Life Sciences, Vol 21, pp. 569-576 Pergamon Press
Printed In The U.S.A.

DIRECTIONAL LIPOPHILIC CHARACTER IN A SERIES OF PSYCHOTOMIMETIC PHENETHYLAMINE DERIVATIVES

David E. Nichols[1], Alexander T. Shulgin[2], and Donald C. Dyer[3]

Department of Medicinal Chemistry and Pharmacognosy[1]
School of Pharmacy and Pharmacal Sciences
Purdue University
West Lafayette, Indiana 47907

1483 Shulgin Road[2]
Lafayette, California 94549

College of Veterinary Medicine[3]
Iowa State University
Ames, Iowa 50011

(Received in final form July 15, 1977)

Summary

Octanol-water partition coefficients (log P) were determined for a series of substituted psychotomimetic phenethylamine derivatives. A relationship was established between log P, steric bulk in the para-position and the ability to stimulate serotonin (5-HT) receptors in an in vitro sheep umbilical artery preparation. It appears that Log P values and in vitro activity in this preparation may be useful in predicting hallucinogenic potency in man.

It was reported recently that a useful correlation exists between 1-octanol-water partition coefficients and hallucinogenic acitivity in man for a series of phenylisopropylamine derivatives (1). Optimum lipophilicity was calculated to be at log P = 3.14. This compares favorably with the log P for LSD of 2.96 (Wm. J. Dunn, III, personal communication) and may support the idea that hallucinogenic amphetamines share a common mechanism of action with LSD. Calculation of log P for the indole hallucinogens N,N-dimethyltryptamine and N,N-diethyltryptamine gives values of 2.36 and 3.40, respectively, and in the range expected for activity. The calculated values of log P for N,N-dipropyltryptamine and N,N-dibutyltryptamine of 4.44 and 5.48 are considerably above the expected optimum and, in fact, these latter two compounds show greatly reduced human activity.

Nichols and Dyer (2) reported that in a series of mescaline analogs a relationship exists between log P and the ability to stimulate serotonin receptors in vitro. Shulgin and Dyer (3) also demonstrated a relationship between activity and the number of carbon atoms in the alkyl chain in a homologous series of 2,5-dimethoxy-4-n-alkylphenylisopropylamines. From principles of additivity, the number of carbon atoms is directly related to log P.

It was apparent from these and other earlier studies that the sheep umbilical artery model showed good correlation with human psychotomimetic activity; not only with respect to correctly assigning relative potencies to the various classes of hallucinogens (4), but also in showing stereo-

selectivity for the R-(-) enantiomer of hallucinogenic phenylisopropylamines (5), the active isomer in man. In view of the possible importance of lipophilicity in determining both hallucinogenic potency in man and in describing activity at in vitro serotonin receptors it was decided to investigate further this parameter. We especially questioned whether this was a non specific or specific hydrophobic effect.

1-Octanol Water Partition Coefficients

Partition coefficients were determined for all compounds in 1-octanol-water as described previously (1) using 0.1 molar phosphate buffer, pH 8, as the aqueous phase. Log P values for some of the compounds reported previously (1) were calculated. Experimental determinations were made for all these. Several others were redetermined to minimize error. With some exceptions, we observed good overall agreement with the earlier values. The present data are listed in table I.

It will be noted from the table that changes in lipophilicity have been brought about chiefly through modification of the para substituent. As first noted by Shulgin (6), both the 2 and 5 positions are sensitive to steric bulk and groups larger than methoxy lead to inactive compounds. The diversity of para substituents, ranging from alkyl to a nitro group, allowed us to examine the dependence on electronic character of this substituent.

Regression equations were generated using a non-weighted polynomial regression program and multivariable linear regression programs and a Hewlett-Packard 9810 programmable calculator.

Effects on Sheep Umbilical Artery Strips

Data for compounds 6-8 are new and comparisons were made on isolated sheep umbilical arteries as described previously (2). The activity values represent the ED_{50} mescaline/ED_{50} compound and were obtained directly from dose response curves in each set of experiments. Log RBR is the $\log_{10}$ of this ratio. Activity data for the other compounds were obtained from the comparable studies of Nichols and Dyer (2), Shulgin and Dyer (3), and of Dyer et al. (5).

Results and Discussion

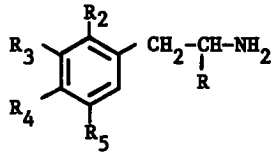
Since it is more common to observe a linear dependence on log P in an in vitro model we first attempted to develop a linear regression equation using log P as the single independent variable. Using the data for compounds 1-17 equation (1) was developed:

$$\log RBR = 0.027 + 0.368 \log P$$
$$n = 17;\ s = 0.44;\ r = 0.66 \qquad (1)$$
$$F_{1,15} = 11.86;\ F_{1,15\ \alpha\ 0.005} = 10.80$$

where n is the number of data points, s is the standard deviation for the regression, and r is the correlation coefficient. Although the equation is significant at the 99.5% level, the standard deviation is high and only 44% ($r^2 = 0.44$) of the variance is explained. The addition of higher order terms did not improve the regression and was not statistically justified, as determined by the F statistic for additional power terms.

