

## SPECTROPHOTOMETRIC STUDY OF THE COMPLEXATION EQUILIBRIA AND DETERMINATION OF COPPER(II) WITH 1-(2-THIAZOLYL-AZO)-2-NAPHTHOL-3,6-DISULPHONIC ACID

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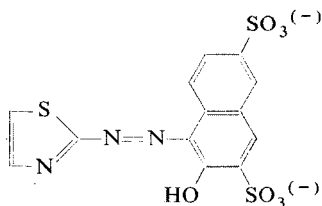
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Received May 23rd, 1974

The complexation equilibria of 1-(2-thiazolylazo)-2-naphthol-3,6-disulphonic acid with  $\text{Cu}^{2+}$  ions have been studied in aqueous solutions. The composition, molar absorption coefficient and stability constant of the 1 : 1 chelate formed have been evaluated from the absorbance curves by graphical methods and by computer calculations. The conditions for the spectrophotometric determination of copper(II) with this reagent have been found.

1-(2-Thiazolylazo)-2-naphthol-3,6-disulphonic acid (TAN-3,6-S) was first prepared by Kawase, who studied the reagent properties and its complexation equilibria with  $\text{Cu}^{2+}$  (ref.<sup>1</sup>) and  $\text{Zn}^{2+}$



(ref.<sup>2</sup>) ions spectrophotometrically. Later, the reagent was proposed for the spectrophotometric determination of palladium(II) (ref.<sup>3</sup>), as an indicator for chelometric determinations of gallium(III), indium(III) and thallium(III) (ref.<sup>4</sup>) and for the mercurimetric determination of chloride, bromide and thiocyanate<sup>5</sup>. The reagent solubility in water is a great advantage.

In the present paper, the complexation equilibria of 1-(2-thiazolylazo)-2-naphthol-3,6-disulphonic acid with  $\text{Cu}^{2+}$  ions were studied spectrophotometrically and evaluated by combined graphical and numerical analysis of the absorbance curves and the conditions for the spectrophotometric determination of  $\text{Cu}^{2+}$  with this reagent in aqueous solutions were specified.

## EXPERIMENTAL AND RESULTS

## Chemicals and Instruments

The titre of the standard solution of *p.a.* cupric nitrate was determined complexometrically. The purity of 1-(2-thiazolylazo)-2-naphthol-3,6-disulphonic acid (sodium salt, dihydrate) prepared by us and of the sample from the Research Institute of Pure Chemicals, Lachema, Brno, was checked by determining the sodium content, water content by drying at 135°C, by paper chromatography in an *n*-butanol–water–acetic acid (2 : 1 : 1) system, by elemental analysis and by spectrophotometric titration with cupric nitrate, using the 1 : 1 chelate at pH 3.5 (a 0.02M formate buffer) with a final reagent concentration of  $2.7 \cdot 10^{-5}$  M and  $\lambda$  580 nm. The ionic strength was adjusted to  $I = 0.1$  during study of the reagent and of the complexation equilibria in aqueous solutions at pH > 1 by additions of potassium nitrate; in some dependences at pH < 1, the sum,  $c_{\text{HNO}_3} + c_{\text{KNO}_3} = I = 1.0$ , was maintained. The spectrophotometric measurements were carried out on a SF-4A spectrophotometer (USSR) in 10 mm cuvettes; the measurements at  $I = 0.1$  (pH > 1.0) were performed in the continuous apparatus<sup>6</sup>. The pH of the solutions was measured with a pHK-1 instrument (Czechoslovakia) with a Radiometer G 202B glass electrode and a saturated calomel electrode.

*Acid-Base Equilibria of 1-(2-Thiazolylazo)-2-naphthol-3,6-disulphonic Acid (TAN-3,6-S)*

The dependence of the absorbance of TAN-3,6-S aqueous solutions on pH at ionic strength  $I = 0.1$  and on  $(-\log [\text{H}^+])$  (not corrected using activity coefficients) at  $I = 1.0$ , at  $\lambda$  510 and 560 nm indicate the dissociation of the thiazolium cation of the dye ( $\text{LH}_2^+$ ) at pH < 3 and the dissociation of the hydroxyl group in *ortho* position with respect to the azo-group (the LH form) in the pH range, 6–10 (Fig. 1). In the series of TAN-3,6-S absorption curves in dependence on pH, the equilibria are

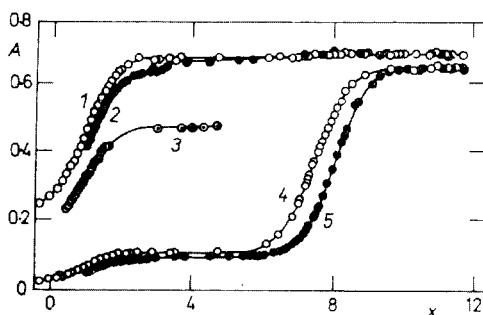


FIG. 1

Absorbance vs pH or  $(-\log [\text{H}^+])$  plots of TAN-3,6-S Solutions

Curve 1  $c_L = 4.10 \cdot 10^{-5}$  M, 510 nm,  $x = -\log [\text{H}^+]$ ,  $I 1.0$ ; 2  $c_L = 4.10 \cdot 10^{-5}$  M, 510 nm,  $x = \text{pH}$ ,  $I 0.1$ ; 3  $c_L = 4.99 \cdot 10^{-4}$  M, 575 nm,  $x = -\log [\text{H}^+]$ ,  $I 1.0$ ; 4  $c_L = 4.10 \cdot 10^{-5}$ , 560 nm,  $x = -\log [\text{H}^+]$ ,  $I 1.0$ ; 5  $c_L = 4.10 \cdot 10^{-5}$  M, 560 nm,  $x = \text{pH}$ ,  $I 0.1$ .

characterized by isosbestic points at 466 and 508 nm (Fig. 2). The dissociation constants of the  $\text{LH}_2^+$  and LH forms of the reagent, obtained by logarithmic analysis of the absorbance *vs* pH (or  $-\log [\text{H}^+]$ ) dependences for solutions with  $c_L = 4.00 \cdot 10^{-5}\text{M}$  and ionic strength  $I = 0.1$  and  $1.0$  and with  $c_L = 5.00 \cdot 10^{-4}\text{M}$  and  $I = 1.0$ , and the values obtained by computation employing the PRCEK II program (ref.<sup>7</sup>), are given in Table I, along with the  $\lambda_{\text{max}}$  and molar absorption coefficient values.

