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COLORIMETRIC DETERMINATION OF PHENOTHIAZINE DRUGS. 3-CORRELATION BETWEEN MOLAR ABSORPTIVITY AND CERTAIN PHYSICOCHEMICAL PARAMETERS OF RING SUBSTITUENTS.

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ABSTRACT

The color intensity developed from the reaction of twelve phenothiazine drugs with I_3^- has been correlated using physicochemical substituent like Hammett constant σ_p, and molecular connectivity $^1\chi^v$. Results indicated a predominant role of electronic parameter in determining color intentsity. The bulkiness described by $^1\chi^v$ played a significant role. However, other parameters of the side chain were also prominent. Working up multiple parametric regression can be used as a reliable tool to predict the sensitivity and scope of the colorimetric method for other phenothiazines.

INTRODUCTION

Recently two colorimetric methods (parts 1 and 2) have been developed for quantitation of phenothiazine derivatives in bulk and in dosage forms[1,2].

Part 1 has been concerned with the optimization of conditions of the reaction involved in the development and elucidation of the structure of the isolated chromogen. This reaction essentially depends on the oxidation of the phenothiazine ring with I_3^- in presence of morpholine using isopropanol as a diluent and the absorbance measurement at the appropriate λ_{max} for each drug[1].

It was observed that the color intensity depends on the nature of the substituents at C-2 and the side chain attached to nuclear N-10. This observation stimulated the interest to examine the contribution of certain numerical parameters of these substituents to the color intensity. The molar absorptivities of the reacted phenothiazines have been correlated separately with : Hammett constant σp, of C-2 substituents and and the molecular conectivity index $({}^1\chi_1^v)^2$ of the side chains at N-10. Furthermore, a multiparametric regression equation has been undertaken to combine both variables, σp and $({}^1\chi_1^v)^2$. This approach can help to decide the scope of application of the suggested method for the analysis of other phenothiazines than those directly involved in the present paper.

EXPERIMENTAL

Log ε_{max} for the analysed phenothiazines (Table 1) have been taken from published data[1]. The parameters for the C-2 functions have been taken from reference[3], and included as electronic contribution variable,

Calculation of molecular connectivity followed the method described by Kier and Hall[4]. The structural representation for the calculation of the

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first order indexes $^{1}\chi^{v}$ is illustrated by Scheme 1. A value for each molecule bond k is computed from each pair of bonded atoms $(\delta_i\ \delta_j)^{-1/2}$, where i and j are the bonded atoms. Finally $^{1}\chi^{v} = \sum_{K=i}^{K=m} (\delta_i\ \delta_j)^{-1/2}$ where δ_i^v and δ_j^v values represent the difference between the number of valency electrons of a given atom and the hydrogen atoms attached to this atom.

RESULTS AND DISCUSSION

Different electronic parameters like Hammett σp and σm; Swain and Lupton $\mathcal{F}$ and $\mathcal{R}$; and molar refraction MR[3] have been correlated to substituents at C-2. The statistical analysis (Compounds II-VI) showed that on keeping the side chain at N-10 unchanged σp was the best variable to correlate with log ε max (eq. 1).

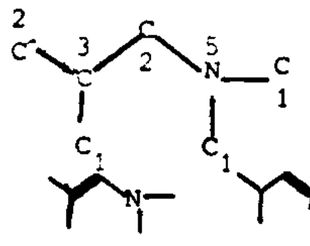
$$\log \varepsilon = 3.907 - 7.169(\pm 4.00)\ \sigma p \qquad \text{eq. 1}$$

$$r = 0.943 \qquad s = 0.667 \qquad n = 5$$

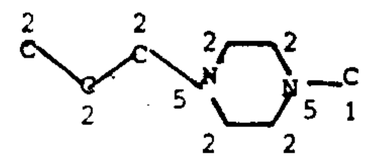
$$F_{(1,3)} = 24.24 \qquad p < 0.05$$

In eq.1 and the following equations, n is the number of compounds included in the analysis, r is the correlation coefficient, and s is the standard deviation while F is the F-ratio between the variances of the observed and calculated values at the given p value. The figure in parantheses expresses the 95% confidence interval.