TABLE I

Log P Values and Activity in Sheep Umbilical Artery 5-HT Receptors



Cmpd	R_2	R_3	R_4	R_5	R	Log P	Log Activity[a] obs	calc	(Log RBR) |Δ|
1	H	OCH_3	OCH_3	OCH_3	H	0.78	0.00	0.20	0.20
2	H	OCH_3	OC_2H_5	OCH_3	H	1.11	0.31	0.39	0.08
3	H	OCH_3	OC_3H_7	OCH_3	H	1.70	0.56	0.75	0.19
4	H	OCH_3	O-i-C_3H_7	OCH_3	H	1.52	0.45	0.64	0.19
5	H	OCH_3	Br	OCH_3	H	2.03	0.88	0.94	0.06
6	OCH_3	H	SCH_3	OCH_3	H	1.81	0.83	0.81	0.02
7	OCH_3	H	SCH_3	OCH_3	CH_3	2.17	1.31	1.03	0.28
8	OCH_3	H	NO_2	OCH_3	CH_3	1.74	0.67	0.77	0.10
9	OCH_3	H	Br	OCH_3	CH_3	2.54	1.57	1.25	0.32
10	OCH_3	H	CH_3	OCH_3	CH_3	2.24	1.00	1.07	0.07
11	OCH_3	H	C_2H_5	OCH_3	CH_3	2.76	1.59	1.40	0.19
12	OCH_3	H	n-C_3H_7	OCH_3	CH_3	3.37	1.84	1.74	0.10
13	OCH_3	H	n-C_4H_9	OCH_3	CH_3	4.00	1.62	1.58	0.04
14	OCH_3	H	n-C_5H_{11}	OCH_3	CH_3	4.43	0.88	1.29	0.41
15	OCH_3	H	t-C_4H_9	OCH_3	CH_3	3.91	1.39	1.52	0.13
16	H	OCH_3	O-n-C_4H_9	OCH_3	H	2.32	0.10	0.04	0.06
17	H	OCH_3	$OCH_2C_6H_5$	OCH_3	H	2.40	0.48	0.09	0.39
18	OCH_3	H	H	OCH_3	CH_3	1.72	-0.98[b]		
19	H	H	OCH_3	H	CH_3	1.78	----[b]		
20	OCH_3	H	OCH_3	OCH_3	CH_3	1.10[c]			
21	H	OCH_3	OCH_3	OCH_3	CH_3	1.21[c]			
22	H	OCH_3	OCH_3	H	CH_3	1.20[c]			
23	H	OCH_3	OCH_3	H	H	0.77[c]			
24	OCH_3	OCH_3	OCH_3	H	CH_3	1.51[c]			
25	H	-OCH_2-		H	CH_3	1.64[c]			
26	H	H	H	H	H	1.52[c]			
27	H	OCH_3	Br	OCH_3	CH_3	2.44[c]			

[a] Calculated from equation (4).

[b] Not included in the regression.

[c] No biological data available for this preparation.

Attempts to incorporate electronic parameters such as Hammett σ values, indicated no importance of electronic character of the para substituent in the series.

The major problem lay in the fact that compounds 16 and 17 were much less active than predicted from their log P values. It has been previously suggested (2) that the length of n-butoxy and benzyloxy may exceed some critical value. This type of situation is not rare but no satisfactory substituent constants have been developed which adequately describe substituent "bulk" in a non-continuous way. The problem of enzyme or receptor tolerance for steric bulk on substrates or drugs has been commented on by many others.

On this basis, points 16 and 17 were dropped. Regression analysis then gave a slightly improved linear equation, using log P as the only independent variable. Addition of a second order term improved the equation greatly, but surprisingly, we found that a third order equation gave an excellent fit to the remaining data. This equation is:

$$\log RBR = 0.23 - 0.89 \log P + 0.95 (\log P)^2 - 0.16 (\log P)^3$$
$$n = 15;\ s = 0.13;\ r = 0.98 \qquad (2)$$
$$F_{3,11} = 77.86;\ F_{3,11\ \alpha\ 0.005} = 7.60$$

where again n is the number of data points, s is the standard deviation of the regression and r is the correlation coefficient. The third order term is significant at the 99.5% level ($F_{3,11} = 25.28$). This equation is illustrated in figure 1. We recognize that there is no theoretical justification for a third order relationship, and that the fit is purely empirical. However, inspection of the curve was useful in that it reveals that what is being described is essentially a linear relationship between activity and values of log P from 0.5 to about 3.0, beyond which activity rapidly falls off. Also, setting the differential of equation (2) equal to zero and solving gives a maximum at log P = 3.41, reasonably close to the *in vivo* optimum reported by Barfknecht *et al.* (1).

