SPECTROSCOPY LETTERS, 29(4), 727-738 (1996)

SPECTROPHOTOMETRIC STUDY OF BASICITY OF SOME SUBSTITUTED α-(BENZAMIDOMETHYL)PHENYLHYDRAZINES

Key words: α-(benzamidomethyl)phenylhydrazine, UV spectrophotometry, protonation constants

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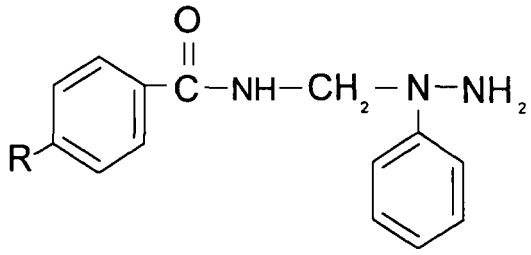
ABSTRACT

The protonation behavior at 25°C in water and in aqueous sulfuric acid solutions of some substituted α-(*p*-*R*-benzamidomethyl)phenyl hydrazines (R=H, CH_3, Cl, Br) was investigated by UV spectrophotometry. The absorption spectra in sulfuric acid solutions were analyzed by the method of multivariate analysis. The protonation constants of hydrazine (pK_1) and amide (pK_2) moieties were determined; pK_2 values were calculated from the reconstituted absorption spectra by Cox - Yates method. Dependence of pK_2 on Hammett's σ constants was discussed.

INTRODUCTION

The hydrazine derivatives belong to a large group of compounds with a potential biological activity. A large number of them are used as

antituberculostatics, antiinflammatics and antidepressives[1-3]. The new group of compounds, α-(*p*-*R*-benzamidomethyl)phenylhydrazines (R=CH_3, H, Cl, Br), was synthesized recently[4]. These compounds have some functional groups that could potentially act as bases. Acording to the literature data, the protonation of -NH_2 group, amide moiety and phenyl substituted hydrazine nitrogen could be expected in water, from neutral media to strongly acidic H_2SO_4 solutions[5-10]. The basicity is very important parameter for understanding the behavior of biochemical systems, so this paper deals with the study of the acid - base properties of the related compounds. The aim of the work was the investigation of the substituent effect on the basic properties of the amide and hydrazine moieties. The substituted α-(*p*-*R*-benzamidomethyl)-phenylhydrazines investigated here are represented by the structural formula:



1 R=H **3** R=Cl

2 R=CH_3 **4** R=Br

EXPERIMENTAL SECTION

Materials

α-(*p*-*R*-benzamidomethyl)phenylhydrazines **1** - **4** were prepared as reported[4]. Stock substrate solutions (concentration of about $1x10^{-3}$ M to $1x10^{-4}$ M) were prepared in 20% aqueous ethanol. The concentration of the substrates in finding pK values was about $5x10^{-5}$ M.

pK Measurements

All measurements for determination of pK values were taken as rapidly as possible after the acid addition. The hydrolysis of the compounds was negligible during the period required to record spectra. The absorption spectra in 3 and 15.6 M H_2SO_4 were repeated after 1 hour's standing and no significant changes were found. On the subsequent dilution the spectra also reproduced closely those of control solution in dilute acid. The absorption curves in concentrated sulfuric acid solutions were reconstituted by the method of multivariate analysis[11], using the original computer program. Absorbance measurements at 5 nm intervals over the wavelength range 210 - 300 nm were taken from at least 15 spectra covering the acidity range from pH 0 to 96 % H_2SO_4.

The concentration of sulfuric acid (Merck, p.a.) was determined from the density measured at 25°C by precise density meter[12]. Acid solutions of various acid concentrations were made up by diluting concentrated sulfuric acid up to the required acidity.

Spectral and pH Measurements

UV spectra were measured in 1 cm matched, quartz cuvette with the cell holder thermostated at 25°C. The spectra were recorded in acidity range from pH 8 to 96% H_2SO_4 aqueous solutions. The reference contained the same quantity of ethanol as the measured solution. The ionic strength was kept constant (0.01 M) by addition of $NaClO_4$.

RESULTS AND DISCUSSION

Absorption Spectra

The absorption spectra of the compounds **1 - 4** (concentration 4 - $5x10^{-5}$ M) were recorded in water (pH 0 -8) and H_2SO_4 aqueous solutions (concentration from 1 to 18.10 M).

