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Ionization constants and water solubilities of some aminoalkylphenothiazine tranquillizers and related compounds

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A simple graphical method is described for deriving the ionization constants of poorly soluble organic bases from the pH dependence of the water solubility. Ionization constants and water solubilities of 13 aminoalkylphenothiazines and related drugs have been determined using this procedure. Potent tranquillizing activity in this group of compounds is shown to be associated with a low water solubility at blood pH.

THE choice of a suitable method for determining the ionization constants of aminoalkylphenothiazine tranquillizers and related drugs is severely restricted by the very poor water solubility of the free bases. This low solubility prohibits the use of conductivity measurements, or of what is normally the most satisfactory method, namely potentiometric titration in aqueous solution. Furthermore, the ultraviolet spectra of the base and cation are too alike for the ionization constant to be calculated from the extinction in buffers of different pH. However, for bases which are much less soluble in water than their corresponding salts, it is possible to derive the ionization constants from the pH dependence of the water solubility. This method has been used here.

Experimental

MATERIALS

The drugs used were kindly supplied by various pharmaceutical companies as follows: amitriptyline hydrochloride (Merck, Sharp & Dohme), desipramine hydrochloride and imipramine hydrochloride (Geigy), chlorpromazine hydrochloride, pecazine hydrochloride, prochlorperazine ethanedisulphonate and trifluoperazine dihydrochloride (Smith Kline & French), fluopromazine hydrochloride (Squibb), perphenazine (Allen & Hanbury), promazine hydrochloride (Rhône-Poulenc), promethazine hydrochloride (May & Baker), thiopropazate dihydrochloride (Searle) and thioridazine hydrochloride (Sandoz).

Buffers were prepared as described by Gomori (1955) from sodium phosphate, tris, 2-amino-2-methylpropane-1,3-diol and glycine.

METHODS

Two methods were used to obtain the solubilities. In the first and simpler method, solid amine salt or free base was shaken with 10 ml of 0·01N sodium hydroxide or 0·01M buffer at room temperature (24 $\pm$ 1°). Sufficient compound was added to ensure that some undissolved material was always present. After 3 hr, this undissolved material was removed

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by centrifuging. The pH of the solution and its extinction at the wavelength of maximum absorption, generally close to 255 or 305 mμ (Warren, Eisdorfer, Thompson & Zarembo, 1966), were then measured. The extinction was converted into concentration of dissolved amine with the aid of the extinction coefficient calculated from a solution of the amine salt at known concentration in 0·01N sodium hydroxide (the compounds obeyed Beer's Law).

Prolonged centrifuging at high speed was generally needed to clarify the solutions adequately; even then the method occasionally failed to give satisfactory results. Consequently, the solubilities were also determined by the following, rather lengthier, procedure.

A series of dilutions of an amine salt in water (4·5 ml) were shaken for 45 min at room temperature (24 ± 1°) with 0·5 ml of 0·1N sodium hydroxide or 0·1M buffer. The extinction of each solution was then measured at 450 mμ. The compounds studied do not absorb light significantly at this wavelength and the observed extinction is due to the scattering of light by the turbid suspension which is formed if the concentration of the free base at the pH of the solution exceeds its solubility. A plot of extinction against concentration of amine salt generally gives two straight lines (one on the horizontal axis) intersecting at the solubility of the base. Typical plots are illustrated in Fig. 1. Occasionally, the turbidity did not

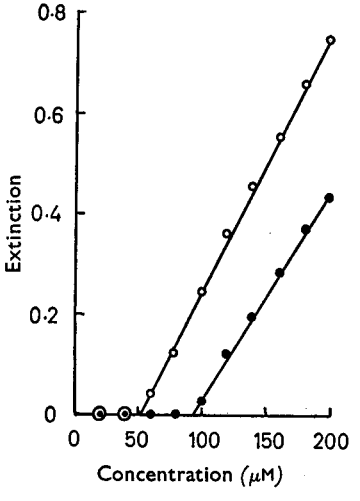


FIG. 1. Plot of extinction against concentration of amitriptyline hydrochloride at pH 9·78 (open circles) and pH 9·20 (filled circles).

vary linearly with concentration; but even so, the point of intersection could usually be determined with fair precision.