### Complexation Equilibria in Solutions of TAN-3,6-S with $\text{Cu}^{2+}$

The absorption curves of  $3.30 \cdot 10^{-5}\text{M}$  TAN-3,6-S solutions with  $\text{Cu}^{2+}$  concentrations increasing up to  $6.53 \cdot 10^{-5}\text{M}$  at pH 3.92 show simple equilibrium between the orange reagent (HL) and the blue-purple  $\text{CuL}$  complex with  $\lambda_{\text{max}}$  575 nm (main maximum) and 415 nm. Because of the existence of an isosbestic point at 523 nm, no other equilibria are probable under the given conditions (Fig. 3). In order to follow the effect of acidity on the complexation equilibria, two procedures were again employed. In equimolar solutions with  $c_L = c_M = 3.30 \cdot 10^{-5}\text{M}$ , with an ascending branch at  $\text{pH} > 1$ , the pH was measured with a glass electrode and constant ionic strength  $I = 0.1$  was maintained by addition of potassium nitrate (Fig. 4, curve 1). In dependences with an excess of one of the components ( $c_M/c_L = 25 - 100$  and  $c_L/c_M = 15$ ) and with the ascending branch at  $\text{pH} < 1$ , defined amounts of nitric acid and potassium nitrate were added to the solutions, so that the sum of their molarities was constant,  $I = c_{\text{HNO}_3} + c_{\text{KNO}_3} = 1.0$ , the acidity being expressed by the  $(-\log [\text{H}^+])$  value, not corrected using activity coefficients (Fig. 4, curves 2–4 and 6). From Fig. 4 it is evident that in solutions with excess metal or ligand the complex begins to form in about 1M acid, while in equimolar solutions it is formed above pH 1. While

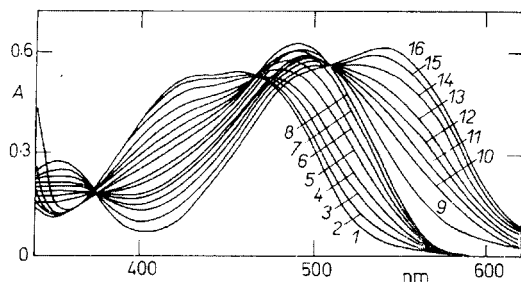


FIG. 2

#### Absorption Spectra of TAN-3,6-S Solutions at Various pH Values

$c_L = 3.44 \cdot 10^{-5}\text{M}$ ,  $I = 0.1$ . pH: curve 1 3M- $\text{HClO}_4$ , 2 0.40, 3 0.75, 4 1.03, 5 1.34, 6 1.64, 7 1.95, 8 2.64, 3.75 and 4.44, 9 7.04 10 7.47, 11 7.67, 12 7.73, 13 8.06, 14 8.40, 15 8.93, 16 10.04 and 11.67.

the equimolar curves and those with excess metal have almost identical horizontal parts in solutions with excess of reagent,  $c_L/c_M = 15$  and  $10$ , the absorbance in the horizontal part is lower (Fig. 4, curves 6 and 7).

### Methods for Studying Complexation Equilibria

The molar absorption coefficient and the equilibrium constant values were determined from the absorbance plots vs pH,  $(-\log [H^+])$ ,  $c_M$  and  $c_L$ , employing the graphical analysis described earlier<sup>7</sup> and using the PRCEK III program<sup>8</sup> and a MSP 2A computer. This program simulates in fact the usual operations carried out during application of classical graphical methods. The only significant difference is that the

TABLE I  
The Molar Absorption Coefficients and Dissociation Constants of TAN-3,6-S

$\lambda_{\max}$ , nm	$\epsilon_{\lambda_{\max}}$ , $\text{cm}^2\text{M}^{-1}$	$\text{p}K_a^a$	$I^b$	Method
430—460	$15\,653 \pm 42^c$	$\text{LH}_2^+$		
		$1.04^d \pm 0.02$	0.1	computer
	$15\,579 \pm 42$	$1.06^e$	0.1	graphical
		$0.97^f \pm 0.02$	1.0	computer
		$0.94^g$	1.0	graphical
		$0.93^h \pm 0.01$	1.0	computer <sup>i</sup>
		$0.95^j$	1.0	graphical <sup>i</sup>
490	$17\,682 \pm 29$	$\text{LH}$		
		$7.86^k \pm 0.02$	0.1	computer
	$17\,989 \pm 51$	$7.96^l$	0.1	graphical
		$7.36^m \pm 0.02$	1.0	computer
	$18\,060$	$7.44^n$	1.0	graphical
540	$17\,255 \pm 211$	$\text{L}^-$		
			0.1	computer
			1.0	computer
	$17\,694$		1.0	graphical

<sup>a</sup>  $K_a = [\text{LH}_{n-1}][\text{H}]/[\text{LH}_n]$ ; <sup>b</sup> the ionic strength, adjusted to  $I = 1.0$  by a constant sum of  $c_{\text{HNO}_3} + c_{\text{KNO}_3} = 1\text{M}$  for the  $A = f(-\log [H^+])$  curves; <sup>c</sup> at 430 nm; <sup>d</sup> the average of the values,  $1.00 \pm 0.01$  (510 nm),  $1.08 \pm 0.03$  (430 nm),  $1.04 \pm 0.02$  (490 nm); <sup>e</sup> at 510 nm; <sup>f</sup> the average of the values,  $0.97 \pm 0.01$  (510 nm),  $0.98 \pm 0.01$  (430 nm),  $0.92 \pm 0.02$  (490 nm),  $1.01 \pm 0.01$  (540 nm); <sup>g</sup> the average of the values,  $0.96$  (510 nm),  $0.92$  (490 nm); <sup>h</sup> the average of the values,  $0.93 \pm 0.01$  (560 nm) and  $0.92 \pm 0.01$  (575 nm); <sup>i</sup>  $c_L = 5.00 \cdot 10^{-4}\text{M}$ , otherwise  $c_L = 4.00 \cdot 10^{-5}\text{M}$ ; <sup>j</sup> at 575 nm; <sup>k</sup> the average of the values,  $7.83 \pm 0.01$  (575 nm),  $7.90 \pm 0.02$  (540 nm); <sup>l</sup> at 560 nm; <sup>m</sup> the average of the values,  $7.36 \pm 0.02$  (560 nm),  $7.35 \pm 0.02$  (575 nm) and  $7.36 \pm 0.02$  (540 nm); <sup>n</sup> the average of the values,  $7.42$  (560 nm) and  $7.46$  (540 nm).

parameters of the linear plots obtained by means of a suitable transformed equation<sup>7</sup> are sought by the objective linear least squares method and the values of the basic characteristics of the complex species (the molar absorption coefficient and the equilibrium constant) are determined by successive approximations. As the initial step  $A_{0i}$  values of the absorbances of the two horizontal parts of the absorbance *vs* pH curve are used and the  $A_{0i}$  values resulting from the previous cycle are employed in each successive approximation. After attaining the required agreement between two

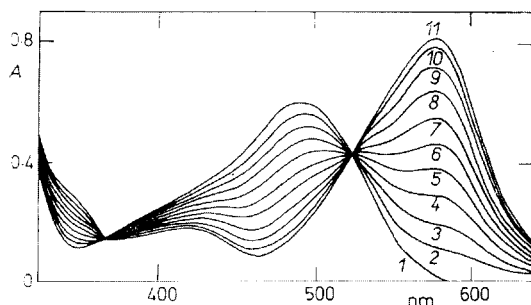


FIG. 3

Absorption Spectra of TAN-3,6-S Solutions with Increasing  $\text{Cu}^{2+}$  concentration

$c_L = 3.30 \cdot 10^{-5} \text{M}$ , pH 3.92,  $10 \cdot 1$ .  $c_M$ : curve 1 0,  $2.3.63 \cdot 10^{-6} \text{M}$ ,  $3.7.25 \cdot 10^{-6} \text{M}$ ,  $4.1.09 \cdot 10^{-5} \text{M}$ ,  $5.1.45 \cdot 10^{-5} \text{M}$ ,  $6.1.81 \cdot 10^{-5} \text{M}$ ,  $7.2.18 \cdot 10^{-5} \text{M}$ ,  $8.2.54 \cdot 10^{-5} \text{M}$ ,  $9.2.90 \cdot 10^{-5} \text{M}$ ,  $10.3.26 \cdot 10^{-5} \text{M}$ ,  $11.6.53 \cdot 10^{-5} \text{M}$ .