The absorption spectra of all compounds have in neutral aqueous solutions (pH 5 - 7) the absorption maximum in the range from 220 - 240 nm. With the increase in acidity the absorption spectra showed from pH 3 to 5 the changes that

were small but measurable. The hypsochromic shifts of the absorption maximum for 1 to 4 nm was followed by the appearance of the clear isobestic point and hypochromic effect. These spectral changes were indicative for the protonation process on $-NH_2$ moiety[6-8].

The increase in acidity from pH 0 to 18 M H_2SO_4 shifted the absorption maximum from 220 - 240 nm to 250 - 270 nm. The experimental absorption spectra of α-(*p*-CH_3-benzamidomethyl)-phenylhydrazine (concentration of 4.17×10^{-5} M) in H_2SO_4 solutions from 1 to 15.6 M are presented on FIG. 1*a*. Spectral changes were followed by the appearance of the poor isobestic point. The similar behavior was found by all investigated compounds. Previous UV studies of some substituted benzamides showed that two processes were taking place over this acidity range[13-15]. One is the protonation of carbonyl group and the other is a pronounced medium effect on the acid or base form of the molecule.

The plot of absorbancies measured at several wavelengths against pH or H_0 was sigmoid curve, and confirmed the formation of BH^+ and BH_2^{2+} species. Table 1 shows the pH and acid concentrations used for the plateau of experimental absorbance vs. acidity curves, with the wavelengths of maximum absorption of the corresponding ionic species.

Calculation of the Protonation Constants

The values of pK_1 of monoprotonated bases were calculated from the dependence of the absorbance vs. pH curves at several wavelengths according the known spectrophotometric method[16]. For each compound the value of pK_1 was obtained from a plot of

$$\log\frac{[BH^+]}{[B]} = \log I = npH + pK_1 \qquad (1)$$

The protolytic process in pH acidity range was separated by more than three pH units from the second one. The first protonation constant was determined independently, as for monoprotic bases. The results are summarized in Table 2. Protonation on the hydrazine $-NH_2$ was totally unaffected by R group changes in

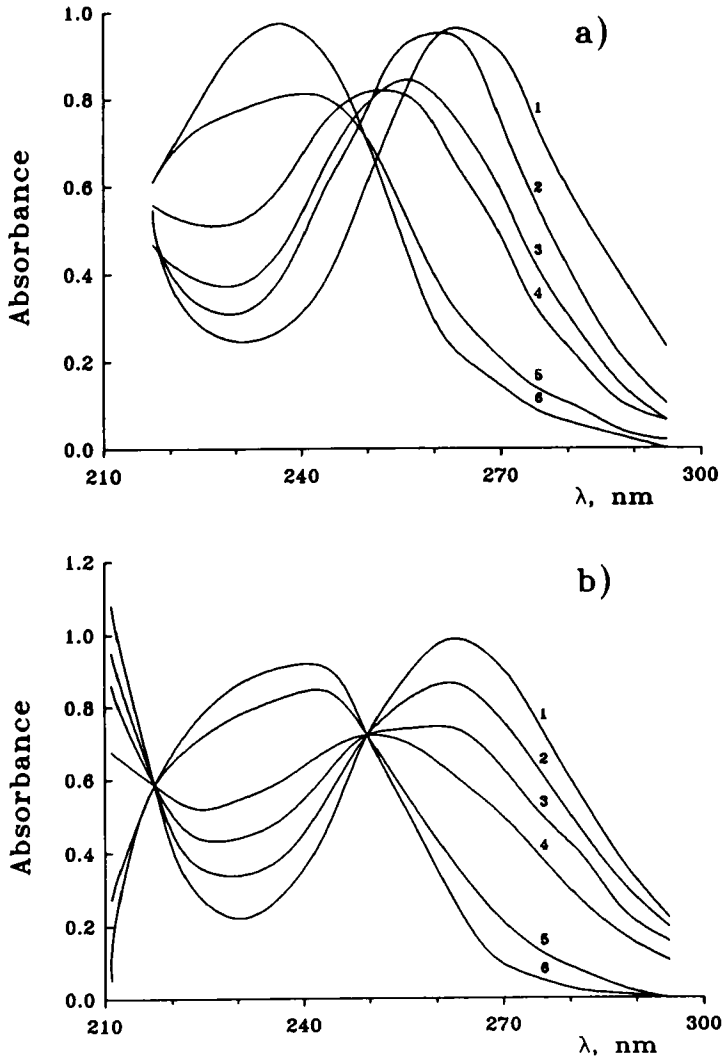


FIG. 1. Absorption spectra of α-(p-CH_3-benzamidomethyl)phenylhydrazine as a function of H_2SO_4 concentration: 1 - 15.6 M; 2 - 10.0 M; 3 - 9.1 M; 4- 8.0 M; 5 - 5.0 M; 6 - 0.8 M. a) experimental data; b) calculated by CVA.