The ionization constants were derived from the pH dependence of the solubilities as follows. At any pH, the total concentration of the base plus salt ([B] + [HB$^+$]) will equal [B] (1 + [H$^+$]/K) where K is the ionization constant (K = [H$^+$] [B]/[HB$^+$]). If the free base is much less soluble

than the salt, the observed total solubility at a particular pH, S, will be related to the hydrogen ion concentration and the solubility of the free base, S_o, by the equation,

$$S = S_o(1 + [H^+]/K)$$

(when $[H^+]$ is small $S = S_o$). This can be rearranged to give

$$[H^+] = (K/S_o)S - K$$

Thus a plot of $[H^+]$ against S should give a straight line with intercepts S_o and $-K$ on the S and $[H^+]$ axes respectively. Two typical plots are shown in Fig. 2. The solubilities in these plots were obtained by the turbidity method, but precisely similar plots were obtained when the more direct method was used.

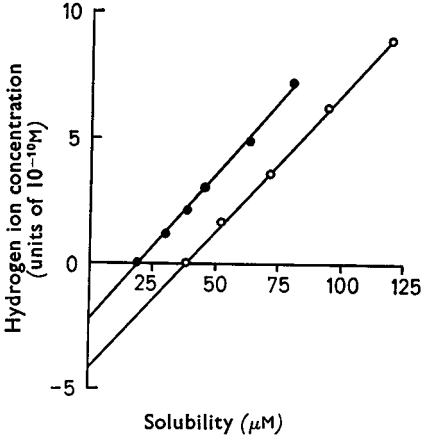
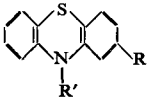
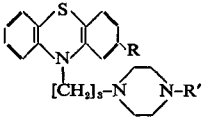
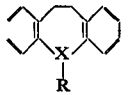


FIG. 2. Relationship between hydrogen ion concentration and solubility of pecazine (filled circles) and amitriptyline (open circles).

Results and discussion

The turbidity method for determining the solubilities gave satisfactory linear plots of hydrogen ion concentration against solubility for all the compounds studied. The more direct method gave good results with most compounds, but failed for pecazine and thiopropazate, for which only very scattered plots could be obtained. Where both methods were used, the pK_a values always agreed within $\pm 0{\cdot}2$ units, and generally within $\pm 0{\cdot}1$ units. Duplicate determinations by either method also gave values agreeing within $\pm 0{\cdot}1$ units. Mean pK_a values for ten aminoalkylphenothiazines and three related antidepressants are given in Table 1. The solubilities of the free bases listed in Table 1 are also the means of those obtained by the two methods. However, the solubilities obtained by the direct method were always lower than those found by the turbidity method. The difference tended to be proportionately greater the lower the solubility, although even with the two least soluble compounds, thioridazine and fluopromazine, the results by the two methods were

TABLE 1. WATER SOLUBILITIES AND pK_a VALUES OF AMINOALKYLPHENOTHIAZINES AND RELATED COMPOUNDS

Structure	Trivial or approved name	pK_a		Solubility (μM)	Calculated relative solubility at pH 7·4
		Solubility method	Chatten & Harris (1962)		
 R = H; R′ = $CH_2{\cdot}CH(Me){\cdot}NMe_2$	Promethazine	9·1	9·1	55	4·5
R = H; R′ = $[CH_2]_3{\cdot}NMe_2$	Promazine	9·4	—	50	8·0
R = Cl; R′ = " "	Chlorpromazine	9·3	9·2	8	1·0
R = CF_3; R′ = " "	Fluopromazine	9·2	9·4	5	0·4
R = H; R′ = CH_2– 	Pecazine	9·7	—	18	5·0
R = SMe; R′ = $[CH_2]_2$– 	Thioridazine	9·5	9·2	1·5	0·3
 R = Cl; R′ = Me	Prochlorperazine	8·1	7·5	40	0·4
R = CF_3; R′ = Me	Trifluoperazine	8·1	8·4	30	0·3
R = Cl; R′ = CH_2CH_2OH	Perphenazine	7·8	—	70	0·35
R = Cl; R′ = CH_2CH_2OCOMe	Thiopropazate	7·3	7·2	20	0·06
 X = N; R = $[CH_2]_3{\cdot}NHMe$	Desipramine	10·2	—	220	250
X = N; R = $[CH_2]_3{\cdot}NMe_2$	Imipramine	9·5	—	65	15
X = C; R = : $CH[CH_2]_2{\cdot}NMe_2$	Amitriptyline	9·4	—	35	6·0

within a factor of 1·7. Aminoalkylphenothiazines have a high surface activity and tendency to form micelles (Scholtan, 1955; Seeman & Bialy, 1963), hence some supersaturation may occur when the free base is precipitated from solution. It is consequently possible that the true solubilities may be slightly lower than those in the Table. There was one curious anomaly which could be accounted for by a supersaturation effect of this kind. Perphenazine was available both as dihydrochloride and as free base. When the dihydrochloride was used, both the direct and the turbidity methods gave the same solubility, 70 μM, but when the direct method was applied to the free base, a solubility of only 26 μM was obtained. The same pK_a value was found irrespective of whether the base or salt was used.