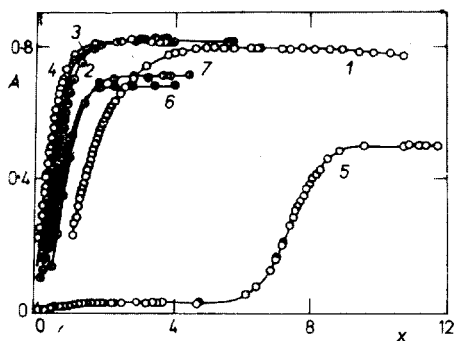


FIG. 4

Absorbance *vs* pH or  $(-\log [\text{H}^+])$  Plots of TAN-3,6-S Solution in Presence of  $\text{Cu}^{2+}$

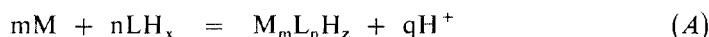
Curves 1–5:  $c_L = 3.30 \cdot 10^{-5} \text{M}$ ; 1  $c_M = c_L$ , 2  $c_M/c_L = 25.3$ , 3  $c_M/c_L = 50.6$ , 4  $c_M/c_L = 100.6$ , 5  $c_M = 0$ , 6  $c_M = 3.30 \cdot 10^{-5} \text{M}$ ,  $c_L/c_M = 15.1$ , 7  $c_M = 3.30 \cdot 10^{-5} \text{M}$ ,  $c_L/c_M = 10$ . 1  $x = \text{pH}$ ,  $I = 0.1$ ; 2–6:  $x = -\log [\text{H}^+]$ ,  $I 1.0$ ; 7  $x = \text{pH}$ ,  $I 0.5$ .

successive  $A_{o_i}$  values or after attaining a maximum number of cycles, the results are printed in tables containing all the necessary data on the studied system. The most probable reaction mechanism is chosen only for a single computing cycle, the values of the coefficients in equation (A) being gradually fed into the machine from a coupled typewriter; on the basis of the correlation coefficient and the sum of squares of the deviations, the most suitable  $m$ ,  $q$ ,  $n$  combination is then selected. More details on the application of the PRCEK program have been published elsewhere<sup>9,10</sup>. The chelate composition was also determined by the variation method in equimolar solutions.

The spectrophotometric analytical curve,  $A = f(c_{Cu})$ , was evaluated on the MSP 2A computer using the LLSQM program<sup>11</sup>. During calculation of the molar absorption coefficients and the constants of the reagent and the chelate, their standard deviations were also calculated by the least squares method and recorded in the tables of results.

### Graphical Analysis of the Absorbance Curves

The ascending parts of the absorbance *vs* pH or absorbance *vs*  $(-\log [H^+])$  plots for solutions of TAN-3,6-S with  $Cu^{2+}$  (Fig. 4, curves 1–4 and 6) were subjected to direct and logarithmic analysis, using equations (1)–(6) derived earlier<sup>7</sup>. For equilibrium



with equilibrium constant  $k = [ML_nH_z][H]^q/[M][LH_x]^n$ , neglecting the metal ion absorbance and on the condition that  $c_M = c_L$ , the equation was used:

$$c_L/A = 1/\varepsilon + \sqrt{(ZA - \varepsilon_L c_L)} [H]^q \sqrt{(Z\varepsilon - \bar{\varepsilon}_L)} / A \sqrt{Z\varepsilon} \sqrt{k}, \quad (1)$$

$$\log(ZA - \bar{\varepsilon}_L c_L) / (\varepsilon c_L - A)^2 Z = qpH + \log k - \log(Z\varepsilon - \bar{\varepsilon}_L). \quad (2)$$

For  $c_M > c_L$ ,

$$A = \varepsilon c_L / n - \{(ZA - \bar{\varepsilon}_L c_L)(n\bar{\varepsilon}_L - \varepsilon Z)^{n-1} [H]^q\} / (nA - \varepsilon c_L)^{n-1} k n c_M^n, \quad (3)$$

$$\begin{aligned} \log(ZA - \bar{\varepsilon}_L c_L) / (\varepsilon c_L - nA)^n + (n-1) \log(Z\varepsilon - n\bar{\varepsilon}_L) = \\ = qpH + m \log c_M + \log k, \end{aligned} \quad (4)$$

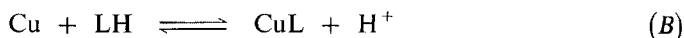
For  $c_L > c_M$ ,

$$c_M / (A - A_{oL}) = 1/\varepsilon + [H]^q Z / k \varepsilon c_L^n, \quad (5)$$

$$\log(A - A_{oL}) Z / (\varepsilon c_M - A + A_{oL}) = \log k + qpH + n \log c_L, \quad (6)$$

In these equations,  $Z = 1 + [\text{H}]/K_{a1}$ ,  $\bar{\epsilon}_L = \epsilon_L + \epsilon_{LH}[\text{H}]/K_{a1}$ ,  $A_{oL} = \bar{\epsilon}_L c_L/Z$ ,  $\epsilon_L$  and  $\epsilon_{LH}$  are the molar absorption coefficients of the LH and  $\text{LH}_2^+$  forms of the reagent, respectively, and  $\epsilon$  is the molar absorption coefficient of the chelate.

The straight lines obtained by analysis of the absorbance vs pH or  $(-\log[\text{H}^+])$  curves, using equations (1), (3) and (5) after substitution of the values  $n = q = 1$ , verified the validity of the equilibrium



(the charges are omitted for the sake of simplicity). From the intercepts of these straight lines with the  $y$ -axis, the molar absorption coefficient of the CuL chelate was determined (Fig. 5, curves 1–5).

