values differ from the Potts and Guy prediction by exactly 1.00. For one compound (diethanolamine) the MW is listed by Cronin et al. (1999) as 77.08; we used the correct value of 105.14. As a result of these calculations—using different reported $\log P$ values and corrected MW in the Potts and Guy equation—five additional compounds had reported $\log K_p$ values that differed from the predicted values by ≤ 0.1 (Table 1). Thus, only seven compounds in the 'OSHA data' set could not be explained as being calculated by the Potts and Guy equation, possibly with transcription errors.

The origin of these data has been confirmed by one of the authors of the Wittaker et al. (1993) abstract. The data were calculated as part of a larger effort to screen and prioritize chemicals for dermal absorption testing, and were never presented as experimental measurements (D.A. Gray, 2002, personal communication).

5. Conclusions

The skin permeability data introduced by Kirchner et al. (1997) and applied by Cronin et al. (1999) should not be used in the development or validation of QSPRs—the resulting QSPR would simply be a model of a model. The apparent statistical improvement reported by Cronin et al. (1999) compared with Potts and Guy's original (1992) formulation is an artifact. Application of Eq. (1) to a data set containing data originally calculated using Eq. (1) would of course be absurd, and any conclusions drawn from such an analysis would be invalid.

Continued utilization of these data would lead to a biased selection of statistical models in favor of those similar in form to Eq. (1), an underestimation of the

Table 1
Compounds with permeability coefficients reported by Kirchner et al. and Cronin et al. (log K_p reported) with residual magnitudes >0.1 (residuals (1)) compared with Potts and Guy prediction (log K_p pred. (1)) using octanol–water partition coefficients from Kirchner et al. (log P (1))

Compound	MW	Log P (1)	Log P (2)	Log P (3)	Log K_p reported	Log K_p pred. (1)	Log K_p pred. (2)	Log K_p pred. (3)	Residuals (1)	Residuals (2)	Residuals (3)
Acetaldehyde	44.05	0.43	−0.22	−0.17	−3.15	−2.68	−3.14	−3.11	−0.47	−0.01	−0.04
Epichlorohydrin	92.53	0.26	0.45	0.63	−3.43	−3.10	−2.96	−2.84	−0.33	−0.47	−0.59
Diethanolamine[a]	77.08	−1.43	−1.31	−1.71	−4.38	−4.21	−4.12	−4.40	−0.17	−0.26	0.02
	(105.14)					(−4.38)	(−4.29)	(−4.57)	(0.00)	(−0.09)	(0.19)
Butyl acrylate	128.17	2.36	2.36	2.20	−2.00	−1.83	−1.83	−1.94	−0.17	−0.17	−0.06
Acrylic acid	72.06	0.36	0.35	0.44	−3.05	−2.90	−2.91	−2.85	−0.15	−0.14	−0.20
Ethylhexyl phthalate	390.56	4.89	7.45	8.39	−1.52	−1.63	0.19	0.85	0.11	−1.71	−2.37
N,N-Dimethyl aniline	121.18	2.31	2.31	2.17	−1.70	−1.82	−1.82	−1.92	0.12	0.12	0.22
Isoamyl alcohol	88.15	1.42	1.16	2.26	−2.00	−2.25	−2.43	−2.36	0.25	0.43	0.36
Phenol	94.11	1.46	1.47	1.51	−2.00	−2.26	−2.25	−2.22	0.26	0.25	0.22
Ethylene dichloride	98.96	1.48	1.47	1.83	−2.00	−2.27	−2.28	−2.02	0.27	0.28	0.02
1,3-Dichloropropane	110.97	1.38	1.76	2.29	−2.00	−2.42	−2.15	−1.77	0.42	0.15	−0.23
Hexachloroethane	236.74	3.34	4.14	4.03	−1.40	−1.79	−1.22	−1.30	0.39	−0.18	−0.10
o-Toluidine	107.16	1.32	1.32	1.62	−1.44	−2.44	−2.44	−2.22	1.00	1.00	0.78
n-Butyl alcohol	74.12	0.88	0.88	0.84	−1.55	−2.55	−2.55	−2.58	1.00	1.00	1.03
Dimethyl acetamide	87.12	−0.77	−0.77	−0.49	−2.80	−3.80	−3.80	−3.60	1.00	1.00	0.80
Propylene dichloride	112.99	2.28	1.99	2.25	2.00	−1.79	−2.00	−1.81	3.79	4.00	3.81

Molecular weights (*MW*) are listed from Cronin et al. Values of log P are given for each compound, as reported by Kirchner et al. (log P (1)), by Cronin et al. (log P (2)), and taken from the Syracuse Research Corporation's web site (log P (3)). Using values of log P from each source, predicted values were calculated using the Potts and Guy model (log K_p pred. (1) through log K_p pred(3)) to give residual differences (residuals (1) through residuals (3)) as compared to the log K_p reported. Units for K_p are (cm/h).

[a] Calculations shown for MW reported by Cronin et al. (77.08) and for correct value (105.14).

variability of statistical results, and an overestimation predictive accuracy.

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