Equation 1 accounts only for 85% ($\tilde{\tilde{r}} = 0.85$) of the variance in log ε_{max} (where $\tilde{r}^2$ unbiased estimator of the population coefficient of determination ρ^2 which is similarly correlated as r^2). The residual 15% of unexplained variance can be interpreted in light of the possible interaction between the electro-

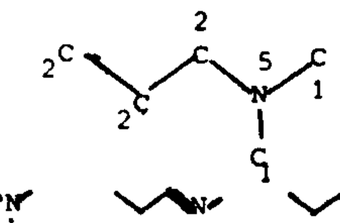
1- 

$^{1}\chi^{v}_{1} = (2x3)^{-\frac{1}{2}} + (3x1)^{-\frac{1}{2}} + (3x2)^{-\frac{1}{2}} + (2x5)^{-\frac{1}{2}} + (5x1)^{-\frac{1}{2}} + (5x1)^{-\frac{1}{2}} =$ =2.603

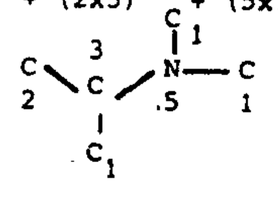
2- 

$^{1}\chi^{v}_{1} = (2x2)^{-\frac{1}{2}} + (2x2)^{-\frac{1}{2}} + (2x5)^{-\frac{1}{2}} + (5x2)^{-\frac{1}{2}} + (2x2)^{-\frac{1}{2}}$

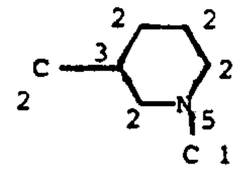
$(2x5)^{-\frac{1}{2}} + (5x1)^{-\frac{1}{2}} + (5x2)^{-\frac{1}{2}} + (2x2)^{-\frac{1}{2}} + (2x5)^{-\frac{1}{2}}$ =4.027

3- 

$^{1}\chi^{v}_{1} = (2x2)^{-\frac{1}{2}} + (2x2)^{-\frac{1}{2}} + (2x5)^{-\frac{1}{2}} + (5x1)^{-\frac{1}{2}} + (5x1)^{-\frac{1}{2}}$ =2.21

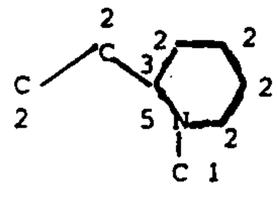
4- 

$^{1}\chi^{v}_{1} = (2x3)^{-\frac{1}{2}} + (3x1)^{-\frac{1}{2}} + (3x5)^{-\frac{1}{2}} + (5x1)^{-\frac{1}{2}} + (5x1)^{-\frac{1}{2}}$ =2.137

5- 

$^{1}\chi^{v}_{1} = (2x3)^{-\frac{1}{2}} + (3x2)^{-\frac{1}{2}} + (2x2)^{-\frac{1}{2}} + (2x2)^{-\frac{1}{2}} + (2x5)^{-\frac{1}{2}}$

$(5x1)^{-\frac{1}{2}} + (5x2)^{-\frac{1}{2}} + (2x3)^{-\frac{1}{2}}$ =3.303

6- 

$^{1}\chi^{v}_{1} = (2x2)^{-\frac{1}{2}} + (2x3)^{-\frac{1}{2}} + (3x2)^{-\frac{1}{2}} + (2x2)^{-\frac{1}{2}} + (2x2)^{-\frac{1}{2}}$