The logarithmic plots constructed by means of equations (2), (4) and (6), with unit slopes also verified that one proton is liberated during the formation of the CuL chelate and were employed for the determination of the equilibrium constant for equi-

TABLE II

The Values of the Molar Absorption Coefficient and the Equilibrium Constant of Chelate CuL, Obtained from the  $A = f(-\log[\text{H}^+])$  Plots at  $I = 1.0$  ( $\text{KNO}_3$ )

$\epsilon(575 \text{ nm})$ $\text{cm}^2 \text{M}^{-1}$	$\log k_2^a$	$r_{xy}^b$	$U^c$	$c_M/c_L^d$ ( $c_L/c_M$ )	Method
24 591	2.03			100.6	graphical, 575 nm
24 863 $\pm$ 1 709	2.09 $\pm$ 0.01	0.99778	0.5877 $\cdot$ 10 <sup>-2</sup>	100.6	computer, 575 nm
24 545 <sup>e</sup>	2.11 $\pm$ 0.06			100.6	computer, average <sup>f</sup>
25454	2.08			50.6	graphical, 575 nm
24 454 $\pm$ 3 297	2.13 $\pm$ 0.07	0.99995	0.2919 $\cdot$ 10 <sup>-1</sup>	50.6	computer, 575 nm
24 545 <sup>e</sup>	2.11 $\pm$ 0.04			50.6	computer, average <sup>f</sup>
24 773	2.11			25.3	graphical, 575 nm
25 350 $\pm$ 1 646	2.15 $\pm$ 0.04	0.99959	0.1244 $\cdot$ 10 <sup>-1</sup>	25.3	computer, 575 nm
24 787 <sup>e</sup>	2.14 $\pm$ 0.03			25.3	computer, average <sup>f</sup>
20.833	2.17			(15.1)	graphical, 575 nm
21.997 $\pm$ 525	2.12 $\pm$ 0.00	0.99982	0.9234 $\cdot$ 10 <sup>-4</sup>	(15.1)	computer, 575 nm
20 697 <sup>e</sup>	2.12 $\pm$ 0.06			(15.1)	computer, average <sup>f</sup>

<sup>a</sup> Constant  $k_1 = [\text{CuL}][\text{H}]/[\text{Cu}][\text{LH}]$  determined graphically and recalculated to  $k_2 = [\text{CuL}] \cdot [\text{H}]^2/[\text{Cu}][\text{LH}_2^+]$ , employing the relation,  $k_2 = k_1 K_{a1}$ , where  $K_{a1}$  is the dissociation constant of the reagent,  $\text{LH}_2^+$  form; <sup>b</sup> correlation coefficient; <sup>c</sup> the sum of the squares of the deviations of the measured and the calculated absorbance; <sup>d</sup> ascending parts in the region,  $(-\log[\text{H}^+]) = 0.0-1.2$ ; <sup>e</sup> the value determined from the horizontal part as  $A/c_L$  or  $A/c_M$  at 575 nm; <sup>f</sup> the average of the values for  $\lambda$  540, 560, 575 and 600 nm.

librium (B),  $k_1 = [\text{CuL}][\text{H}^+]/[\text{Cu}][\text{LH}]$ . Transformation (5) for solutions with excess reagent,  $c_L/c_M = 15.1$ , is also linear; the original absorbance curve,  $A - A_{oL} = f(-\log[\text{H}^+])$  (Fig. 4, curve 6) has a lower horizontal part. This anomaly was manifested only in a lower molar absorption coefficient value compared to the other results.

The resulting values of the molar absorption coefficient of the CuL chelate at 575 nm and its equilibrium constant for  $I = 1.0$ , obtained by the graphical methods, the molar absorption coefficient values obtained from the horizontal parts of the absorbance curves and the equilibrium constant values obtained by computer treatment are summarized in Table II. The values obtained from the equimolar pH-curves are given in Table III for  $I = 0.1$ . The absorbance dependences of solutions of  $3.30 \cdot 10^{-5}\text{M}$  TAN-3,6-S on the  $\text{Cu}^{2+}$  concentration in the range  $0 - 5 \cdot 10^{-4}\text{M}$  at pH 1.12 and 1.63, and the absorbance vs TAN-3,6-S concentration plots for

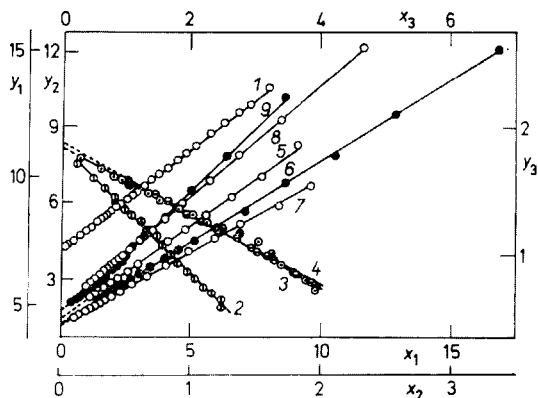


FIG. 5

Graphical Analysis of the Absorbance vs pH, or  $(-\log [\text{H}^+])$ ,  $c_M$  and  $c_L$  Plots in Solutions of TAN-3,6-S with  $\text{Cu}^{2+}$  at 575 nm

1 direct analysis of the curve,  $A = f(\text{pH})$ ,  $I = 0.1$ ,  $c_M = c_L = 3.30 \cdot 10^{-5}\text{M}$ ,  $x_1 = \sqrt{[(ZA - \bar{\epsilon}_L c_L) \cdot [\text{H}](Z\bar{\epsilon} - \bar{\epsilon}_L)]/A} \cdot \sqrt{(Z \times 10)}$ ,  $y_2 = (c_L/A) \cdot 10^{-5}$ . 2-5 analysis of the curves,  $A = f(-\log [\text{H}^+])$ ,  $I = 1.0$ ; 2  $c_L = 3.30 \cdot 10^{-5}\text{M}$ ,  $c_M/c_L = 25.3$ ,  $x_1 = (ZA - \bar{\epsilon}_L c_L) [\text{H}] \cdot 10$ ,  $y_2 = A \cdot 10$ ; 3  $c_L = 3.30 \cdot 10^{-5}\text{M}$ ,  $c_M/c_L = 100.6$ ,  $x_2 = (ZA - \bar{\epsilon}_L c_L) [\text{H}] \cdot 10$ ,  $y_2 = A \cdot 10$ ; 4  $c_L = 3.30 \cdot 10^{-5}\text{M}$ ,  $c_M/c_L = 50.6$ ,  $x_1 = (ZA - \bar{\epsilon}_L c_L) [\text{H}] \cdot 10$ ,  $y_2 = A \cdot 10$ ; 5  $c_M = 3.30 \cdot 10^{-5}\text{M}$ ,  $c_L/c_M = 15.1$ ,  $x_2 = Z [\text{H}]$ ,  $y_3 = c_M/(A - A_{oL}) \cdot 10^4$ ; 6, 7 analysis of the curves,  $A = f(c_M)$ ,  $c_L = 3.30 \cdot 10^{-5}\text{M}$ ,  $I = 0.1$ ; 6 pH 1.12,  $x_2 = 1/[c_M - (A - A_{oL})/(\bar{\epsilon} - \bar{\epsilon}_L/Z)] \cdot 10^{-4}$ ,  $y_2 = c_L \cdot 10^5/(A - A_{oL})$ ; 7 pH 1.63,  $x_1 = 1/[c_M - (A - A_{oL})/(\bar{\epsilon} - \bar{\epsilon}_L/Z)] \cdot 10^{-4}$ ,  $y_1 = c_L \cdot 10^5/(A - A_{oL})$ ; 8, 9 analysis of the curves,  $A = f(c_L)$ ,  $c_M = 3.29 \cdot 10^{-5}\text{M}$ ,  $I = 0.1$ ; 8 pH 1.16,  $x_3 = 1/[c_L - (A - A_{oL})/(\bar{\epsilon} - \bar{\epsilon}_L/Z)] \cdot 10^{-4}$ ,  $y_1 = c_M \cdot 10^5/(A - A_{oL})$ ; 9 pH 1.65,  $x_2 = 1/[c_L - (A - A_{oL})/(\bar{\epsilon} - \bar{\epsilon}_L/Z)] \cdot 10^{-5}$ ,  $y_1 = c_M \cdot 10^5/(A - A_{oL})$ .



