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[^{13}C]NMR Chemical Shifts and Calculated Electronic Structures of Serotonin Congeners: Relation to Biological Activity

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SUMMARY

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[^{13}C]NMR and quantum mechanical calculations of the electronic structure of some substituted tryptamines were applied to study the molecular determinants for the activity of these compounds on the LSD/serotonin receptor. A good correlation was observed between the measured [^{13}C]chemical shifts and the gross atomic charges obtained from *ab-initio* and near *ab-initio* (pseudopotential) calculations. These parameters, however, did not correlate with the biological activity of the compounds measured by their potency in contracting the rat fundus or by their ability to displace D-LSD from a high-affinity binding site in brain homogenates from guinea pig. For example, 5-fluorotryptamine (5-FT) resembles tryptamine more than serotonin in its biological potency but is most similar to serotonin in both the measured [^{13}C]chemical shifts and the calculated charge distribution. However, the frontier electron density maps above the indole portion of 5-FT bear a striking resemblance to those of tryptamine. The similar biological potency of 5-FT and tryptamine is in full accord with these findings and with results from our previous work from which the *localization pattern* of the electron density in the highest occupied molecular orbital emerged as a useful indicator of the relative affinity of tryptamines for the LSD/serotonin receptor. These conclusions support the use of judiciously chosen molecular reactivity criteria for the prediction of biological potency of drugs in this series.

INTRODUCTION

The wide distribution and potent pharmacological activity of 5-hydroxytryptamine (5-HT)[3] on a variety of tissues has

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[3] The abbreviations used are: 5-HT, 5-hydroxy-

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attracted attention to the pharmacological properties of tryptamine derivatives. In low concentrations (0.1 nM to 1 μM), 5-HT, like many other tryptamines, contracts the isolated fundus of the rat, an effect that is blocked by D-lysergic acid diethylamide (D-LSD) and its 2-bromo derivative (BOL) (1–3). Early experiments with various congeners and derivatives of tryptamine probed the relation between the molecular structure and the action of these compounds (1–4). Theoretical efforts were also made to understand the molecular requirements for the biological activity of the compounds, based mainly on the electronic characteristics of the indole ring portion of the molecules (5–8). A correlation was found between the electronic structure of the tryptamines and their potency on the fundus receptor (9), which could be used to account quantitatively for the potencies of a small number of tryptamines on that receptor (10). These studies demonstrated several correlations of potencies with charge densities and frontier electron densities at the N1, C4 and C5 positions of the indole ring, and indicated that the greater potency of 5-HT (compared to other members of the congeneric series) was due to the effect of the hydroxyl substitution at the C5 position on the electronic structure of the ring. These correlations predicted accurately the potencies of the 4-NH_2, 5-F and 5-OH-7-Cl derivatives measured subsequently for their ability to contract the fundus. In addition, this analysis prompted the study of the action of 12 additional tryptamine derivatives on the fundus (10–12). The activity of the new compounds was found to obey the same qualitative picture (11), providing additional support for the hypothesis of an "electron-donor" role for the tryptamines in the complex with the receptor (13). More recently, it was shown that the rank order of potencies of these tryptamines in contracting the fundus was nearly identical to their potencies in blocking high affinity binding of LSD to brain membranes, implying that the two receptors are very similar (12). Not surprisingly, the potencies in this latter system correlated with the same electronic indices.

These results are, however, subject to the fundamental shortcoming of most correlation approaches of this type: the lack of a rigorous theoretical basis for the choice of the molecular reactivity indices used in the correlation. For example, although the energy of the Highest Occupied Molecular Orbital (HOMO) is intuitively considered to represent the "electron-donating" ability of a molecule, it must be only a very crude indicator if the specific interaction between a drug and its receptor happens to be *localized* in *particular* regions of the molecule. Such a localized "electron-donating" ability of a molecule would be closer to its local polarizability than to its total ionization potential (12, 14). It thus becomes important to identify and use indices of chemical and biological reactivity that bear a close relationship to the physical mechanism assumed for the interaction. For that reason, attempts have been made to correlate superdelocalizability indices derived from the Huckel Molecular Orbital (HMO) method to molecular complexation (5) and Mulliken charge and frontier electron distributions from CNDO and INDO calculations (11) with activity on isolated tissue. The most significant shortcomings of these approaches, however, are related to the very approximate nature of the molecular orbital methods from which the indices were obtained and to the basic inaccuracy inherent in an attempt to describe the effects of interactions on the continuous electronic density function by sample points calculated according to an approximation scheme (such as the point charges in the Mulliken population analysis).