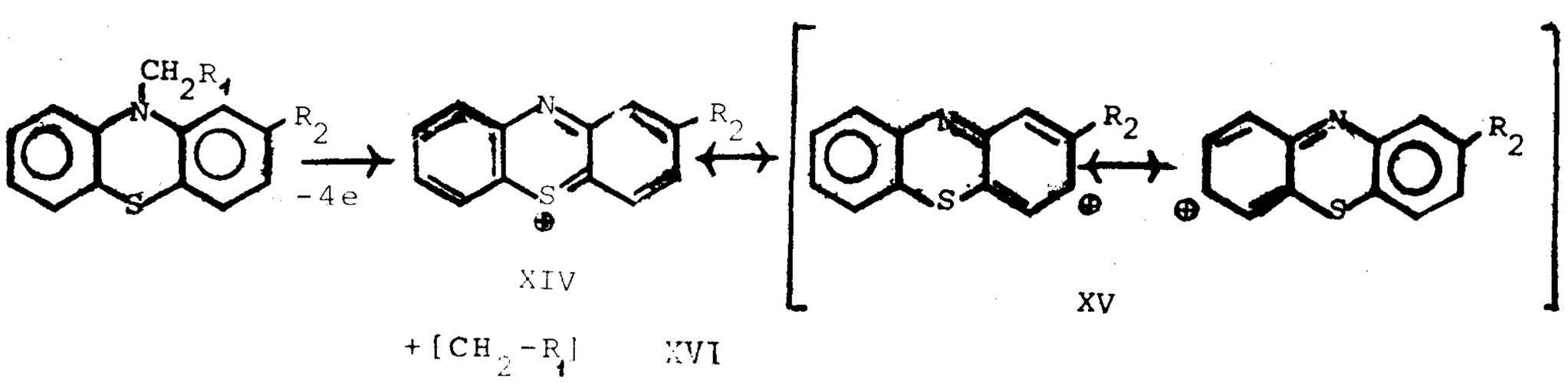
$(2x2)^{-\frac{1}{2}} + (2x5)^{-\frac{1}{2}} + (5x1)^{-\frac{1}{2}} + (5x3)^{-\frac{1}{2}}$ =3.837

Scheme 1

philic group at C-2 and the available n-electrons of the unprotonated piperazine moiety (Table 1). Such intramolecular interaction has been already reported for similarly substituted compounds and leads to significant modifications of the IR spectrum[5].

Equation 1 shows that log ξ_{max} decreases by increasing the electronegativity of substituent C-2. At higher σp values ≥ 0.48, the calculated $\xi < 3$, and practically no color can be noticed under the reaction conditions. On the contrary to that expected from eq. 1 levomepromazine (I) carrying electron donating group (C-2= OCH_3;) displayed a log ξ_{max} value less than alimemazine (VIII) (C-2 = H). We can infer from this remark that chromogen synthesis is controlled by a dual electronic effect of C-2 substituents. Thus strong electron withdrawing groups may hinder the formation of the phenazathionium ion XIV (Scheme 2), which takes place via the abstraction of four electrons[6]. Mild electron withdrawing group will not block the intiation of ion XIV, at the same time will relatively facilitate nucleophilic attack by the amine reagent (morpholine) to give the chromogen already isolated and identified[1].

In case of levomepromazine with an electron-donating group at C-2, it seems that the compound XIV is easily formed and the mesomeric structure XV is more stabilized to give a better chance for attack by abundant nucleophiles in the reaction medium. Under these conditions in lieu of morpholine other nucleophiles will compete for the reactive sites to yield products besides the target chromogen.

