Salicylidene-2-iminobenzohydroxamic acid and its analytical application

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This paper deals with preparation and analytical properties of the new agent — the salicylidene-2-iminobenzohydroxamic acid. The dissociation constants were determined and the mechanism of complex formation with Fe(III) of the new agent was studied. Further, the possibility of use of this agent for the proof of cations and as a chelometric indicator for Fe(III) was investigated.

В работе описываются получение и аналитические свойства нового реагента — салицилиден-2-иминобензогидроксамовой кислоты. Были определены ее константы диссоциации и изучен механизм комплексообразования с Fe(III). Исследована также возможность применения этого реагента как индикатора присутствия катионов и в качестве хелатометрического индикатора на Fe(III).

In the previous paper [1] the preparation of salicylidene-2-iminodiacetohydroxamic acid (SIAH) and some of its complexes was described. Its certain modification is the salicylidene-2-iminobenzohydroxamic acid (SIBH), which is the object of present study.
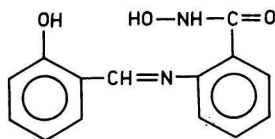
SIBH

A common feature of both agents is the presence of azomethine, hydroxamic, and phenolic group in their molecule. Until now, there were prepared and studied the tens of polydonor analytical agents possessing azomethine and hydroxamic groups [2—4]. With the exception of our previous paper [1], however, none of them involves the mutual combination of azomethine and hydroxamic group.

In this paper the preparation of SIBH is described as well as the study of its acid-base properties together with the study of the complex formation with Fe(III).

Experimental

All the agents used were of anal. grade purity. The elemental analysis was performed with C. Erba 1102 analyzer. Infrared spectra were measured with the Perkin—Elmer 377 spectrophotometer using KBr pellets technique, and electronic absorption spectra with the Unicam SP-700 one. For the pH measurements the PHM 25 (Radiometer, Copenhagen) and combined electrode GK 2301C were used.

Salicylidene-2-iminobenzohydroxamic acid (SIBH)

This agent was prepared by the reaction of stoichiometric amount of anthranilehydroxamic acid with salicylaldehyde in methanol. Anthranilehydroxamic acid was prepared from the 2-aminobenzoic acid according to *Scott* and *Wood* [5].

To the suspension of anthranilehydroxamic acid (15.2 g; 100 mmol) in methanol (100 cm³) salicylaldehyde (12.2 g; 100 mmol) being freshly distilled, in methanol (20 cm³) was dropwise added. After the aldehyde addition, the reaction was left to proceed for 4 h at ambient temperature. The raw product was yielded by evaporation of the reaction mixture at reduced pressure. The SIBH agent was recrystallized from water—methanol ($w = 50\%$) medium. It is poorly soluble in water, and very good in methanol, ethanol, and dimethyl sulfoxide. The yield of SIBH was 12.8 g, *i.e.* 80%, m.p. = 183 °C.

For $C_{14}H_{12}N_2O_3$ ($M_r = 256.3$) w_i(calc.): 65.62% C, 4.72% H, 10.39% N; w_i(found): 65.67% C, 4.74% H, 10.43% N.

Determination of dissociation constants

The dissociation constants pK_{a1} and pK_{a2} of SIBH were determined spectrophotometrically by measuring the absorption of agent under study as a dependence on pH, in 50 mass % methanol in the range of $\lambda = 250$—333 nm. The acidity of solution was maintained and measured simultaneously with the absorbance in 100 cm³ quartz cells of 35 mm absorption thickness [6]. The calibration of electrode was performed with acetate and phosphate buffer, respectively, in 50 mass % methanol with NaCl as a background electrolyte [7]. The pK_{a0} value for the protonated agent $[H_3L^+]$ was determined potentiometrically by titrating 50 cm³ of solution (c(SIBH) $= 5 \times 10^{-3}$ mol dm⁻³ in 50 mass % methanol) with the solution of c(HCl) $= 1 \times 10^{-3}$ mol dm⁻³. The pK_{a1} was determined by the same method by titrating the solution (c(SIBH) $= 1 \times 10^{-3}$ mol dm⁻³ in 50 mass % methanol) with the solution of c(NaOH) $= 0.1$ mol dm⁻³. The electrodes were calibrated by the buffers as used in spectrophotometric measurements.