TABLE 1

Ultraviolet Spectral Data of α-(p-R-Benzamidomethyl)Phenylhidrazines

R[a]		acidity	λ_{max} nm	$10^{-4}\epsilon_{max}$ $dm^3mol^{-1}cm^{-1}$
H	B	pH 5 - 7	228	2.10
	BH^+	1.25 M H_2SO_4	227	1.80
	BH_2^{2+}	14.0 M H_2SO_4	250	1.66
CH_3	B	pH 5 - 7	240	1.97
	BH^+	pH 0	236	1.66
	BH_2^{2+}	13.5 M H_2SO_4	263	1.66
Cl	B	pH 5 - 7	234	2.20
	BH^+	pH 1.33	232	1.70
	BH_2^{2+}	14.1 M H_2SO_4	257	1.92
Br	B	pH 5 - 7	242	1.90
	BH^+	pH 1.23	240	1.60
	BH_2^{2+}	16.7 M H_2SO_4	267	2.20

[a] B - neutral form; BH^+ and BH_2^{2+} - monoprotonated and diprotonated forms

TABLE 2

Protonation Constants of α-(p-R-Benzamidomethyl)Phenylhydrazines at 25°C

R	pK_1[a]	$-pK_2$[b]	m*
H	4.28±0.16	2.95±0.02	0.66±0.13
CH_3	4.12±0.02	2.56±0.04	0.57±0.02
Cl	4.29±0.05	3.18±0.14	0.58±0.04
Br	3.85±0.15	3.21±0.08	0.59±0.03
ρ[c]		-1.60	
r[c]		0.980	

[a] I=0.01 M ($NaClO_4$); [b] Calculated from reconstituted spectral curves; [c] Parametars of Hammett's plot.

the other ring, which is a long way away. The pK_1 values are almost constant, showing that there are no proximity interactions between benzamide part and the reaction center. The investigated compounds are weaker bases than phenylhydrazine itself[6]. The substitution of $PhCONH_2CH_2$- group for hydrogen in the phenylhydrazine lowered the base strength for approximately one pK unit.

pK Values in Concentrated H_2SO_4 Solutions

For each compound the values of pK_2 were calculated using the Cox-Yates approach[17] (EAFM):

$$\log I - \log c_{H^+} = m*X + pK_2 \qquad (2)$$

$$I=\frac{[BH_2^{2+}]}{[BH^+]}=\frac{A - A_{BH^+}}{A_{BH_2^{2+}} - A} \qquad (3)$$

$[BH^{2+}]$ and $[BH^+]$ are the concentrations of diprotonated and monoprotonated base. X represents Cox - Yates "excess acidity" function, and log c_{H+} proton concentration[17].

The slopes of ionization curves by compounds **1 - 4**, were dependent on wavelength, and the method of multivariate analysis[11,13] was considered to separate the effect of protonation from the generalized medium effect. The absorbance (A) at wavelength λ could be mathematically described by the eq. 1:

$$A_\lambda = \overline{A_\lambda} + c_1 v_1 + c_2 v_2 \qquad (4)$$

where A_λ is absorbance at wavelength λ, $\overline{A_\lambda}$ is the average A at that wavelength over the acidity range, *v*'s are vectors, independent of acidity, and *c*'s are weighthing coefficients, independent of wavelength. The first vector (v_1) is associated with the effect of protonation and the second vector (v_2) with the medium effect. Although the method of multivariate analysis does not recognize

the effects of medium changes on the conjugate acid and on the free base as independent factors, this treatment was sufficient to account for the small medium effect that was found by the investigated compounds. FIG. 2*a* shows the coefficients of two characteristic vectors, c_1 and c_2, plotted as the dependence of the Hammett acidity function for primary aromatic amines, H_o[18]. In the case of all investigated compounds the protonation behavior as a function of acidity was implied in the resulting coefficients of the first vector, c_1; c_2 had no obvious physical or chemical meaning[15]. FIG. 2*b* shows the dependence of characteristic vectors, v_1 and v_2, as the function of wavelength. The most suitable wavelengths for the calculation of pK values were determined from v_i vs. λ dependencies for all investigated compounds[13].