solutions of  $3.30 \cdot 10^{-5} \text{M}$   $\text{Cu}^{2+}$  up to  $5 \cdot 10^{-4} \text{M}$  at pH 1.16 and 1.65 and  $I = 0.1$  (Fig. 6, curves 1–4) were treated graphically employing equations (7)–(10) for direct and logarithmic analysis of the concentration dependences, derived earlier<sup>7</sup>. It holds for the  $A = f(c_M)$  dependence that

$$c_L / (A - A_{oL}) = 1 / (\varepsilon - \bar{\varepsilon}_L / Z) + [\text{H}^+]^q Z^n / \{c_M - n(A - A_{oL})\} / (\varepsilon - \bar{\varepsilon}_L / Z)^m (\varepsilon - \bar{\varepsilon}_L / Z) k, \quad (7)$$

$$\log \{(A - A_{oL}) / (\varepsilon c_L - A)\} = m \log \{c_M - m(A - A_{oL}) / (\varepsilon - \bar{\varepsilon}_L / Z)\} + q \text{pH} + \log k - \log Z. \quad (8)$$

For the  $A = f(c_L)$  dependence it is valid that

$$c_M / (A - A_{oL}) = 1 / (\varepsilon - n\bar{\varepsilon}_L / Z) + [\text{H}^+]^q Z^n / \{c_L - n(A - A_{oL}) / (\varepsilon - \bar{\varepsilon}_L / Z)\}^n (\varepsilon - n\bar{\varepsilon}_L / Z) k, \quad (9)$$

$$\log (A - A_{oL}) / [c_M - (A - A_{oL}) / (\varepsilon - n\bar{\varepsilon}_L / Z)] = \log k + q \text{pH} + n \log [c_L - n(A - A_{oL}) / (\varepsilon - n\bar{\varepsilon}_L / Z)] + \log (\varepsilon - n\bar{\varepsilon}_L / Z) - n \log Z. \quad (10)$$

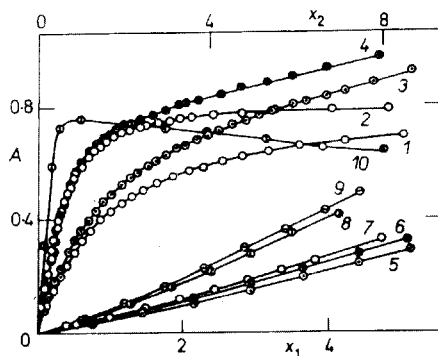


FIG. 6

Absorbance vs  $c_M$  or  $c_L$  Plots in Solutions of TAN-3,6-S with  $\text{Cu}^{2+}$

Curves 1–7  $I$  0.1, 575 nm; 8–10  $I$  0.5 or  $I$  1.0, 580 nm. 1  $c_L = 3.30 \cdot 10^{-5} \text{M}$ ,  $x_1 = c_M \cdot 10^4$ , pH 1.12; 2  $c_L = 3.30 \cdot 10^{-5} \text{M}$ ,  $x_1 = c_M \cdot 10^4$ , pH 1.63; 3  $c_M = 3.29 \cdot 10^{-5} \text{M}$ ,  $x_1 = c_L \cdot 10^4$ , pH 1.16; 4  $c_M = 3.29 \cdot 10^{-5} \text{M}$ ,  $x_1 = c_L \cdot 10^4$ , pH 1.65; curves 5–9:  $c_M = 0$ ; 5  $x_1 = c_L \cdot 10^4$ , pH 1.16; 6  $x_1 = c_L \cdot 10^4$ , pH 1.65; 7  $x_1 = c_L \cdot 10^4$ , pH 2.85 (0.05M chloroacetate),  $I$  0.1; 8  $x_2 = c_L \cdot 10^4$ , pH 3.84 (0.05M chloroacetate),  $I$  0.5; 9  $x_2 = c_L \cdot 10^4$ , pH 2.80 (0.05M chloroacetate)  $I$  1.0; 10  $c_M = 3.30 \cdot 10^{-5} \text{M}$ ,  $x_2 = c_L \cdot 10^4$ , pH 2.66 (0.05M chloroacetate),  $I$  0.5, reagent absorbance subtracted.

The straight lines related to the concentration dependences with the help of equations (7) and (9) after substituting  $m = n = q = 1$  verified equilibrium (B) and yielded the value of the molar absorption coefficient of the CuL chelate from the intercept on the  $y$ -axis (Fig. 5, curves 6–9). The unit slope of the logarithmic plots according to equations (8) and (10) verified that one ligand is bound to central ion  $\text{Cu}^{2+}$ ; equilibrium constant  $k_1$  for chelate CuL was also determined from these straight lines. The distortion of the boundary parts of the straight lines obtained during the analysis of the  $A = f(c_L)$  plot in a region of  $c_L > 1.07 \cdot 10^{-4} \text{M}$  (Fig. 5, curve 9) is apparently connected with the above-mentioned anomalous decrease in the absorbance of solutions containing excess reagent. The resulting values of the molar absorption coefficient of chelate CuL at 575 nm and the equilibrium constant for  $I = 0.1$  are summarized in Table III, together with the values obtained by the computer.

#### Interpretation of the Absorbance Curves Using the PRCEK III Program

Analysis of the absorbance *vs* pH or ( $-\log [\text{H}^+]$ ) curves was carried out for various concentration ratios on the basis of similar assumptions as those made in the graphical

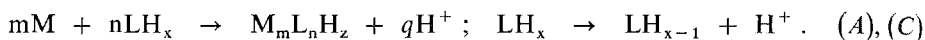
TABLE III

The Values of the Molar Absorption Coefficient and the Equilibrium Constant of Chelate CuL, Obtained from the pH-Curves of Equimolar Solutions and from the Concentration Dependences at  $I = 0.1$  ( $\text{KNO}_3$ )