Qualitative test

The formation of coloured complexes of the agent under study with Cu(II), Ni(II), Co(II), Fe(III), V(V), U(VI), W(VI), Mo(VI), and Zr(IV) was studied in the pH range from 3 to 11

units ($c_L = 5 \times 10^{-3}$ mol dm^{-3}, $c_M = 2 \times 10^{-3}$ mol dm^{-3}, 50 mass % methanol). The changes in colour were examined visually.

Spectrophotometric study of the Fe(III)—SIBH complex formation

This was studied by the absorption curve method in the range of pH = 0.5—12. Solutions used were of $c_M = 5 \times 10^{-5}$ mol dm^{-3} and $c_L = 5 \times 10^{-4}$ mol dm^{-3} in 50 mass % methanol, measured in the spectral range of $\lambda = 370$—770 nm. The composition of complexes was evaluated by the Job's method and by the mole ratios one.

Chelometric determination of Fe(III) using SIBH as an indicator

The titration was as follows: to the titration flask a 10 cm^3 portion of ($c(Fe(ClO_4)_3) = 0.01$ mol dm^{-3}) solution was pipetted, to which 40 cm^3 of water and 1 cm^3 of $w(SIBH) = 1$ mass % methanolic solution was added. Fe(ClO$_4$)$_3$ solution was gravimetrically standardized with NH$_4$OH as a precipitant. After the pH adjustment with $c(HClO_4) = 0.5$ mol dm^{-3} the content of Fe(III) was titrated by c(Chelaton III) $= 1 \times 10^{-2}$ mol dm^{-3} solution. The violet colour of titrated solution is turned to pale yellow at the end point.

Results and discussion

The salicylidene-2-iminobenzohydroxamic acid was prepared by the addition--elimination reaction of 2-aminobenzohydroxamic acid with salicylaldehyde in methanolic medium. For verification of the agent constitution the elemental analysis and IR spectroscopy were used.

In the case of IR spectrum the attention was paid to several bands, which might contribute to the verification of the compound constitution. The vibration of the N—H fragment of —CONHOH was found at $\tilde{v} = 3410$ cm^{-1} and 3230 cm^{-1}, respectively, in agreement with Ref. [8]. In both cases the bands are of moderately strong intensities. The most intensive band at $\tilde{v} = 1638$ cm^{-1} is characteristic of the stretching vibration of C = O (amide *I*). The C = N stretching vibration is observed as a shoulder of the former band at $\tilde{v} = 1653$ cm^{-1}. The band at $\tilde{v} = 1495$ cm^{-1} is characteristic of bending vibration of N—H (amide *II*). The amide *III* band was observed at $\tilde{v} = 1275$ cm^{-1}. Absorption of the C—O of phenolic group was observed at $\tilde{v} = 1232$ cm^{-1}.

The dissociation constants pK_{a1} and pK_{a2} were determined by the evaluation of absorption curves of SIBH from spectrophotometric measurements in the UV range. Agent in the range of $\lambda = 200$—333 nm reveals several maxima of absorption, from which the main one is positioned at $\lambda = 224$ nm. For the dissociation constant evaluation, the range of $\lambda = 263$—333 nm was selected with the absorp-

tion maximum at $\lambda = 296$ nm possessing the most pronounced difference in absorption of individual forms of the agent under study (H_2L, HL^-, and L^{2-}). The graphical construction of $A = f(pH)$ dependence (Fig. 1) shows the three different forms of this agent. By the logarithmic analysis [9] it was unambiguously proved that in the range of pH = 8.0—9.0 and pH = 10.9—11.8, respectively, the dissociation of one proton always takes place. The dissociation constants as determined from the $A = f(pH)$ dependence are listed in Table 1 (an average from five measurements). The molar absorption coefficients of the H_2L and L^{2-} species were calculated from horizontal parts of the $A = f(pH)$ dependence curves. The HL^- molar absorption coefficient was found by the graphical analysis using relationships described in our previous paper [1].
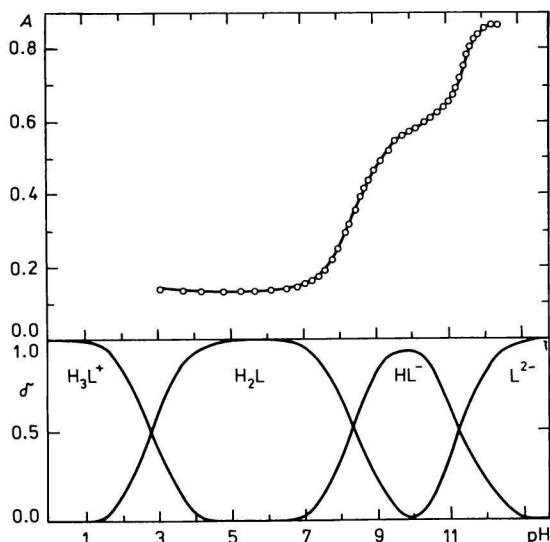