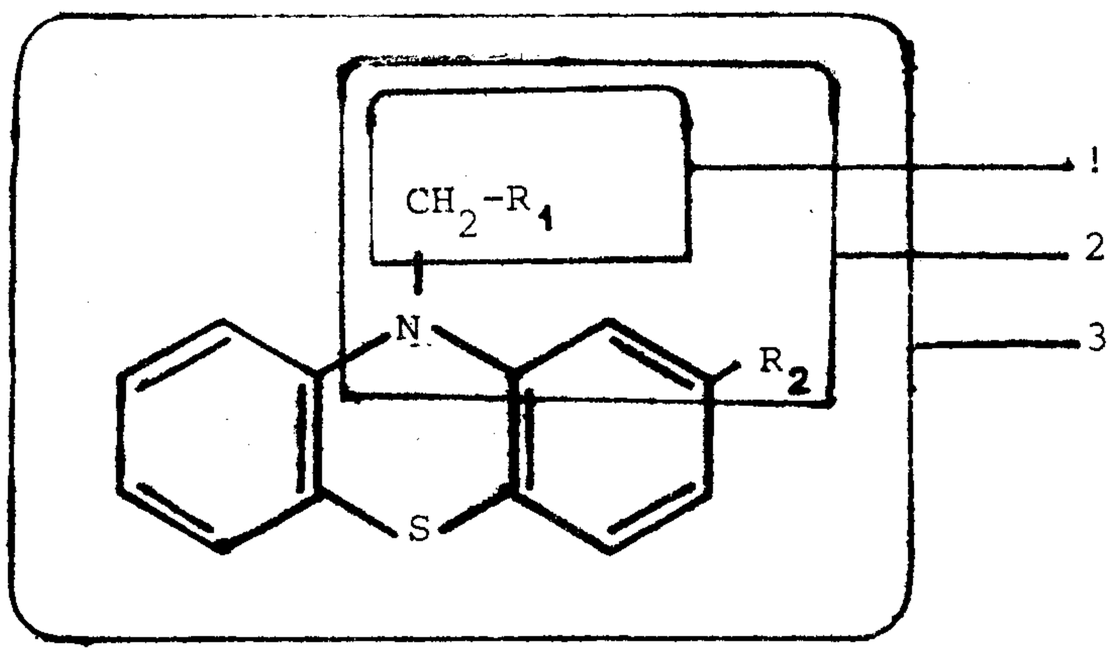


Scheme 2

Further confirmation of our suggestion about the role of C-2 substituent can be searched in the optimum value of σp that gives maximum color intensity. This study demands more probes with electron donating properties. For this reason compound I was excluded from the regression computation of eq. 1.

The second approach is to correlate log ξ_{max} with the molecular connectivity. The connectivity calculation used a set of indexes to describe molecular structure $^{n}\chi_{p}^{v}$. The superscript n is called the index order and is simply the number of bonds in the skeletal fragment[4], n=0,1,2,3..; v is the number of valence electrons of the given atom minus those involved in hydrogen bonds. The subscript p defines the cluster or the pathway by which the bonds were traced. Three clusters were calculated for $^{n}\chi^{v}$ values, however, the best corre-

lation was that found with cluster 1 including N-10 side-chain only (Scheme 3).



Scheme 3

Log ξ_{max} was correlated separately with $^{n}\chi^{v}_{1}$, where n=0,1,2 and 3 using simple linear regression analysis. where n=0,1, the χ index essentially represents the molecular volume of substituent at N-10. To account for branching effect, the higher order connectivity indexes n=2,3 were used.

The statistical analysis showed that $(^{1}\chi^{v}_{1})^{2}$ is the best variable to correlate with log ξ_{max} (eq. 2).

$$\log \xi_{max} = 4.827-0.0691(\pm 0.0585)\ (^{1}\chi^{v}_{1})^{2} \qquad \text{eq. 2}$$

$$r = 0.840 \quad s=0.522 \quad n= 6 \text{ (VI-XI)}$$

$$F_{(1,4)}=9.17 \qquad p<0.05$$

It is clear from eq. 2 that the negative effect displayed by the bulkiness of N-10 moiety represents 62% ($\tilde{r}$ = 0.62) of the explained variance of log ξ_{max} . It seems that other parameters not reflected by the χ index were significantly effective too. Such parameters like the conformation of the N-10 side chain may influence to different extents the phenazathionium ion formation XIV. Further, in the reaction medium, the abundance of the cleaved fragment XVI and the conjugated base of the acid used in salt formation of the drug (chloride, maleate or tartrate) may affect the color intensity too. In a computarised radiocrystallographic study, Reboul and Cristau[7] showed the folding of the central heterocyclic structure as well as the conformation variation of the N-10 substituents in different phenothiazines. They have reported that the conformation of the basic N-10 substituent was affected by salt formation and the type of acid used.

Working up the two variable second order regression, equation 3 revealed high quality and give better account of the correlation of log ξ_{max} than that represented by eq. 1 or 2

$$\log \xi_{max} = 5.044 - 6.697\ \sigma p - 0.073\ (^{1}\chi_{1}^{v})^{2} \qquad \text{eq. 3}$$

$$r = 0.950 \quad s = 0.583 \quad n = 12$$

$$F_{(2,9)} = 41.65 \quad , \quad p < 0.005$$

Equation 3 accounts for 90% ($\tilde{r}$ = 0.9025) of the variance in log ξ max of twelve compounds tested (II-VIII).