Schill (1965) has also obtained water solubilities and pK_a values at 20° for promazine and chlorpromazine by a procedure similar to my direct method, but by use of a non-linear instead of a linear plot. His pK_a values, 9·4 and 9·3 respectively, and his solubility for promazine (50 μM) are in agreement with my values, but not so his solubility for chlorpromazine (3·6 μM) which is considerably lower.

A common expedient for obtaining ionization constants of bases which are poorly soluble in water, but freely soluble in organic solvents, is to titrate them potentiometrically in mixtures of organic solvent and water, and then to extrapolate the pK_a values so obtained to zero organic solvent content. This procedure is open to serious errors (Albert & Serjeant, 1962) but pK_a values for some aminoalkylphenothiazines in water have been determined by extrapolation from values obtained by potentiometric titration in aqueous methanol (Marshall, 1955; Chatten & Harris, 1962). Marshall obtained values of 9·5, 9·3 and 9·0 for promazine, chlorpromazine and promethazine respectively; the values obtained by Chatten & Harris are included in Table 1. The agreement between the titration and solubility methods is generally good. The only serious discrepancy is found with prochlorperazine and trifluoperazine, which have the same pK_a values when these are determined by the solubility method, but have markedly different values when measured by titration. Since chlorpromazine has about the same pK_a as fluopromazine when measured by either method, it seems improbable that there would be any large difference between the pK_a values of the corresponding *N*-methylpiperazine analogues.

The major factor controlling the ionization constant of these compounds is the nature of the aminoalkyl side-chain. Substitution in the phenothiazine nucleus has only a minor effect on the pK_a although it causes a sharp drop in solubility. Replacement of the phenothiazine group of promazine by the dibenzazepine system in imipramine also results in only small increases in pK_a and solubility. Even replacement of the $>N—CH_2—$ group of imipramine by $>C=CH—$ in amitriptyline causes no major change in either the pK_a or the solubility. The effect of variation in the structure of the amino-group is much the same as found for other substituted aliphatic amines. Demethylation of imipramine to give desipramine increases the pK_a by 0·7 units, and replacement of the

dimethylamino-group by *N*-methylpiperidine, in pecazine and thioridazine, raises the pK_a by about 0·3 units. These differences are typical of those normally found between monomethylamino- and dimethylamino-compounds, and between dimethylamino- and *N*-methylpiperidino-compounds (Clark & Perrin, 1964; Perrin, 1965). Insufficient pK_a values have been published for piperazine derivatives to enable any quantitative correlation with the present series, but the values obtained for prochlorperazine, perphenazine and thiopropazate are consistent with the increasing electron-withdrawing properties of the substituent on the terminal nitrogen atom. Piperazine derivatives are dibasic, but as the second pK_a value is much lower than the first (Chatten & Harris, 1962), the equations on pages 11–12 need no modification for solubilities determined at pH 7 or above.

No very extensive correlation is discernable between ionization constant, water solubility and tranquillizing activity, but there is a trend for depressant activity to be associated with a low pK_a or low water solubility. A rough guide to the likely joint influence of these two factors is the water solubility at blood pH (7·4). The calculated solubilities at pH 7·4 relative to that of chlorpromazine are given in the final column of Table 1. The potent tranquillizing drugs all have relative solubilities of 1 or less, whereas the antidepressant drugs imipramine and desipramine have relative solubilities greater than 10. The antidepressant drug amitriptyline, with a relative solubility of 6, also produces sedation and can be used as a combined antidepressant-tranquillizer (Freed, 1960). Promazine and pecazine, with relative solubilities of 8 and 5, have weak tranquillizing activity, but are used far less for this purpose than the more active, ring-substituted compounds. Promethazine, which has a relative solubility of 4·5, causes some sedation, but it is not a tranquillizer. However, this drug has only 2 carbon atoms separating the phenothiazine ring from the terminal amino-group, and consequently does not conform to one of the generally accepted structural requirements for tranquillizing activity, namely a trimethylene chain between the ring system and the amino-group (Gordon, Craig & Zirkle, 1964). The relative solubility at pH 7·4 may thus provide a useful indication of whether tranquillizing or antidepressant activity will predominate in any series of compounds having appropriate structures for these types of activity; but, the converse, that bases with a low solubility at pH 7·4 will have tranquillizing properties, will generally not be true.

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