Fig. 1. The $A = f(pH)$ dependence of SIBH and the distribution diagram.
$c_L = 3.270 \times 10^{-5}$ mol dm^{-3}; $\lambda = 296$ nm; $l = 35$ mm; $I_c(NaCl) = 0.1$ mol dm^{-3}; water—methanol ($w = 0.5$).

Except the use of spectrophotometric method the dissociation constants were also determined by the potentiometric measurements (pK_{a1}, pK_{a0} values) using well known calculations. These results are listed in Table 1 as the average values of five measurements. We suppose that pK_{a0} is connected with deprotonation of the —CH=$\overset{+}{N}$H— azomethine group, pK_{a1} and pK_{a2} with deprotonation of the (—CONHOH) and phenolic ones, respectively.

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Table 1

Dissociation constants of SIBH
$I_c(NaCl) = 0.1$ mol dm^{-3}; $\theta = 20$ °C; water—methanol ($w = 0.5$)

Equilibrium	Constant	$-\log K$	
[H₂L][H]/[H₃L]	K_{a0}	2.8 ± 0.1	(p)
[HL][H]/[H₂L]	K_{a1}	8.35 ± 0.04	(p)
		8.44 ± 0.05	(s)
[L][H]/[HL]	K_{a2}	11.36 ± 0.08	(s)

(p) — potentiometrically, (s) — spectrophotometrically.

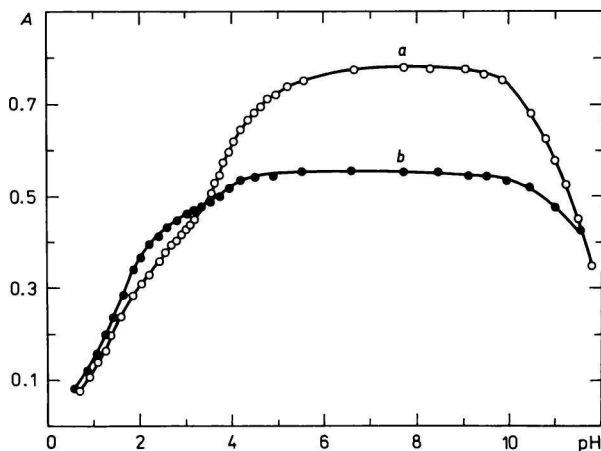*Fig. 2.* The $A = f(pH)$ dependence for the Fe(III)—SIBH system.
$c_M = 5 \times 10^{-5}$ mol dm^{-3}; $c_L = 5 \times 10^{-4}$ mol dm^{-3}; $l = 35$ mm; $I_c(NaCl) = 0.1$ mol dm^{-3},
water—methanol ($w = 0.5$).
a) $\lambda = 477$ nm; *b)* $\lambda = 538$ nm.

In spectrophotometric investigation of the complex equilibria Fe(III)—SIBH the series of solutions containing a tenfold excess of agent with respect to Fe(III) was used. From the $A = f(pH)$ dependence curves at wavelengths of the absorption maxima of complexes formed (Fig. 2) one may conclude that in the range of pH = 0.5—3.3 proceeds the formation of two violetly coloured reaction products, which are not separated by the evident horizontal part. The next red-brown coloured complex is formed at pH = 3.7—5.0. It was proved that in alkaline medium the hydroxo complexes are formed.