More accurate computational methods for these molecular systems, at the *ab-initio* LCAO-SCF-MO calculational level, have made possible the formulation of a more direct link between the electronic structure of the active compounds and the proposed mechanisms of interaction with their receptors, by introducing reactivity criteria that are based on the explicit contribution from the electrons and the nuclei of the molecule as a whole (14).

tryptamine; D-LSD, D-lysergic acid diethylamide; BOL, 2-bromo derivative of D-LSD; HOMO, Highest Occupied Molecular Orbital; HMO, Huckel Molecular Orbital; TRYP, tryptamine; NMR, nuclear magnetic resonance; TMS, tetramethyl silane.

These studies led to an integral representation of the molecular reactivity characteristics: i) the position in space of the reactive sites in the molecule (e.g., the minima in the electrostatic potential) (14); ii) criteria predicting the orientation of these molecular active sites toward interacting molecular models representing receptor sites (e.g., electrostatic orientation vectors) (14, 15); and, iii) the "response" of the molecular charge distribution to perturbations that could be caused by the environment or by the receptor itself (e.g., the E_2 polarization maps) (12, 16). Clearly, this is a more realistic attempt toward understanding the molecular properties that determine biological activity.

A comparison of the reactivity characteristics of several tryptamine congeners indicated that the primary interaction of the protonated side chain with an anionic receptor site prepares the drugs for the formation of a "stacking complex" between their indole portions and a second site of the receptor (14). Based on the electrostatic orientation vectors obtained from the electrostatic potentials generated by the various tryptamine congeners it was predicted (14) that there will be a difference in the preferred orientation that the indole portion of each of the tryptamine congeners will assume toward the second site of the receptor: The orientation of these vectors is different in the indole portions of the different congeners. Assuming that optimal affinity requires an optimal alignment of this vector with a complementary receptor site (14), the preferred orientation of the indole rings of various congeners is expected to differ. For example, with the side chain interacting at an anionic site, the vector describing an optimal electrostatic alignment between the indole portion of 5-HT and a second receptor site would be almost perpendicular to the vector describing the optimal alignment of the indole fragment of 6-HT with the *same site* (12, 14). The energy expenditure required to reorient the indole portion of any congener so as to make it "electrostatically equivalent" in its orientation to the configuration assumed by the indole portion of 5-HT at the receptor, would affect the probability for optimal binding. This should be measurable as lower affinity to the receptor (16). Accordingly, if 6-HT is to assume an orientation that is electrostatically equivalent to that of 5-HT at the receptor, its apparent affinity should be lower (15). This is indeed the experimental situation.

Such predicted differences in the preferred configurations that various tryptamine derivatives would assume in stacking complexes were found to be consistent with results of theoretical calculations (15, 16): distinct differences were observed in the energy dependence of various geometrical configurations of stacking complexes between 5-hydroxy or 6-hydroxy substituted indole derivatives with reactants such as imidazolium (15, 16). Experimental data from crystallographic studies of "charge-transfer" complexes of tryptamine derivatives (17), and results from NMR spectroscopy (18) also indicate differences in the mutual configurations that 5-hydroxy, and 6-hydroxyindole derivatives assume in stacking complexes. Such verification and mutual reinforcement between theoretical and experimental findings have played an important role in our studies on the molecular determinants for the pharmacological action of tryptamine derivatives (12, 14–16). In particular, the calculated molecular charge distribution and properties derived from it have been shown to be valuable predictors of differences in the biological activity of the various tryptamine congeners. We therefore studied the relation between such theoretical parameters and the molecular physicochemical properties observed from [^{13}C]nuclear magnetic resonance spectroscopy for various tryptamine derivatives. Results are presented here for some representative monosubstituted tryptamines, with special emphasis on the correlation between the molecular properties and the biological activities of the drugs.

