Ternary Ion-Association Complexes Of Cobalt with 4-(2-pyridylazo)Resorcinol and Tetrazolium Salts

Vidka V. Divarova,[1] Vanya D. Lekova,[1,]* Petya V. Racheva,[2] Kirila T. Stojnova[1] and Atanas N. Dimitrov[1]

[1] *Department of General and Inorganic Chemistry, University of Plovdiv"Paisii Hilendarski",*
24 Tsar Assen Street. 4000 Plovdiv

[2] *Department of Chemistry and Biochemistry, Medical University Plovdiv, 15A Vasil Aprilov Boulevard,*
4002 Plovdiv, Bulgaria

* *Corresponding author: E-mail: vanlek@uni-plovdiv.bg*
tel.: +35932261420

Received: 04-04-2014

Abstract

The formation of ternary ion-association complexes of Co(II) with 4-(2-pyridylazo)resorcinol (PAR) and tetrazolium salts (TS) – (3-(4,5-dimethylThiazol-2-yl)-2,5-diphenyl-2H-tetrazolium bromide (MTT) and 3-(2-naphtyl)-2,5-diphenyl-2H-tetrazolium chloride (TV) – were investigated with the aid of the extraction-spectrophotometric method. The optimum conditions for the extraction of Co(II) were found. On the basis of the experimental data and their mathematical treatment the association process in aqueous phase and extraction process was investigated quantitatively. The molar ratios of the components in the associates were determined. The results show that the extracted species could be represented with the general formula (TS)[CoH(PAR)$_2$]. The following key constants were calculated: association constant, distribution constant, extraction constant and recovery factor. The validity of Beer's law was checked and some analytical characteristics were calculated.

Keywords: Cobalt-PAR chelate; tetrazolium salts; ion-association complexes; spectrophotometry; extraction

1. Introduction

Our investigations carried out by means of tetrazolium salts showed that they could be applied as reagents for extraction-spectrophotometric determination of metals.[1–9] The obtained compounds are ternary ion-association complexes being easily and quickly extracted in the organic phase.

Of interest to us are negatively charged complexes of Co(II) with the azodyestuffs [Co(PAR)$_2$]$^{z-}$ or [Co(TAR)$_2$]$^{z-}$ (z = 1 or 2) interact with onium cations and form ion- associate. [10–27] These compounds are slightly soluble in water and readily in organic solvents.

In the present paper, we described the complex formation and liquid-liquid extraction from water to chloroform in system containing Co(II)–PAR–tetrazolium salts (TS)–H$_2$O–CHCl$_3$, [TS: 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyl-2H-tetrazolium bromide (MTT) and 3-(2-naphtyl)-2,5-diphenyl-2H-tetrazolium chloride (TV)],

with a view to their further application to extraction-spectrophotometric determination of Co(II).

2. Experimental

2. 1. Reagents and Equipment

– CoSO$_4 \cdot$ 7H$_2$O (Sigma-Aldrich, p.a.), 1000 mg L^{-1} aqueous solution. Working solution ($C = 1.7 \times 10^{-4}$ mol L^{-1}) was prepared by dilution.

– PAR (96%, Sigma-Aldrich), dissolved in slightly alkalized distilled water, 2×10^{-3} mol L^{-1}.

– MTT (Merck) – 3.0×10^{-3} mol L^{-1} aqueous solution.

– TV (Fluka) – 3.0×10^{-3} mol L^{-1} aqueous solution.

– The acidity of the aqueous medium was set using the buffer solution prepared by mixing 2.0 mol L^{-1} aqueous solutions of CH$_3$COOH and NH$_4$OH.

– The pH was checked by HI 83140 pH meter (Romania).
– A Camspec M508 spectrophotometer (United Kingdom), equipped with 10 mm path length cells, was employed for reading the absorbance.
– Organic solvent – CHCl$_3$ (additionally distilled).

2. 1. Procedure for Establishing the Optimum Operating Conditions

Aliquots of Co(II) solution, PAR solution, TS solution and buffer solution (pH ranging from 4 to 8) were introduced into separatory funnels. The resulting solutions were diluted with distilled water to a total volume of 10 mL. Then 10 mL of chloroform was added and the funnels were shaken. A portion of the organic extract was filtered through a filter paper into a cell and the absorbance was read against a blank.