The first part of $A = f(pH)$ dependence in the range of $pH = 0.72$—1.76 was evaluated by the graphical and logarithmic analyses, whereas the relationships for calculations were taken from Ref. [9]. Performing the calculations, it was taken into account that agent in this range of acidity is in the H_3L^+ form (Fig. 1). The obtained results lead to conclusion that the complex formation is connected with the dissociation of one proton. Molar absorption coefficients of this complex and other ones formed in the reaction of SIBH with Fe(III), as gained by graphical analysis, are listed in Table 2. The composition of complex was determined by Job's method (Fig. 3, curve a). Basing on the results obtained it is possible to describe the complex formation by the following scheme
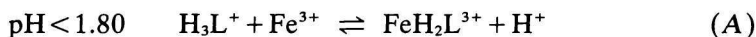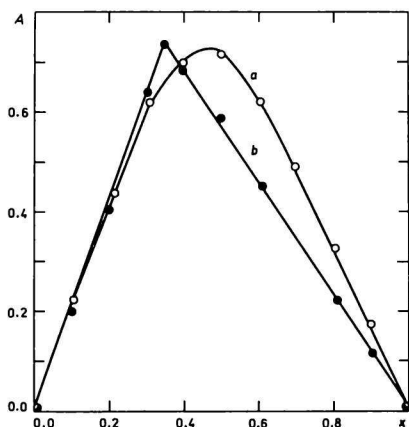
$$pH < 1.80 \qquad H_3L^+ + Fe^{3+} \; \rightleftharpoons \; FeH_2L^{3+} + H^+ \qquad (A)$$

Table 2

Molar absorption coefficients of the Fe(III)—SIBH complexes

Complex	λ/nm	$\varepsilon/l\ mol^{-1}\ cm^{-1}$
FeH_2L^{3+}	477	2040
	510	2280
	538	2320
$Fe(HL)_2^+$	477	2720
	510	2880
	538	2700
$Fe(HL)_3$	477	4360
	510	4030
$FeOH(HL)_3^-$	477	1960

Fig. 3. The Job's curves for the Fe(III)—SIBH system.
$x = c_M/(c_M + c_L)$; $c_0 = c_M + c_L$.
a) $c_0 = 3 \times 10^{-4}\ mol\ dm^{-3}$; $l = 20\ mm$;
$\lambda = 538\ nm$; $pH = 1.80$.
b) $c_0 = 2 \times 10^{-4}\ mol\ dm^{-3}$; $l = 20\ mm$;
$\lambda = 510\ nm$; $pH = 3.20$.

The equilibrium constant and the stability one for a given complex and others, which are formed by SIBH and Fe(III), are listed in Table 3.

Further, a complex formed in the range of pH = 2.3—3.3 was analyzed. By the logarithmic analysis the dissociation of two protons was proved, and by Job's method the composition of complex $n(M):n(L) = 1:2$ (Fig. 3, curve b). The complex formation may be described as follows

$$pH = 2.3\text{---}3.3 \qquad FeH_2L^{3+} + H_2L \; \rightleftharpoons \; Fe(HL)_2^+ + 2H^+ \qquad (B)$$

By analysis of the $A = f(pH)$ function of the next complex it was proved that in the range of pH = 3.7—4.8 one proton is split off. The composition of complex formed was evaluated by the mole ratios method, and it is $n(M):n(L) = 1:3$. Regarding this, the complex formation may be expressed as follows

$$pH = 3.7\text{---}4.8 \qquad Fe(HL)_2^+ + H_2L \; \rightleftharpoons \; Fe(HL)_3 + H^+ \qquad (C)$$
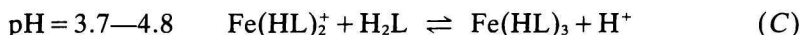
In the range of pH = 9.0—12.0 the absorbance and its maximum is gradually reduced and at $\lambda = 477$ nm the latter disappears. In this pH range the dissociation of one proton was proved from the results of logarithmic analysis. In this case the equilibrium may be described as follows

$$pH = 9.0\text{---}12.0 \qquad Fe(HL)_3 + H_2O \; \rightleftharpoons \; FeOH(HL)_3^- + H^+ \qquad (D)$$

The colour reactions of SIBH with selected cations were investigated in the further part of study. In all cases, there proceeds the formation of coloured complexes, whereas in some cases the complexes formed are poorly soluble (for instance those of Cu(II), Ni(II), Co(II)). The most intensive colour is observable in the case of iron and vanadium complexes. The determination of boundary sensitivity gave these values: for Fe(III) pD = 5.6 (pH = 2—6), for vanadium(V) pD = 5.7 (pH = 2—5). In the cases of other cations the pD value is in the range of 3.5—4.5.