Application of the method of multivariate analysis resulted in very similar behavior for all investigated compounds. The first vector was accounting for 94-96%, and the sum of the first and the second vectors for > 99,5% of the total variance. FIG. 1*b* shows the absorption spectra calculated by the method of multivariate analysis. The spectra, reconstituted using the calculated vectors and coefficients, pass through well-defined isobestic points and are in good agreement with the originals. The root - square residual error is about ± 0.010 absorbance units.

The ionization ratios obtained from the reconstituted absorption spectra were independent of the wavelength. The same values were also obtained directly from the coefficients of the first characteristic vector, c_1. Table 2 gives the pK_2 values of α-(*p*-*R*-benzamidomethyl)- phenylhydrazine **1 - 4** calculated by eq. 2.

Substituent Effect

The plot of pK_2 at 25° C against Hammett σ constants is linear, and its parametars are listed in Table 2. Although the substituent range is very small (two points fall approximately in the same range), a good linear correlation is obtained (r=0.980). The value of ρ by the method of least square is -1.60, which is very close to that found by a series of substituted benzamides (ρ=-1.30)[5]. It is the

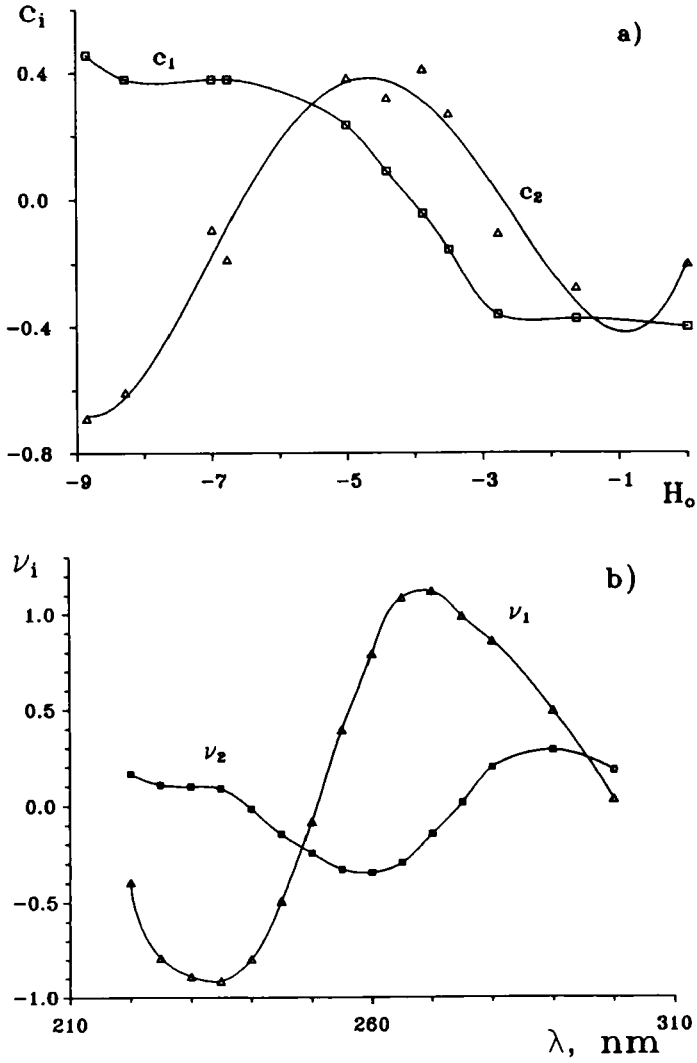


FIG. 2. a) Coefficients of the characteristic vectors of α-(p-$_C$-benzamidomethyl)-phenylhydrazine (c_i) vs. Hammett acidity function, H_o; b) characteristic vectors (v_i) as a function of wavelength.

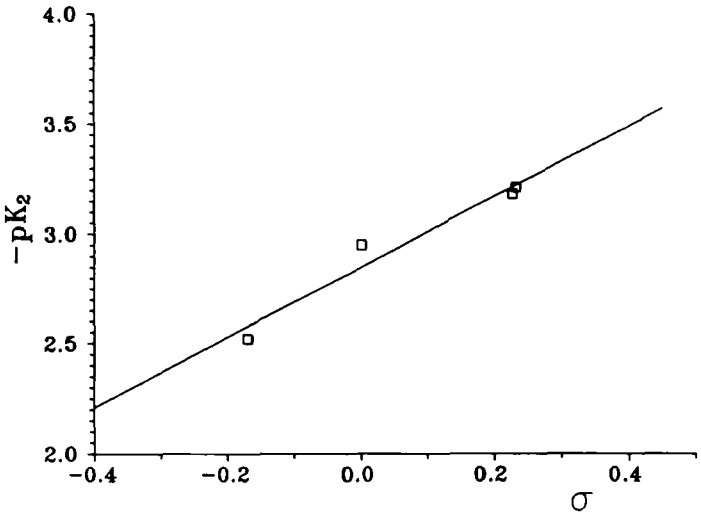


FIG. 3. Plot of pK_2 at 25° C calculated by the HAFM against Hammett's σ values.

indication that the electronic interactions from the protonated $-NH_2$ to the reaction center does not exist, because the reaction center is a long way away.