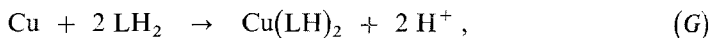
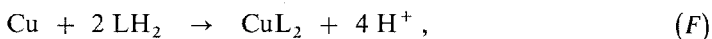
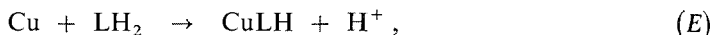
$\epsilon(575 \text{ nm})$ $\text{cm}^2 \cdot \text{M}^{-1}$	$\log k_2^a$	$r_{xy}^b$	$U^c$	pH	Method
24 155	2.39			1.1–4.2 <sup>d</sup>	graphical, 575 nm
24 071 $\pm$ 1 024	2.31 $\pm$ 0.02	0.99975	0.4819 $\cdot 10^{-2}$	1.1–4.2	computer, 575 nm
23 939 <sup>e</sup>	2.36 $\pm$ 0.03			1.1–4.2	computer, average <sup>f</sup>
24 213	2.27			1.12	graphical, 575 nm
21 586 $\pm$ 863	2.39 $\pm$ 0.02	0.99881	0.4897 $\cdot 10^{-1}$	1.12	computer, 575 nm
	2.27 $\pm$ 0.03			1.12	computer, average <sup>f</sup>
24 879	2.29			1.63	graphical, 575 nm
24 722 $\pm$ 314	2.27 $\pm$ 0.05	0.99819	0.2706 $\cdot 10^{-1}$	1.63	computer, 575 nm
	2.30 $\pm$ 0.04			1.63	computer, average <sup>f</sup>
21 688	2.34			1.16	graphical, 575 nm
23 357 $\pm$ 490	2.21 $\pm$ 0.04	0.99619	0.1442 $\cdot 10^{-1}$	1.16	computer, 575 nm
	2.29 $\pm$ 0.07			1.16	computer, average <sup>f</sup>
23 042	2.33			1.65	graphical, 575 nm
24 078 $\pm$ 1924	2.27 $\pm$ 0.01	0.99740	0.4498	1.65	computer, 575 nm
	2.24 $\pm$ 0.02			1.65	computer, average <sup>f</sup>

<sup>a,b,c,e,f</sup> See notes at Table II, <sup>d</sup> the absorbance–pH curve, otherwise concentration dependences.

analysis and employing equations analogous to equations (3)–(6) for general equilibrium (A), simultaneously considering the ligand dissociation, (C)



The following equilibria were considered as possible:



For the calculation values read directly from the graphs were employed as the input absorbance values for both horizontal parts of the absorbance–pH curves. Because the calculation is time-consuming, the selection of the most probable reaction mechanism, (D)–(H), was carried out only for a single wavelength, namely that of the complex absorption maximum, 575 nm. The correlation coefficients and the sum of the squares of the deviations were used as criteria for evaluating the suitability of particular mechanisms. Correlation coefficients closest to unity and the smallest values of the sum of the squares of the deviations were obtained for reaction mechanism (D), *i.e.* the formation of complex CuL, which is in agreement with the results of the graphical methods.

All the absorbance *vs* pH or ( $-\log [H^+]$ ) curves were then tested at wavelengths of 540, 560, 575 and 600 nm for reaction mechanism (D). The values of the molar absorption coefficient of chelate CuL, equilibrium constant  $k_2 = [CuL][H]^2/[Cu][LH_2]$  at 575 nm and  $I = 1.0$ , their standard deviations, the  $r_{xy}$  and  $U$  values and the average values of  $k_2$  for all wavelengths studied are given in Table II, together with the values obtained by the graphical methods.

The concentration dependence curves,  $A = f(c_L)$  and  $A = f(c_M)$ , at a constant pH were tested analogously for equilibrium (D), *i.e.* for coefficients  $m = n = 1$ ,  $q = 2$ , using modified equations (7) and (9). The values of the molar absorption coefficient of chelate CuL, equilibrium constant  $k_2$  for  $I = 0.1$  and  $\lambda$  575 nm, their standard deviations, the  $r_{xy}$  and  $U$  values and the average values of  $k_2$  for  $\lambda$  540, 560, 575 and 600 nm are summarized in Table III.

The values of the stability constant of chelate CuL,  $\beta_{11} = [CuL]/[Cu][L]$ , calculated from the relation,  $\beta_{11} = k_2/K_{a1}K_{a2}$ , using the average values of  $k_2$ ,  $K_{a1}$

and  $K_{a2}$  obtained by the computer, are  $\log \beta_{1,1} = 11.19$  for  $I = 0.1$  and  $\log \beta_{1,1} = 10.43$  for  $I = 1.0$ , in this case the acidity being expressed in terms of  $(-\log [H^+])$ . The average value of the molar absorption coefficient of chelate  $CuL$  at 575 nm is  $\epsilon = 24.414$ , not considering the anomalous values obtained with excess reagent.

#### Job Plots in Equimolar Solutions

In solutions with  $c_M + c_L = 6.60 \cdot 10^{-5}M$  and pH 1.05 to 9.45, the existence of the 1 : 1 complex was verified by the method of continuous variations (Fig. 7). The plot at pH 9.45 is lower than that at pH 4.99, although the same chelate is formed in both cases. This decrease is due to subtraction of higher absorbance values of the purple ligand form in the alkaline medium over the whole range of the plot, while, with the stable complex, it would be more suitable to subtract the reagent absorbance only in the right-hand half of the variation curve.

#### The Spectrophotometric Determination of Copper(II) with TAN-3,6-S

A pH range from 2.5 to 4.5 can be recommended for the determination of cupric ions with TAN-3,6-S, since the absorbance of the complex ( $CuL$ ) and the reagent (the LH form) are constant within this range (Fig. 4, curve 7). 0.5M chloroacetate, 0.2M formate and 0.1M acetate buffers do not affect the absorbance. Because of the presence of traces of metal impurities in the chemicals (buffers and potassium nitrate used for adjustment of the ionic strength) and possibly some other metals in the samples analyzed, the interference from which increases with increasing pH, it is desirable to maintain the conditions constant, especially the concentrations of the chemicals added and the acidity (the latter is best kept at the beginning of the above given range) and to check the blank determination. During the following experiments, the solution acidity was adjusted to pH 2.6 with a 0.05M chloroacetate buffer.

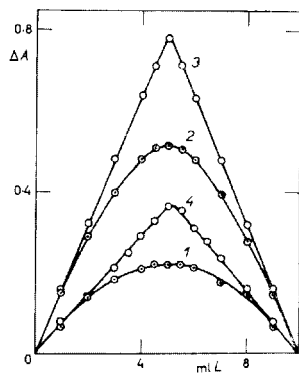


FIG. 7  
Job Curves of TAN-3,6-S Solutions with  $Cu^{2+}$   
 $c_0 = 6.60 \cdot 10^{-5}M$ ,  $I 0.1$ , 575 nm. pH: 1 1.05, 2 1.65,  
3 4.99, 4 9.45.