Application of the null hypothesis to test that there is no difference between average observed (μ_o) and average calculated (μ_c) of log ξ values i.e.

$$H_o : \mu_o = \mu_c$$

$$H_A : \mu_o \neq \mu_c$$

the test statistic computed from data (II-XIII), z = -0.0014, at the level of significance α = 0.05 where the tabulated value of z = $\pm$ 1.96 for the above two sided hypothesis. It is clear that we do not reject H_o and conclude that there is no significant difference between observed and calcuated log ξ values at the 0.05 level of significance or even higher than that.

From equation 3, it can be pointed out that both substituents C-2 and N-10 play a definite role in the color development. This role can take the form of electronic and steric involving the ring.

Development of the second order regression equation 3 is of special value in the prediction of the sensitivity and the limit of application of the reported colorimetric method for the assay of other phenothiazine derivatives.

Table 1: Phenothiazines in the study and their observed and calculated log ξ max from the appropriate equations.

No.	Name	R_1^+	R_2^x	Log ξ max			
				Obs.	Eq.1	Eq.2	Eq.3
I	Levomepromazine maleate	1	OCH_3	3.8921	----	----	----
II	Thiethylperazine maleate	2	SC_2H_5	3.4423	3.6923	----	3.6574
III	Prochlorperazine maleate	2	Cl	3.3501	2.2796	----	2.3382
IV	Butaperazine maleate	2	$COC_3H_7^{xx}$	0.0000	0.4664	----	0.6438
V	Trifluperazine hydrochloride	2	CF_3	0.0000	0.0363	----	0.2420
VI	Perazine maleate	2	H	3.5688	3.9070	3.7064	3.8584
VII	Phenothiazine base	H	H	4.7243	----	4.8271	5.0440
VIII	Alimemazine tartrate	1	H	4.0465	----	4.3588	4.5493
IX	Promazine hydrochloride	3	H	4.4314	----	4.4896	4.6874
X	Promethazine hydrochloride	4	H	4.8484	----	4.5115	4.7106
XI	Mepazine hydrochloride	5	H	4.3581	----	4.0747	4.2490
XII	Colorpromazine hydrochloride	3	Cl	4.0669	----	-----	3.1672
XIII	Thioridazine hydrochloride	6	SCH_3	4.2679	----	-----	3.9687

\+ The number in column refers to the side chain scheme 1.

x σp of substituents have been taken from reference 3.

xx σp of COC_2H_5 has been taken in correlation.

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التحليل الطيفى لمركبات الفينوثيازين

٣ ـ مدى العلاقة بين قيمة لوغاريتم شدة الامتصاص الجزيئى وبعض المعايير الفيزيوكيميائية للمجموعات المستبدلة فى حلقة الفينوثيازين

سلوى رزق الشابورى ـ عادل فوزى يوسف ـ فردوس عبد الفتاح محمد ـ عبد المعبود اسماعيل

قسم الكيمياء الصيدلية ـ كلية الصيدلة ـ جامعة اسيوط

فى هذا البحث تم ربط شدة اللون من تفاعل اثنا عشر عقارا من عقارات الفينوثيازين مع ثلاثى اليوديد ببعض المعايير الفيزيو كيميائية للمجموعات المستبدلة فى حلقة الفينوثيازين وهى المعيار الالكترونى سيجما σ (ثابت هاميت) وعامل الترابط الجزيئى (كاى χ) والنتائج توضح دورا فعالا للثابت الالكترونى سيجما فى تحديد شدة اللون .

كما ثبت ان حجم المجموعة الفرعية ممثلا بالثابت كاى يلعب دورا هاما ايضا وقد تم استنباط معادلة انحدار متعددة المجاهيل وتم استخدامها بنجاح فى حساب مدى حساسية الطريقة ومجال تطبيقها فى التحليل الطيفى لبعض مركبات الفينوثيازين.

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