Table 3

Values of the Fe(III)—SIBH stability constants
$I_c(NaCl) = 0.1$ mol dm^{-3}; $\theta = 20$ °C; water—methanol ($w = 0.5$)

Equilibrium	Constant	log of numerical value of constant
$[FeH_2L][H]/[Fe][H_3L]$	k_{1H}	2.07
$[FeH_2L]/[Fe][H_2L]$	β_{1H}	4.87
$[Fe(HL)_2][H]^2/[FeH_2L][H_2L]$	k_{22}	-2.46
$[Fe(HL)_2]/[Fe][HL]^2$	β_2	13.56
$[Fe(HL)_3][H]/[Fe(HL)_2][H_2L]$	k_{31}	-0.97
$[Fe(HL)_3]/[Fe][HL]^3$	β_3	20.94
$[FeOH(HL)_3][H]/[Fe(HL)_3]$	k_{1OH}	-11.7

The use of SIBH as a metallochromic indicator for chelometric determination of Fe(III) was studied more thoroughly. Looking for a suitable pH for the titration it was found that determination is possible in the range of $pH = 1.0—1.8$. With increase in the acidity of medium near to the $pH = 1.0$ the standard deviation became higher due to the reduced sensitivity of indication. The most suitable acidity for titration is about $pH = 1.5$, at which the interfering influences of other cations present are minimal. The determination is suitable for 2—10 mg Fe(III) using the solution $c(\text{Chelaton III}) = 1 \times 10^{-2}$ mol dm^{-3} as a titrant at 20—30 °C. It is also possible to titrate the probes with higher content of Fe(III), however, the standard deviation becomes higher (Table 4). The interval of reliability was calculated for the significance level $1 - \alpha = 0.95$ [10]. It was found that results of the analysis are correct and within the interval of reliability.

Table 4

Effect of the amount m of Fe(III) upon the determination
$pH = 1.5$; $\theta = 20$ °C; $n = 8$

$\dfrac{m(\text{calc.})}{\text{mg}}$	$\dfrac{m(\text{found})}{\text{mg}}$	$\dfrac{m(\text{found})/m(\text{calc.})}{\%}$	s/mg
2.635	2.625	99.6	0.39
5.270	5.259	99.8	0.33
7.905	7.886	99.7	0.44
10.540	10.474	99.4	0.41

By investigating the influence of cations and anions upon the Fe(III) titration results it was found that usual anions (Cl^-, ClO_4^-, NO_3^-, SO_4^{2-}, CH_3COO^-, and $(COO)_2^{2-}$) do not interfere the determination even in fifty-fold excess. Basing on the results from study of the cation interference it was found that alkaline metals and NH_4^+ ion do not affect the titration even in fifty-fold excess. The tenfold excess of Ca(II), Ba(II), Cd(II), Zn(II), Pb(II), Mn(II), and Cr(III) has no effect, either. In the case of Hg(II), Al(III), Co(II) the determination of Fe(III) is possible, if cations as mentioned do not exceed twofold excess. In the presence of Cu(II), Ni(II), and Bi(III) cations the correct results may be obtained, if content of these cations is lower than a half of that of Fe(III) being determined.

The chelometric determination of Fe(III) using SIBH as an indicator was successfully applied to the analysis of drugs containing iron: Ce-ferro drops, Ferronat dragee, Erypher dragee, and Conferon capsules [11].

References

1. Kopecká, B., Špringer, V., and Majer, J., *Chem. Zvesti 35*, 641 (1981).
2. Jungreis, E. and Thabet, S., in *Chelates in Analytical Chemistry*, Vol. *II*. (Flaschka, H. A. and Barnard, A. J., Editors.) Pp. 149—177. M. Dekker, New York, 1969.
3. Rodriquez, J. M. B., *Talanta 27*, 923 (1980).
4. Garcia-Vargas, M., Gallege, M., and de La Quardia, M., *Analyst* (London) *105*, 965 (1980).
5. Scott, A. W. and Wood, B. L., *J. Org. Chem. 7*, 508 (1942).
6. Karlíček, R., *Spectrovision 23*, 7 (1970).
7. Bates, R. G., *Anal. Chem. 40*, 29A (1968).
8. Horák, M. and Papoušek, D., *Infračervená spektra a struktura molekul.* (Infrared Spectra and Structure of Molecules.) Academia, Prague, 1976.
9. Sommer, L., Kubáň, V., and Havel, J., *Folia Fac. Rerum Natur. Univ. Brno*, Vol. *11*, Part 7 (*Chemia*), p. 3, 1970.
10. Vláčil, F. et al., *Příklady z chemické a instrumentální analýzy.* (Examples from Chemical and Instrumental Analysis.) Nakladatelství technické literatury. (Publishing House of Technical Literature.) Prague, 1983.
11. Špringer, V. and Kopecká, B., *Československ. Farm. 33*, 6 (1984).