2. 3. Procedure for Determination of the Distribution Constants

The distribution constants K_D was found from the ratio $K_D = A_1/(A_3–A_1)$ where, A_1 is the light absorbance for chloroform obtained after a single extraction (at the optimum operating conditions – see Table 2) and A_3 is the absorbance for chloroform obtained after a triple extraction under the same conditions.[27, 28] The single extraction and the first stage of the triple extraction were performed with 10 mL chloroform. The organic layers were transferred into 25 mL calibrated flasks and the flask for the single extraction was brought to volume with chloroform. The second stage of the triple extraction was performed by adding a 7 mL of chloroform to the aqueous phase that remained after the first stage. The third stage was performed in the same manner. The two successive organic layers were transferred to the flask containing the organic layer obtained after the first stage. The volume was brought to the mark with chloroform and shaken for homogenization. Absorbencies A_1 and A_3 were measured against a blank.

3. Results and Discussion

3. 1. Absorption Spectra

The absorption spectra of the investigated ion-association complexes (IAC) Co–PAR–TS in chloroform are shown in Figure 1. As seen from Figure 1 the maxima in both cases are in the λ-range 515–520 nm. They are shifted to 5–10 nm as compared to the maximum of the binary Co–PAR complex in aqueous medium (510 nm; pH = 3.5 ÷ 10.0).[29–36] As optimum wavelength we used $\lambda = 520$ nm in both cases.
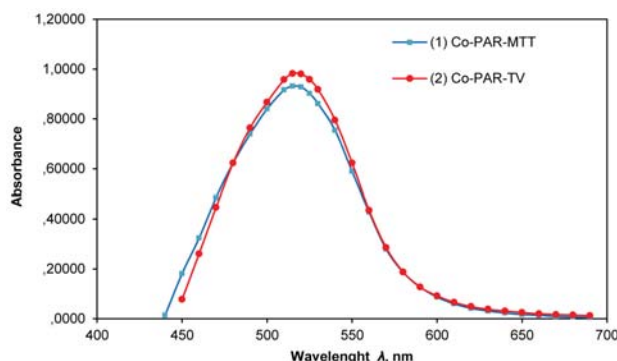

Figure 1. Absorption spectra of the ternary complexes of Co-PAR-MTT and Co-PAR-TV in CHCl$_3$ against blanks: $C_{Co(II)} = 1.7 \times 10^{-5}$ mol L^{-1}; $C_{PAR} = 2.0 \times 10^{-4}$ mol L^{-1}; $C_{MTT} = 3.6 \times 10^{-4}$ mol L^{-1}; $C_{TV} = 3.0 \times 10^{-4}$ mol L^{-1}

3. 2. Effect the Acidity on the Extraction

The most important factor influencing the extraction Co(II) as IAC is acidity of the aqueous phase. Our investigations shown that the IAC of Co–PAR complex with TS are extracted in both cases in a maximum extent at pH 5.0 ÷ 6.0 (Figure 2).
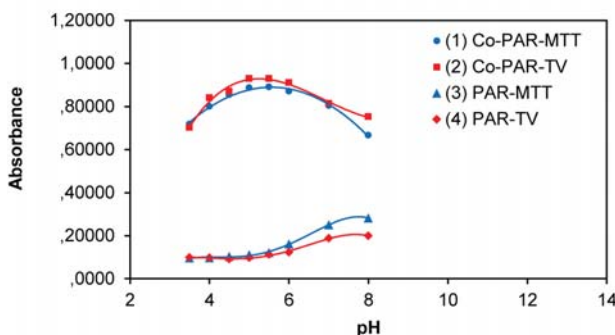

Figure 2. Absorbance of Co-PAR-TS extracts against PAR-TS extracts and absorbance of PAR-TS against CHCl$_3$ vs.pH of the aqueous phase plots: $C_{Co(II)} = 1.7 \times 10^{-5}$ mol L^{-1}; $C_{PAR} = 2.0 \times 10^{-4}$ mol L^{-1}; $C_{MTT} = 3.6 \times 10^{-4}$ mol L^{-1}; $C_{TV} = 3.0 \times 10^{-4}$ mol L^{-1}

3. 3. Effect of Shaking Time

The extraction equilibrium for Co–PAR–TV system is reached for about 30 s and that for Co–PAR–MTT system is reached for about 90 s. In both cases longer shaking time does not affect the absorbance. We extracted in our farther experiments for 2 min in both cases.