TABLE IV

The Statistical Parameters for the Spectrophotometric Calibration Curve for the Determination of Copper(II) with TAN-3,6-S, Obtained by the MSP 2A Computer Using the LLSQM Program

$\lambda$ , nm	$\epsilon_{\text{CuL}}^{\text{CuL}}$ $\text{cm}^2 \text{M}^{-1}$	$\epsilon_{\text{LH}}^{\text{LH}}$ $\text{cm}^2 \text{M}^{-1}$	$q^a$ A
575	$20\,916 \pm 59$	$836 \pm 56$	$0.276 \pm 0.002$
580	$21\,303 \pm 38$	$588 \pm 35$	$0.194 \pm 0.001$
590	$19\,657 \pm 43$	$334 \pm 16$	$0.110 \pm 0.001$

The reagent concentration of  $c_L = 3.30 \cdot 10^{-4} \text{M}$  was employed for the determination of  $\text{Cu}^{2+}$  because of the anomalous shape of the absorbance *vs* TAN-3,6-S concentration dependence for solutions containing  $3.30 \cdot 10^{-5} \text{M}$   $\text{Cu}^{2+}$ , where the horizontal part is not developed, but a maximum is reached at  $c_L = 1 \cdot 10^{-4} \text{M}$  followed by a slight decrease (Fig. 6 curve 10). This phenomenon is probably connected with the anomalous shape of the concentration dependence for the reagent alone, where absorbances higher than that corresponding to a linear dependence are obtained at higher concentrations, especially at a higher ionic strength (Fig. 6, curves 7–9). Fluctuations in the ionic strength of the solutions affect the absorbance of  $3.30 \cdot 10^{-4} \text{M}$  reagent solutions, especially at low values,  $I < 0.5$ , where the dependence of the reagent absorbance on the ionic strength exponentially increases. The absorbance of the complex ( $\Delta A = A_{\text{total}} - A_{\text{OL}}$ ) first slightly decreases with increasing ionic strength and is then constant from  $I = 0.5$ . Therefore it is suitable to adjust the ionic strength with potassium nitrate to  $I = 0.5$ . The characteristic constants of the calibration curve for the determination of  $\text{Cu}^{2+}$  with TAN-3,6-S, obtained on the MSP 2A computer using the LLSQM program for a  $\text{Cu}^{2+}$  concentration range of 0.017 to 2.80  $\mu\text{g/ml}$  for  $c_L = 3.30 \cdot 10^{-4} \text{M}$ , pH 2.6 (0.05M chloroacetate buffer),  $I = 0.5$  ( $\text{KNO}_3$ ),  $\lambda$  575–590 nm and a 10 mm cuvette, are given in Table IV. The limiting concentrations of some ions, causing an error of  $\pm 2\%$  rel. at a  $\text{Cu}^{2+}$  concentration of 1.37  $\mu\text{g/ml}$  and under the recommended conditions, are summarized in Table V.

Organic solvents decrease the reagent absorbance somewhat and, on the other hand, increase the complex absorbance. For example, the reagent absorbance decreases by 15–20% rel. and that of the complex increases by 5 or 9% rel. in the presence of 20% vol. ethanol or dimethylformamide, respectively.

## DISCUSSION

TAN-3,6-S forms a 1 : 1 chelate with  $\text{Cu}^{2+}$  ions in aqueous solutions with  $c_L < 10^{-4} \text{M}$  and pH from 0 to 9.5, with an absorption maximum at  $\lambda_{\text{max}}$  575 nm, the molar

TABLE IV  
(continued)

$s^b$ μg/ml	$m^c$ μg/ml	Sensitivity <sup>d</sup> μg/cm <sup>2</sup>	$r_k^e$	$U^f$
0.0093	0.0279	0.0304	0.9999	$2.46 \cdot 10^{-4}$
0.0060	0.0176	0.0298	1.0000	$1.02 \cdot 10^{-4}$
0.0068	0.0218	0.0323	0.9999	$1.33 \cdot 10^{-4}$

<sup>a</sup> Section  $q = (\sum y_i - k \sum x_i)/n$ ; <sup>b</sup> standard deviation  $s = 1000 s_{xy}$  at weight Cu/ $\epsilon_{CuL}$ , where  $s_{xy} = \sqrt{[(Y_i - y_i)^2/(n-2)]}$ ,  $x_i$  the concentration values,  $y_i$  and  $Y_i$  the measured and calculated absorbance values, respectively,  $n$  the number of experimental points ( $n = 14$ ); <sup>c</sup> detection limit<sup>12</sup>  $m_s = 3 s$ , simplified for the statistical evaluation of the calibration curve; <sup>d</sup> the sensitivity index according to Sandell<sup>13</sup> (for  $A = 0.010$ ); <sup>e</sup> correlation coefficient<sup>14</sup>  $r_k = [\sum x_i \sum y_i - n \sum x_i y_i] / \sqrt{[(\sum x_i)^2 - n \sum x_i^2][(\sum y_i)^2 - n \sum y_i^2]}$ ; <sup>f</sup> the sum of the squares of the deviations,  $U = \sum (Y_i - y_i)^2$ .

TABLE V  
The Effect of Some Ions on the Determination of  $Cu^{2+}$  with TAN-3,6-S

Taken 1.37 μg/ml  $Cu^{2+}$ ,  $3.30 \cdot 10^{-4}$  M TAN-3,6-S, pH 2.6 (0.05 M chloroacetate buffer), 0.5 (KNO<sub>3</sub>),  $\lambda$  580 nm, 10 mm cuvettes.

Ion	μg/ml <sup>a</sup>	Ion/Cu <sup>b</sup>	Ion	μg/ml	Ion/Cu <sup>b</sup> or M
Bi <sup>3+</sup>	0.115	0.084	Ba <sup>2+</sup>	1 724	1 258
Ni <sup>2+</sup>	0.060	0.044	Mg <sup>2+</sup>	5 382	3 928
Fe <sup>3+</sup>	0.339	0.24	SO <sub>4</sub> <sup>2-</sup>	8 136	0.085
Zn <sup>2+</sup>	1.12	0.82	Cl <sup>-</sup>	6 360	0.18
Cd <sup>2+</sup>	49.3	40.0	Br <sup>-</sup>	11 760	0.15
Cr <sup>3+</sup>	23.4	17.1	PO <sub>4</sub> <sup>3-</sup>	15 270	0.16
Mn <sup>2+</sup>	48.8	35.6	C <sub>2</sub> O <sub>4</sub> <sup>2-</sup> <sup>c</sup>	170	0.0019
Al <sup>3+</sup>	129	94.2	citric acid <sup>c</sup>	12 640	0.066
Ca <sup>2+</sup>	1 700	1 240			
Sr <sup>2+</sup>	532	388	sulphosalicylic acid	7 320	0.029

<sup>a</sup> The amount increasing the absorbance by 2% rel.; <sup>b</sup> the weight ratio given for the cations, the resulting molarity for the anions; <sup>c</sup> absorbance decreased by 2% rel., the other anions increase the absorbance in the same manner as increasing ionic strength.

absorption coefficient  $\varepsilon = 2.44 \cdot 10^4$  (at 575 nm), the equilibrium constant value,  $k_1 = [\text{CuL}][\text{H}]/[\text{Cu}][\text{LH}]$ ,  $\log k_1 = 3.33$  at  $I = 0.1$  and  $\log k_1 = 3.07$  at  $I = 1.0$ , in this case the acidity being expressed in terms of  $(-\log [\text{H}^+])$  and the stability constants,  $\log \beta_{11} = 11.19$  at  $I = 0.1$  and  $\log \beta_{11} = 10.45$  at  $I = 0.1$ . Similar results have been obtained for this reagent and  $\text{Cu}^{2+}$  in a 20% dioxane medium<sup>1</sup>, where a 1 : 1 chelate with  $\lambda_{\text{max}}$  577 nm,  $\varepsilon = 2.23 \cdot 10^4$  and  $\log \beta_{11} = 10.7$  have been found.