In view of the interdependence of log P and certain "bulk" terms (7) it seemed possible that the drop in activity in compounds 13-15 might be due to excessive steric bulk in the para position, rather than to increased hydrophobic character *per se*. This would be similar to the explanation for lowered activity in compounds 16 and 17. The only substituent constant which promised reasonable independence from log P, but which might be a crude measure of such "steric bulk" was molar refraction, MR (8). Accordingly, multivariable linear regression equations were developed which included all compounds active in this preparation. The use of log P and MR_4 (molar refraction of the para substituent) as independent variables led to equation (3):

$$\log RBR = 0.354 - 0.043\ MR_4 + 0.501 \log P$$
$$n = 17;\ s = 0.39;\ r = 0.81 \qquad (3)$$
$$F_{2,14} = 13.96;\ F_{2,14\ \alpha\ 0.005} = 7.92$$

Clearly, addition of a bulk term improved the correlation coefficient, and explained additional variance in the biological data. However, the standard deviation was still relatively high, and simple inspection of the data seemed to indicate the need for a non-continuous, or stepped, function which would take into account a sharp limit to the size of the hydrophobic binding site on the receptor.

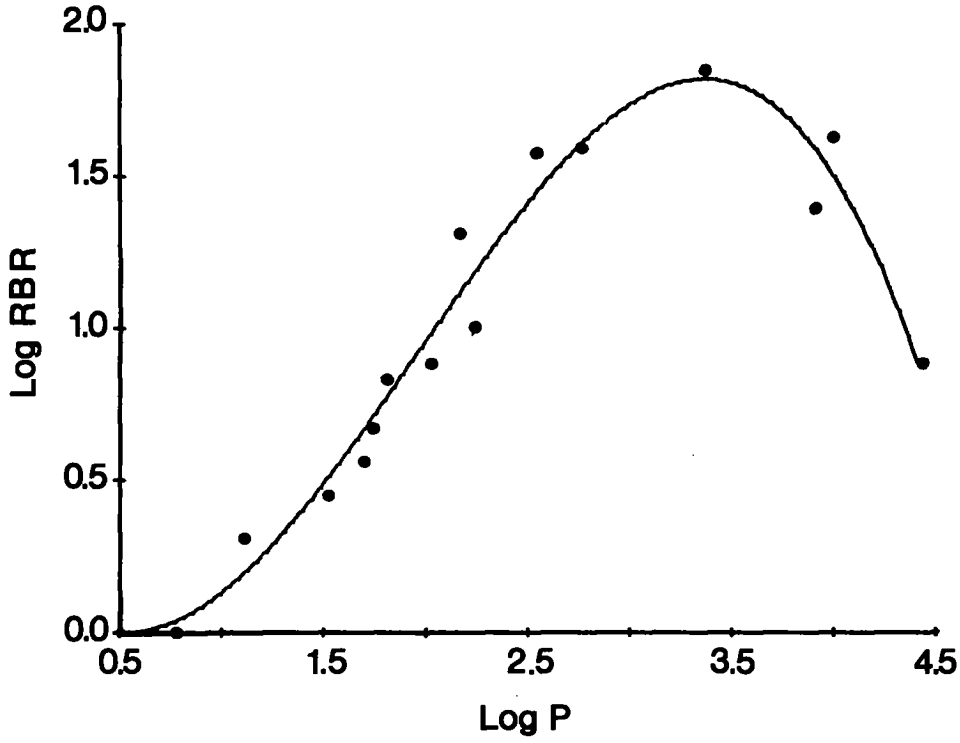


Figure 1

The plot of log in vitro activity in the sheep umbilical artery preparation against log P, using equation 2. Note the apparent optimum at log P = 3.41.

In an attempt to do this, an indicator parameter, I_4, was defined approximately as I_4 = (number of atoms in para substituent - 3). I_4 was defined as 0 for compounds 1 - 12, as 1 for compounds 13 and 15, and equal to 2 for compounds 14, 16, and 17. Linear regression analysis then resulted in equation (4):

$$\log RBR = -0.265 - 0.539\ I_4 + 0.595 \log P$$
$$n = 17;\ s = 0.23;\ r = 0.926 \qquad (4)$$
$$F_{2,14} = 42.05;\ F_{2,14\ \alpha\ 0.005} = 7.92$$

This represented the best equation which we were able to obtain using two independent variables, having reduced the standard deviation and increased the explained variance to 86% ($r^2 = 0.86$). The activities listed in table I are calculated from equation (4).