METHODS

Experimental. Tryptamine (TRYP) hydrochloride was obtained from Fluka. The creatinine sulfate complex of 5-hydroxy- and of 6-hydroxytryptamine and 5-fluorotryptamine (5-HT, 6-HT, and 5-FT, respectively) hydrochloride were purchased from Sigma Chemical Co. The [^{13}C]NMR spec-

tra were obtained of saturated solutions in d_6-dimethyl sulfoxide on Varian CFT-60 (20 MHz) or XL-100-12 (25.16 MHz) spectrometers in the pulse Fourier transform mode at probe temperature (ca. 35°). The spectrometers were locked to the deuterium signal of the solvent and chemical shifts were determined with respect to internal tetramethylsilane (accuracy ± 0.03 ppm). Spectra were measured both with ^1H on-resonance noise-decoupling, and for determination of quaternary carbons, with off-resonance noise decoupling (21). ^1H-coupled [^{13}C]spectra were run with setting the decoupler such that the nuclear Overhauser signal enhancement was retained (22). The ^1H-coupled spectra, through vicinal [^{13}C, ^1H] coupling constants, yielded information about the number of vicinal protons for the unsubstituted carbons and thus facilitated signal assignments (23, 24). C-2 could be assigned from its larger one-bond (^{13}C, ^1H) coupling constant (ca. 180–183 Hz) as compared to the outer carbon atoms (ca. 160 Hz). Selective ^1H-decoupling experiments and consideration of substituent effects on chemical shifts (23, 25) allowed complete signal assignment.

Theoretical. The calculations reported here were performed with the *ab-initio* LCAO-SCF-MO methods described previously (14), and with a model potential near *ab-initio* method (15). Results for TRYP, 5-HT and 6-HT were obtained from the *ab-initio* calculations; results for 5-FT are from near *ab-initio* calculations. We have shown (19) that the two methods yield virtually identical results for tryptamine derivatives such as the ones discussed here when the same valence atomic basis sets are used. The atomic basis used in the *ab-initio* calculations consists of five s- and three p-type Gaussian functions on the heavy atoms and two s-type Gaussians on the hydrogens, contracted to a minimal basis. The exponents for the (5s, 3p, 2s/2s, p, s) basis are the optimized values from Whitman and Hornback (20). The same valence atomic basis was used for the model potential calculations (15).

All calculations were performed with the tryptamine in the Falkenberg averaged geometry, as described before (14). The charge distribution patterns were studied in planes parallel to the indole ring system of each tryptamine derivative, at a distance of 1Å (1.9 a.u.) from the molecular plane, a distance chosen so as to reflect mainly contributions from the polarizable π-electron density. The atom numbering scheme and the methods used for the calculation of density maps and charges are consistent with all our previous definitions (14, 15, 19). The relevance of calculations on the cationic and/or neutral species in relation to the biological activity has been discussed in detail (14).

RESULTS

The combination of several techniques was used for peak assignment in the [^{13}C] spectra of the tryptamine derivatives. As an example, the assignment of the [^{13}C] signals of 5-HT hydrochloride is decribed briefly: The off-resonance [^{13}C], ^1H decoupled spectra showed the signals at δ = 108.38, 127.39, 130.73 and 150.26 to belong to quaternary carbons, i.e., to C3, C5, C8 and C9. The characteristic low field shift allows the signal at δ = 150.26 to be assigned to C5 which is bonded to oxygen (22). The high field signal at δ = 108.38 is characteristic for carbon atoms of the β-pyrrolic type and thus belongs to C3. A distinction between the C8 and C9 signals at δ = 127.39 and 130.72 is possible by considering the shifts of these carbons in tryptamine hydrochloride where the assignment presents no problems. Here, δ_8 = 136.22 is to low field of δ_9 = 126.75 because C8 is connected to nitrogen, cf., the situation in aniline (26a) and pyrrole (26b). Because a hydroxy group exercises a strong shielding effect on the *para* carbon (ca. −5 ppm in the structurally related 2-hydroxynaphthalene) and a slight deshielding effect on the *meta* carbons (ca. +1 ppm on C9 in 2-hydroxynaphthalene) (31), the signal at δ = 130.72 in 5-hydroxytryptamine is assigned to C8 and the one at δ = 127.93 to C9.

Of the signals for the proton-bearing carbons, the one at δ = 123.61 exhibits a one-bond (^{13}C, ^1H) coupling of ca. 182 Hz, which is characteristic for α-pyrrolic carbons (26c) and therefore allows assignment of this signal to C2. The remaining signals at δ =

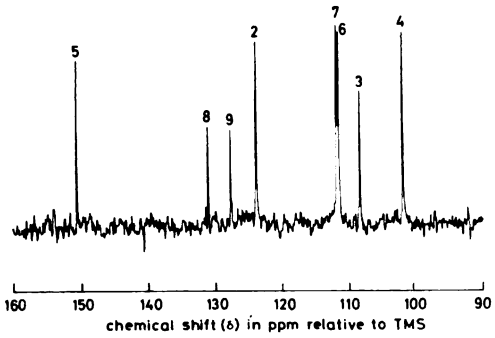

FIG. 1. *Proton decoupled [^{13}C]NMR spectra of 5-hydroxytryptamine*

Chemical shifts (δ) are given in ppm relative to tetramethyl silane (TMS) as an internal standard. Spectra were taken in saturated solution in d_6-dimethyl sulfoxide.