The chelate CuL of TAN-3,6-S has properties similar to the chelate of cupric ions with 1-(2-thiazolylazo)-2-naphthol, for which the values,  $\lambda_{\text{max}}$  580 nm,  $\varepsilon = 2.3 \cdot 10^4$  and  $\log k_1 = 3.73$  have been found in 50% dioxane<sup>19</sup>.

The coordination of another ligand particle to the metal atom with formation of a 1 : 2 chelate, which is usually accompanied by a pronounced increase in the molar absorption coefficient (even up to twice the value) with heterocyclic azo-dyes without any substantial change in the absorption maximum, has not been observed in the studied system, even in solutions with excess reagent; however, another, unusual equilibrium was found in these solutions, marked by a decrease in the absorbance

TABLE VI  
Thiazolylazo-Dyes as Reagents for the Spectrophotometric Determination of Cu(II)

Reagent	Complex			$\log k^a$	pH	Medium	Ref.
	M : L	$\lambda_{\text{max}}$ , nm	$\varepsilon \cdot 10^{-4}$ $\text{cm}^2\text{M}^{-1}$				
1-(2-Thiazolylazo)- -2-naphthol-3,6- -disulphonic acid	1 : 1	575	2.13	3.38	2.6	water	—
4-(2-Thiazolylazo)- resorcinol	1 : 1	560	1.94	2.13	2.7—3.4	30% v/v ethanol	15
2-(2-Thiazolylazo)- -4-methoxyphenol	1 : 1	631	1.50	1.66	4.0	1% ethanol and 0.1% PVA	16
	1 : 2	625	3.70		7.1—9.1	extraction into benzene	
2-(4-Methyl- -2-thiazolylazo)- -4-methoxyphenol	1 : 1	634		1.83	5.0	8% v/v methanol	17
	1 : 2	626			8—9	extraction into isopentylalcohol	
2-(2-Thiazolylazo)- -5-dimethylamino- phenol	1 : 2	570	3.94	$-5.6^b$	0.1M- -NaOH	extraction into chloroform	18

<sup>a</sup>  $k = [\text{ML}][\text{H}]/[\text{M}][\text{LH}]$ , <sup>b</sup> the instability constant.

compared to equimolar solutions and solutions with excess metal, also without a perceptible change in the absorption maximum. This equilibrium is most markedly manifested by a decrease in the horizontal parts of the pH curves for  $\text{Cu}^{2+}$  solutions with excess reagent (Fig. 4, curves 6 and 7) and by a decrease of the absorbance of the  $A = f(c_L)$  plot, in the presence of  $\text{Cu}^{2+}$ , starting from  $c_L = 1 \cdot 10^{-4} \text{M}$  (Fig. 6, curve 10). However, the absorbance *vs* concentration plot for the reagent alone has a rather uncommon shape: it exhibits positive deviations from linearity at higher  $c_L$  values (Fig. 6, curves 7–9). The most probable explanation of these equilibria is the assumption of association of the reagent particles in solutions containing more than  $10^{-4} \text{M}$  reagent; this phenomenon has already been described for sulphonated dyes<sup>20</sup>. The decrease in the absorbance in solutions containing  $\text{Cu}^{2+}$  ions (complex  $\text{CuL}$ ) could be caused by reagent association with complex  $\text{CuL}$ , the additional ligand not being coordinated to the  $\text{Cu}^{2+}$  central ion but associated with the ligand bound in the complex, forming a chelate  $\text{Cu}(\text{L} \cdot \text{LH})$  with a lower chromophoric effect of its ligand component. Reliable explanation of these equilibria would require, however, a more extensive experimental background. Therefore, in the spectrophotometric determination of copper with TAN-3,6-S it is desirable to avoid large reagent excess because of these equilibria, even if they do not unfavourably affect the reproducibility of the results.

Comparison of some thiazolylazo-dyes proposed for the spectrophotometric determination of copper (Table VI) and of some other reagents of this type, whose chelates with copper were studied for different purposes<sup>21,22</sup>, shows that TAN-3,6-S ranks among promising reagents for the determination of  $\text{Cu}^{2+}$  because of the relatively high value of the molar absorption coefficient and the equilibrium constant of its chelate with cupric ions. Its main advantage is the solubility and the possibility of its application in aqueous media.

*The authors are very obliged to Professor L. Sommer for valuable discussions and continued interest in this work. They also thank Dr V. Svoboda from the Research Institute of Pure Chemicals, Lachema, Brno, for supplying a sample of TAN-3,6-S.*

#### REFERENCES

1. Kawase A.: Japan Analyst 11, 628 (1962).
2. Kawase A.: Talanta 12, 195 (1965).
3. Busev A. I., Krysina L. S., Zholondkovskaya T. N., Pribylova G. A., Krysina E. P.: Zh. Anal. Khim. 25, 1575 (1970).
4. Busev A. I., Zholondkovskaya T. N., Krysina L. S., Golubkova N. A.: Zh. Anal. Khim. 27, 2165 (1972).
5. Ciba J., Langová M., Kubičková L.: This Journal 38, 3405 (1973).
6. Kubáň V.: Scripta Fac. Sci. Nat. Univ. Brno, in press.
7. Sommer L., Kubáň V., Havel J.: Folia Fac. Sci. Nat. Univ. Brno, Vol. XI (1970), Chemia No 1, 33–62.
8. Kubáň V.: Unpublished results.



9. Kubáň V.: *Scripta Fac. Sci. Nat. Univ. Brno, Chemia* 2, 2, 81 (1972).
10. Havel J., Kubáň V.: *Scripta Fac. Sci. Nat. Univ. Brno, Chemia* 2, 1, 87 (1971).
11. Kubáň V.: Unpublished results.
12. Kaiser H.: *Z. Anal. Chem.* 209, 1 (1965).
13. Sandell E. B.: *Colorimetric Determination of Traces of Metals*. Interscience, New York—London 1959.
14. Gottschalk G.: *Statistik in der Quantitativen Chemischen Analyse*. Enke, Stuttgart 1962.
15. Hniličková M., Sommer L.: *Talanta* 13, 667 (1966).
16. Chromý V., Sommer L.: *Spisy přírodověd. fak. univ. Brno, č.* 517, 407 (1970).
17. Yanagihara T., Matano N., Kawase A.: *Trans. Nat. Res. Inst. Metals* 2, 51 (1960).
18. Minczewski J., Kasiura K.: *Chem. Anal.* 10, 719 (1965).
19. Gusev S. I., Ketova L. A., Glushkova I. N.: *Zh. Anal. Khim.* 25, 2099 (1970).
20. Coates E., Rigg B.: *Trans. Faraday Soc.* 57, 1637 (1961); 58, 2058 (1962); 58, 88 (1962).
21. Langová M., Sommer L.: *Folia Fac. Sci. Nat. Univ. Brno, Vol. IX* (1968), *Chemia* No 2, 1.
22. Anderson R. G., Nickless G.: *Analyst* 92, 207 (1967).

Translated by M. Štulíková.