The basicity of amide group in α-(*p*-*R*-benzamidomethyl)phenylhydrazines is lower than in the other benzamides, widely investigated by many authors (pK about -1.5)[7,19]. The basic strength has been weakened, because the protonated hydrazine $-NH_3^+$ group behaves as an electronic withdrawing substituent, including also the repulsive columbic effect of the protonated amine nitrogen.

Since only a few data were available in the high acidity region, no qualitative treatment of the third protonation was attempt. The subsequent process in strongly acidic solutions seems to be the protonation of phenylhydrazine nitrogen, rather than the tautomeric equilibria between N- and O-protonated forms[20]. This statement agrees well with the literature data. They show that the first and the second protonation stage in the series of ring substituted phenylhydrazine are separated by ten H_o units[6].

Our investigations showed that monoprotonation of the investigated hydrazine derivatives occurs in the pH range from 3 to 5, as also found by some acyl and benzoylhydrazines[7-9]. The basicity of the investigated compounds is lower for about one pK unit, compared with ring substituted phenylhydrazines[6]. Decreasing in pK values with investigated compounds could be explained by the effect of the amide part of molecules.

ACKNOWLEDGMENTS

The financial support of Scientific Fund of Serbia (Yugoslavia) is gratefully acknowledged. The authors thank to P.Jelić for the help in experimental work.

REFERENCES

1. Grekov A.P., Veselov V.J. *Fizicheskaja himia gidrazina,* Kiev, 1979.
2. Myadera T. The Chemistry of the hydrazo, azo and azoxy groups. In: Patai S. ed. *The Chemistry of Functional Groups.* London: 1975. Part 1, Chapter 13, p.495.
3. Kruger P.J. The Chemistry of the hydrazo, azo and azoxy groups. In: Patai S. ed. *The Chemistry of Functional Groups.* London: 1975. Part 1, Chapter 7, p.153.
4. Lazarević M., Popovska Lj., Klisarova L., Ivanovski T. Bull.Chem. Technol. Macedonia 1992; 11: 23.
5. Edward J.T., Chang H.S., Yates K., Stewart R. Can.J.Chem. 1960; 38: 1518.
6. Brignell P.J., Johnson C.D., Karitzky A.R., Shakir N., Tarhan H.O., Walker G. J.Chem.Soc.(B) 1967; 1235.
7. Mashima M., Ikeda F. Bull.Chem.Soc.Jpn. 1973; 46: 1366.
8. Mashima M., Ikeda F., Doi T., Nishikawa S. *ibid.* 1981; 54: 3659.
9. Zarakhani N.G., Borisova E.N., Nechaev P.P., Yaikov G.E. Izv.Akad.Nauk SSSR, Ser.Khim. 1981; 2476.
10. Cox R.A., Yates K. Can.J.Chem. 1984; 62: 1613.
11. Edward J.T., Wong S.C. J.Amer.Chem.Soc. 1977; 99: 4229.
12. Lurie Ju. *Handbook of Analytical Chemistry*, Moscow: Mir, 1978.
13. Zalewski R.J. J.Chem.Soc.Perkin Trans. 2 1979; 1637.
14. Edward J.T., Wong S.C. Can.J.Chem. 1977; 55: 2492.
15. Cox R., Yates K. Can.J.Chem. 1981; 59: 1560.
16. Asuero A.G., Navas M.J., Jimenez-Trillo J.L. Talanta 1986; 33: 195.
17. Cox R.A., Yates K. J.Amer.Chem.Soc. 1978; 100: 3861.

18. Rochester C.H. *Acidity Functions,* London: Academic Press, 1970.
19. Cox R.A., Druelt L.M., Klausner A.E., Modro T.A., Wan P., Yates K. Can.J.Chem. 1981; 59: 1568.
20. Liler, M. J.Chem.Soc.Perkin Trans. 2 1974; 71.

Received: November 27, 1995
Accepted: January 11, 1996