101.97, 111.39, and 111.72 then belong to carbons 4, 6 and 7. Carbons 4 and 6 can be differentiated by selective ^{13}C–^{1}H decoupling. Protons 4 and 6 absorb at δ_H = 6.84 and 6.63,[4] respectively, and show characteristic coupling patterns: a *meta* coupling of 2.2 Hz for H4, and an *ortho* coupling of 8.5 Hz and a *meta* coupling of 2.2 Hz for H6. Irradiation at δ_H = 6.84 collapses the signal at δ_C = 101.97 to a singlet. This signal therefore belongs to C4. Similarly, irradiation at δ_H = 6.63 decouples the signal at δ_C = 111.39 which then must belong to C6. Carbon 7 absorbs at 111.72.

The ^{13}C spectra for the remaining compounds were assigned in a similar fashion. Figure 1 shows a typical assigned spectrum. The spectra of the creatinine sulfate salts were found to be identical to those of the hydrochlorides of the same compounds. Notably, the spectra of the substituted indoles are also virtually identical to those of the indole portions of the corresponding tryptamines.[5] The chemical shifts for the four tryptamine derivatives are given in Table 1.

Calculated electronic structures. The distribution of the total electronic charge in the indole portion of the tryptamine derivatives is represented by the net atomic charges calculated for the cationic species (Table 1). The effect of the substituent and of its position in the indole ring is evident from the differences between the distribution of the net charges in the various derivatives. The relation between these charges and the [^{13}C]chemical shifts has been extensively discussed (27–30) with the general conclusion that the measured chemical shifts are proportional to the calculated total electron density at the carbon atoms. This relation has been shown to be valid at least in a comparative sense (28) for series of saturated hydrocarbons (27–30). A linear relation between the [^{13}C]chemical shift (δ_i) and the net charge (q_i) on atom i was also observed here for the carbon atoms in the indole portions of the tryptamine derivatives. A regression analysis yielded the following relations

$$\text{5-HT: } \delta_i = (0.009 \pm 0.002)q_i + 129. \pm 3. \quad (r = 0.903) \quad (1)$$

$$\text{6-HT: } \delta_i = (0.010 \pm 0.002)q_i + 129. \pm 3. \quad (r = 0.910) \quad (2)$$

$$\text{5-FT: } \delta_i = (0.010 \pm 0.002)q_i + 130. \pm 3. \quad (r = 0.918) \quad (3)$$

Of particular interest in these correlation is the fact that both the slopes and the intercepts of the lines are *nearly identical* for all three compounds, suggesting a common origin for the relation between the charges and the chemical shifts.

The chemical reactivity characteristics established for the indolealkylamines (14), and the nature of their interactions in model complexes (15, 16) have lead to the suggestion that these drugs would be likely to interact with the LSD/serotonin receptor by forming geometrically specific polarization complexes (for a review of these concepts see (12, 14, 15, 31)). It is therefore important to identify the localization of the sites in these molecules that are most susceptible to polarization interactions. Based on a quantum mechanical perturbation formalism (32, 33) it was shown (14) that the single largest contribution to the polarization of these molecules by an electrophilic interaction comes from a term involving mainly the highest occupied molecular orbital (HOMO): The perturbation expansion contains a term (E_2) that represents the change in energy due to polarization. The

[4] The solvent effect is responsible for the difference from previously reported values for δ_H of H4 and H6 (34).

[5] L. Ernst and S. Kang, unpublished results.