Many people question the relevance of in vitro or peripheral models for central processes. It was nevertheless felt that, at the least, this preparation might represent a useful screening method. On the other hand, it also seemed possible that the serotonin receptors in this preparation might resemble those in the central nervous system which are presumed to be involved in the action of hallucinogens. In that case, a detailed analysis of the

structure-activity-relationships for activity in this preparation might extrapolate directly to consideration of requirements for action at central receptors.

In view of such possible relevance, several points bear comment:

1. The serotonin receptors in this preparation appear to have a hydrophobic region which can bind to the para substituent of phenethylamine derivatives, but which is only able to accommodate a substituent less than 5-6 angstroms in length (2), or which is not highly branched. This represents a case of directional hydrophobicity.

2. The lack of dependence on electronic character of the para substituent leads to questions about the importance of molecular orbital calculations in describing activity.

3. The similar optimum log P for this preparation and for in vivo activity would seem to strengthen the arguments for the use of this model. The apparent optimum log P in this preparation appears to be largely determined by the steric bulk of the para substituent. It is not possible to assess the extent to which the optimum for human activity is dependent on this phenomenon. If the two receptors are similar, it suggests that a specific hydrophobic interaction may be more important in determining activity in man than the requirements for simple passive diffusion.

4. As suggested by Nichols and Dyer (2) 3,4,5-substituted compounds are about as active in this preparation as 2,4,5-substituted compounds of comparable log P. This work however, shows that this is independent of whether or not the compound possesses an alpha methyl group. (In contrast an alpha ethyl destroys hallucinogenic activity, reduces 5-HT agonist activity and imparts 5-HT antagonist activity to the molecule (9).)

This model has now been used successfully for predictive purposes. For example, 3,5-dimethoxy-4-bromophenylisopropylamine, compound 27, was predicted to have high potency based on its log P value and in vitro activity in this preparation for its 2 carbon homolog 5. Subsequent testing revealed that this compound elicits central effects in man at a dose in the range 3-6 mg. Likewise, this model predicts that in the series of compounds, 1-2-3 and 16, highest activity should reside in compounds 2-3 with a sharp drop in activity at compound 16. Although testing data is not complete, this prediction is also confirmed. Finally, compound 7 was originally predicted to be highly potent based solely on its log P value. Testing in this preparation further indicated activity in the expected range. It has now been demonstrated that this compound is indeed quite active in man (10).

We are quick to point out that this model will not be predictive for compounds which do not involve direct stimulation of serotonin receptors as their major mode of action such as compounds 18 and 19 (11-13). In this regard it is more likely that active compounds will be missed, than that activity will be predicted where none exists.

References

1. C. F. BARFKNECHT, D. E. NICHOLS, and W. J. DUNN, III, J. Med. Chem., 18, 208-210 (1975).
2. D. E. NICHOLS and D. C. DYER, J. Med. Chem., 20, 299-301 (1977).

3. A. T. SHULGIN and D. C. DYER, J. Med. Chem., 18, 1201-1204 (1975).
4. D. C. DYER and D. W. GANT, J. Pharmacol. Exp. Ther., 184, 366-375 (1973); D. C. DYER, ibid, 188, 336 (1974).
5. D. C. DYER, D. E. NICHOLS, D. B. RUSTERHOLZ, and C. F. BARFKNECHT, Life Sci., 10, 885-896 (1973).
6. A. T. SHULGIN, J. Med. Chem., 11, 186-187 (1968).
7. P. N. CRAIG, J. Med. Chem., 14, 680-684 (1971).
8. C. HANSCH, A. LEO, S. H. UNGER, K. H. KIM, D. NIKAITANI, and E. J. LIEN, J. Med. Chem., 16, 1207-1216 (1973).
9. D. C. DYER, Res. Comm. Chem. Path. Pharm., 14, 449-454 (1976).
10. A. T. SHULGIN and D. E. NICHOLS, in Psychopharmacology of Hallucinogens, ed. R. L. Stillman and R. E. Willette, Pergamon Press, in press, (1977).
11. H. C. CHENG, J. P. LONG, D. E. NICHOLS, and C. F. BARFKNECHT, J. Pharmacol. Exp. Ther., 188, 114-213 (1974).
12. H. C. CHENG, J. P. LONG, D. E. NICHOLS and C. F. BARFKNECHT, Arch. Int. Pharmacodyn. Ther., 212, 83-88 (1974).
13. D. E. NICHOLS, M. ILHAN, and J. P. LONG, Arch. Int. Pharmacodyn. Ther., 214, 133-140 (1975).