TABLE 1

[^{13}C]chemical shift (δ)[a] and Mulliken net charge distribution ($q \times 10^{-4}$ e) for tryptamine derivatives[b]

Position	TRYP		5-HT		5-FT		6-HT	
	δ	q	δ	q	δ	q	δ	q
C2	123.20	−695	123.61	−663	125.48 (~0)[c]	−662	121.07	−736
C3	109.46	−840	108.38	−870	109.95 (4.8)	−874	109.21	−809
C4	118.01	−2090	101.97	−2414	102.97 (23.1)	−2507	118.30	−1943
C5	118.34	−2163	150.26	1512	156.83 (231.0)	2217	109.02	−2668
C6	120.99	−2059	111.39	−2489	109.23 (25.7)	−2407	153.05	1630
C7	111.48	−2277	111.72	−2132	112.55 (9.6)	−2190	96.51	−2564
C8	136.22	1034	130.73	879	133.04 (~0)	969	137.39	1139
C9	126.75	−556	127.39	−454	127.14 (9.3)	−493	120.25	−706

[a] Given in ppm relative to high frequency of internal tetramethyl silane (TMS). Positive values mean deshielding.

[b] Spectra were taken in saturated solutions (≤8 mol %) in d_6-dimethyl sulfoxide. See text for details on the *ab-initio* method for the calculation of the molecular wave functions and charges.

[c] Values in parenthesis are [^{19}F]–[^{13}C]coupling constant in Hz.

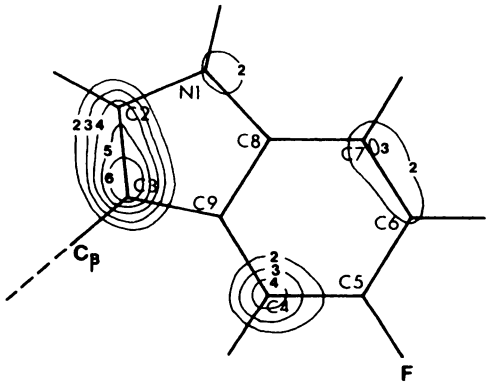

FIG. 2. *Map of the frontier electron density distribution*

The density generated by the highest occupied molecular orbital – HOMO in a plane 1Å above the indole ring system of 5-fluorotryptamine (5-FT). Contour values are n × 10^{-3} electrons/a.u.^{3}. See text for a description of the *ab-initio* method of calculation and the geometry.

matrix elements from the HOMO contribute up to 30% of the numerical value of the E_2 term (14).

Not surprisingly, the largest contribution from the HOMO to the polarization term (E_2) originates from the regions in which HOMO is localized (i.e., the regions in which HOMO places most of its electron density). This finding establishes a theoretical relation between the localization of the HOMO in tryptamine derivatives, and the sites in those molecules that would be most reactive in polarization interactions (12, 14). We have therefore compared the localization patterns in the HOMO of the tryptamine congeners by mapping the HOMO charge distributions (14). The map of electron density in the HOMO of 5-FT is shown in Figure 2. A comparison with Figure 6 of (14) indicates the striking similarity with the HOMO density of TRYP. The commonly used (albeit less suggestive and less accurate) representation of charge distributions by net charges obtained from the Mulliken population analysis is shown in Table 2 for the HOMO of 5-FT compared to those of TRYP, 5-HT and 6-HT.

It is possible to quantitate the similarities between the molecules with respect to the measured [^{13}C]chemical shifts and the calculated charge distributions: Linear regression analyses were performed on the measured (or computed) quantities for the same atoms in the different molecules. The highest possible similarity is expressed by a maximal correlation coefficient ($r = 1.0$) and a slope of unity. As an example, Figure 3 shows the correlation between [^{13}C] chemical shifts for 5-HT and 5-FT. The correlation coefficients for other regressions are given in Tables 3 and 4.

DISCUSSION

Results in Table 1 indicate a great similarity between 5-HT and 5-FT in both the distribution of the total electronic charge

TABLE 2

Distribution of electronic charge[a] in HOMO (frontier density) on atoms in the indole portion of neutral tryptamine derivatives (in $q \times 10^4$ e)

Molecule	Position							
	C2	C3	C4	C5	C6	C7	C8	C9
TRYP	−3846	−4663	−2793	−199	−2581	−2055	−745	−367
5-HT	−658	−3656	−4381	−2491	−61	−2568	−368	−234
5-FT	−2921	−5140	−3411	−657	−1588	−2152	−224	−29
6-HT	−4047	−3158	−1824	−123	−2722	−2392	−1121	−1320

[a] See text for details on the *ab-initio* method for the calculation of the molecular wave functions and the charges.

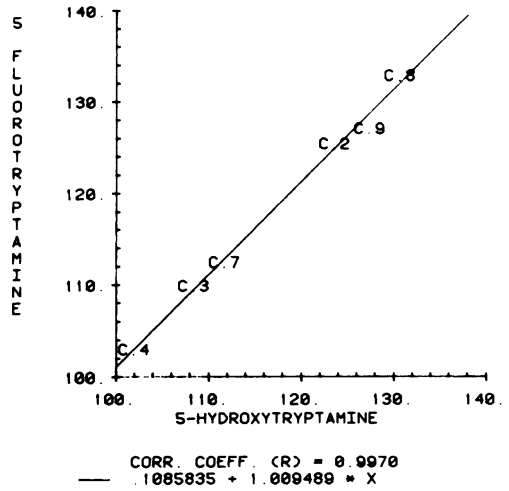

FIG. 3. *Linear regression analysis of [^{13}C]chemical shifts of 5-HT on those of 5-FT*

The chemical shifts of carbons 2, 3, 4, 7, 8 and 9 in the indole portion of each molecule were included in the regression.

and in the related [^{13}C] chemical shifts (Equations 1-3). If only the unsubstituted atoms of the indole portions in the two molecules are considered (i.e., carbons 2, 3, 4, 7, 8, and 9), the correlation coefficient for a statistical regression analysis of the similarity between the [^{13}C] chemical shifts in 5-HT and 5-FT is 0.997 (Fig. 3). The next best correlation is between the chemical shifts in 6-HT and TRYP (see Table 3). The correlation between the net charges on these molecules reveals the same qualitative picture (Table 3). It thus appears that the calculated charge distribution is a good descriptor of the electronic properties that are related with the chemical shifts of the molecules in solution. However, the biological activity of these molecules *does not* appear to be predictable from the distribution of the total charge nor from the measured chemical shifts: On the pharmacolog-

TABLE 3

Correlation coefficients[a] (r) from linear regression on net atomic charges (lower triangle)[b] and on [^{13}C]chemical shifts[c] (upper triangle)

	TRYP	5-HT	5-FT	6-HT
TRYP	1.0	0.824	0.830	0.908
5-HT	0.989	1.0	*0.9970*	0.630
5-FT	0.990	*0.9998*	1.0	0.652
6-HT	0.993	0.966	0.966	1.0

[a] Atoms included in the correlations are: C2, C3, C4, C7, C8, and C9 on the indole portion of the molecules.

[b] The correlation coefficients result from a linear regression analysis on the total charges on the *same* atoms in the *different* molecules.

[c] The correlation coefficients result from a linear regression analysis on the measured [^{13}C]chemical shifts of the *same* atoms in *different* molecules. See Figure 3 for an example of how the (5-HT, 5-FT) element of the upper triangle was obtained.

TABLE 4

Correlation coefficients[a] (r) from a linear regression on charges from the HOMO (lower triangle) and the NHOMO (upper triangle)[b]

	TRYP	5-HT	5-FT	6-HT
TRYP	1.0	0.649	*0.9428*	0.744
5-HT	0.578	1.0	0.841	0.149
5-FT	*0.9549*	0.783	1.0	0.506
6-HT	0.856	0.162	0.698	1.0

[a] Atoms included in the correlation are: C2, C3, C4, C7, C8, and C9 on the indole portion of the molecules.

[b] The correlation coefficients result from a linear regression analysis on charges on the *same* atoms in the *different* molecules. See footnote [c] in Table 3 and Figure 3 for an illustration of the procedure.

ical preparation from the rat fundus (11, 12), and from the affinity of the tryptamines for the receptors labeled by [^{3}H]-LSD in rat brain homogenate (12), 5-FT has similar potency to TRYP (both are ca. 10 times less potent than 5-HT), while 6-HT is at least 10–15 times less potent than TRYP.

On the other hand, the charge distribution in the HOMO has been shown to be directly correlated to the biological activity of the compounds (11, 12). The calculation for 5-FT presented here (Fig. 2) is consistent with this finding: Table 4 shows that the distribution of HOMO charges on the atoms of the indole fragment of 5-FT is much more similar to TRYP (correlation coefficient = 0.955) than to 5-HT (correlation coefficient = 0.783). In fact, the entire statistical picture offered by the utilization of HOMO charges as predictors of activity is close to the ranking of the biological potency of these compounds: TRYP and 5-FT are the closest, 5-FT is closer in potency to 5-HT than TRYP is, 6-HT is furthest in potency from 5-HT. The fact that the distribution of charges from NHOMO (the molecular orbital that is next to HOMO in energy) also correlates well with the biological activity, has been discussed previously (12) on the basis of the orthogonality properties of molecular orbitals. It has been shown (12) that the NHOMO is localized on the atoms that carry *the least* charge from HOMO, and that the correlation with the biological activity is therefore *implicit* in the correlation obtained with HOMO. In fact, Johnson and Green (11) observed this relation as a good *negative* correlation (i.e., with negative coefficients) between the pharmacological potency of the compounds on the rat fundus muscle and the charges on atoms C2, C6 and C9 of the indole portions. NHOMO of 5-HT was shown to be localized on these atoms (12). The results provide an illustration of the "polarizability localization" criterion as an indicator for the activity of tryptamines on an LSD/serotonin receptor (11, 12, 14–16).

It becomes clear that molecular structural and electronic parameters (e.g., HOMO energies, frontier densities) that are commonly used for correlation with biological activity, such as in studies on quantitative structure-activity relations (QSAR), have to be chosen with great care and with reference to the underlying physical origins of the molecular mechanism by which the drugs interact with their receptors (31).

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REFERENCES

1. Vane, J. R. The relative activities of some tryptamine analogues on the isolated rat stomach strip preparation, *Br. J. Pharmacol.* **14**: 87–98, 1959.
2. Barlow, R. B. and I. Kahn. Actions of some analogues of tryptamine on the isolated rat uterus and on the isolated rat fundus strip preparations, *Br. J. Pharmacol.* **14**: 99–101, 1959.
3. Barlow, R. B. and I. Kahn. Actions of some analogues of 5-hydroxytryptamine on the isolated rat uterus and the rat fundus strip preparations, *Br. J. Pharmacol.* **14**: 265–272, 1959.
4. Offermeier, J. and E. J. Ariens. Serotonin. II. Structural variation and action, *Arch. int. Pharmacodyn.* **164**: 216–245, 1966.
5. Green, J. P. and J. P. Malrieu. Quantum chemical studies of charge-transfer complexes of indoles, *Proc. Nat. Acad. Sci. U.S.A.* **54**: 659–664, 1965.
6. Snyder, S. H. and C. R. Merril. A relationship between the hallucinogenic activity of drugs and their electron configuration. *Proc. Nat. Acad. Sci. U.S.A.* **54**: 258–266, 1965.
7. Millie, P., J. P. Malrieu, J. Benaim, J. Y. Lallemand, and M. Julia. Researches in the indole series. XX. Quantum mechanical calculations and charge-transfer complexes of substituted indoles, *J. Med. Chem.* **11**: 207–211, 1968.
8. Kier, L. B. Preferred conformation of serotonin and a postulate on the nature of its receptor from molecular orbital calculations. *J. Pharm. Sci.* **57**: 1188–1191, 1968.
9. Kang, S. and J. P. Green. Resonance constants and the activities of indole-alkylamines on stomach muscle. *Nature* **222**: 794, 1969.
10. Kang, S. and J. P. Green. Steric and electronic relationships among some hallucinogenic compounds, *Proc. Nat. Acad. Sci. U.S.A.* **67**: 62–67, 1970.
11. Johnson, C. L. and J. P. Green. Molecular orbital

studies on tryptamines active on the LSD receptor of the rat fundus strip, *Int. J. Quantum Chem.* QBS **1**: 159–167, 1974.

12. Green, J. P., C. L. Johnson, H. Weinstein, S. Kang, and Chou, D. Molecular determinants for interaction with the LSD receptors: biological studies and quantum chemical analysis. In *The Psychopharmacology of Hallucinogens*, edited by R. E. Willette and R. C. Stillman. New York, Pergamon Press, 1978, 28–60.
13. Doukas, P. H. The role of charge-transfer processes in the action of bioactive materials, in *Drug Design*, Vol. 5, edited by Ariens, E. J. Academic Press, New York, 1975, 133–167.
14. Weinstein, H., D. Chou, S. Kang, C. L. Johnson, and J. P. Green. Reactivity characteristics of large molecules and their biological activity: Indole-alkylamines on the LSD-Serotonin receptor. *Int. J. Quant. Chem.* **QBS3**: 134–150, 1976.
15. Weinstein, H. and R. Osman. Models for molecular mechanisms in drug-receptor interactions: serotonin and 5-hydroxyindole complexes with imidazolium cation. *Int. J. Quant. Chem.* **QBS4**: 253–268, 1977.
16. Weinstein, H., R. Osman, W. D. Edwards, and J. P. Green. Theoretical models for molecular mechanisms in biological systems: tryptamine congeners acting on an LSD/serotonin receptor. *Int. J. Quant. Chem.* **QBS5**: 449–461, 1978.
17. Thewalt, U. and C. E. Bugg. The crystal and molecular structure of serotonin picrate monohydrate. *Acta Cryst.* **B28**: 82–92, 1972.
18. Sapper, H. and W. Lohmann. Self-association and binding sites of some psychotomimetic tryptamine derivatives and related compounds: nuclear magnetic resonance investigations. *Mol. Pharmacol.* **12**: 605–611, 1976.
19. Osman, R. and H. Weinstein. Near ab-initio methods for the calculation of large molecules: comparison of pseudopotential and ab-initio results. *Chem. Phys. Letters* **49**: 69–74, 1977.
20. Whitman, D. R. and C. J. Hornback. Optimized gaussian basis SCF wave-functions for first-row atoms. *J. Chem. Phys.* **51**: 398–402, 1969.
21. Wenkert, E., A. O. Clouse, D. W. Cochran, and D. Doddrell. Carbon-13 NMR spectroscopy of naturally occurring substances. I. A direct analysis of nonprotonated carbon centers. *J. Amer. Chem. Soc.* **91**: 6879–6880, 1969.
22. Freeman, R. and H. D. W. Hill. Nuclear overhauser effect in undecoupled NMR spectra of C-13. *J. Magn. Res.* **5**: 278–280, 1971.
23. Ernst, L. ^{13}C NMR-spectroscopy of polycyclic aromatics. 1. Naphtols, naphthalenediols and methoxynaphthalenes—complete assignment of ^{13}C NMR spectra and investigation of substituent effects on chemical shifts. *Chem. Ber.* **108**: 2030–2039, 1975.
24. Gunther, H., H. Schmickler, and G. Jikeli. Applications of carbon-13 resonance spectroscopy. V. Fingerprints for the assignment of carbon-13 resonance signals. *J. Magnet. Reson.* **11**: 344–351, 1973.
25. Ernst, L. ^{13}C NMR spectroscopy of polycyclic aromatics. 2. ^{13}C NMR spectra of 1-fluoronaphthalene and 2-fluoronaphthalene. Reexamination of signal assignments and investigation of substituent effects on chemical shifts. *Z. Naturforsch.* **30**: 788–793, 1975.
26. Strothers, J. B. *Carbon-13 NMR Spectroscopy.* Academic Press, New York, 1972, a) p. 197; b) p. 253; c) 343.
27. Sardella, D. J. Substituent-induced ^{13}C chemical shifts. A perturbation approach. *J. Am. Chem. Soc.* **95**: 3809–3811, 1973.
28. Fliszar, S. Charge distributions and chemical effects. XII. A unifying concept. *Can. J. Chem.* **54**: 2839–2846, 1976.
29. Fliszar, S., A. Goursot, and H. Dugas. Charge distributions and chemical effects. V. A modified population analysis. Application to ^{13}C NMR shifts. *J. Am. Chem. Soc.* **96**: 4358–4359, 1974.
30. Kean, G., D. Gravel, and S. Fliszar. Charge distributions and chemical effects. On the charge dependence of ^{13}C chemical shifts in adamantane and related six-membered polycyclic molecules. *J. Am. Chem. Soc.* **98**: 4749–4754, 1976.
31. Weinstein, H., Green, J. P., Osman, R. and Edwards, W. D. Recognition and activation mechanisms on the LSD/serotonin receptor: The molecular basis of structure activity relationships. In: *QuaSAR-Quantitative Structure Activity Relationships of Analgesics, Narcotic Antagonists and Hallucinogens*, eds. G. Barnett, M. Trsic and R. E. Willette, NIDA Research monograph #22, National Institute on Drug Abuse, pp. 333–358, 1978.
32. Bartlett, R. J. and H. Weinstein. Theoretical treatment of multiple site reactivity in large molecules. *Chem. Phys. Letters* **30**: 441–445, 1975.
33. Chang, S. Y., H. Weinstein, and D. Chou. Perturbation treatment of multiple site reactivity: molecule-molecule interactions. *Chem. Phys. Letters* **42**: 145–150, 1976.
34. Kang, S., T. H. Witherup, and S. Gross. Acid catalyzed deuterium exchange of the indole ring protons in tryptamine derivatives. *J. Org. Chem.* **42**: 3769–3772, 1977.