3. 4. Effect of Reagents Concentration

The complete bonding of Co(II) into a chelate complex requires a 5–fold excess of PAR for IAC with MTT and 6.5–fold excess for IAC with TV (Figure 3).
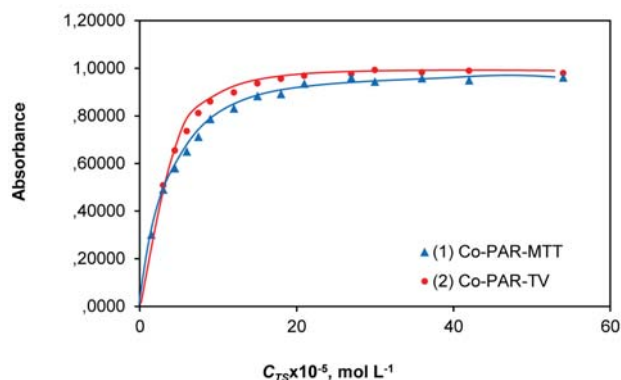
Figure 3. Absorbance of the extracted ternary Co-PAR-TS complexes vs. concentration of the TS plots: $C_{Co(II)} = 1.7 \times 10^{-5}$ mol L^{-1}; $C_{PAR} = 2.0 \times 10^{-4}$ mol L^{-1}
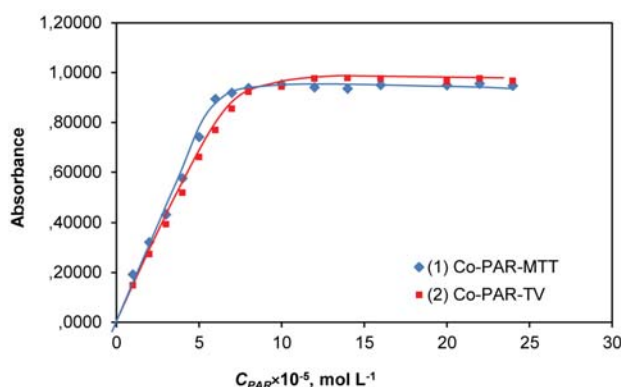

Figure 4. Absorbance of the extracted ternary Co-PAR- TS complexes vs. concentration of the PAR plots: $C_{Co(II)} = 1.7 \times 10^{-5}$ mol L^{-1}; $C_{MTT} = 3.6 \times 10^{-4}$ mol L^{-1}; $C_{TV} = 3.0 \times 10^{-4}$ mol L^{-1}

For a maximum association and extraction the amount of tetrazolium salts should not be lower than a 14–fold excess of MTT and 15–fold excess of TV (Figure 4).

3. 5. Beer's Law and Analytical Characteristics

The dependency between Co(II) concentration in the aqueous phase and the absorbance of the organic layer (corresponding to definite Co(II) concentrations in the aqueous phase) is linear up to 2.6 μg mL^{-1} for Co–PAR–MTT system and up to 2.8 μg mL^{-1} for Co–PAR–TV system,

respectively. Some important analytical characteristics are shown in Table 1.

3. 6. Molar Ratios of the Complexes

By employing the widely used spectrophotometric methods, the mobile equilibrium method and the straight – line method of Asmus it was established that under optimum extraction conditions the molar ratios, n(Co(II)): n(PAR):n(TS), were 1:2:1 (Figures 5, 6, 7, 8). [37, 51]
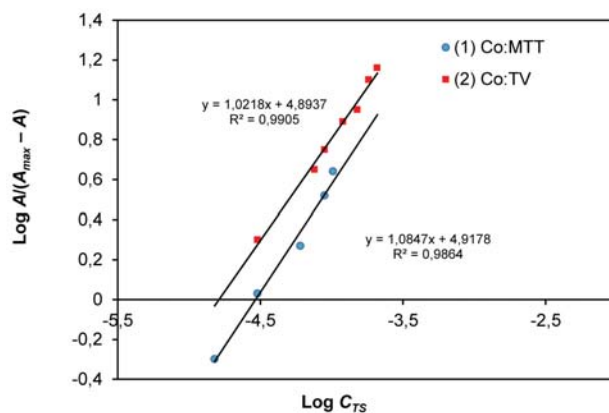

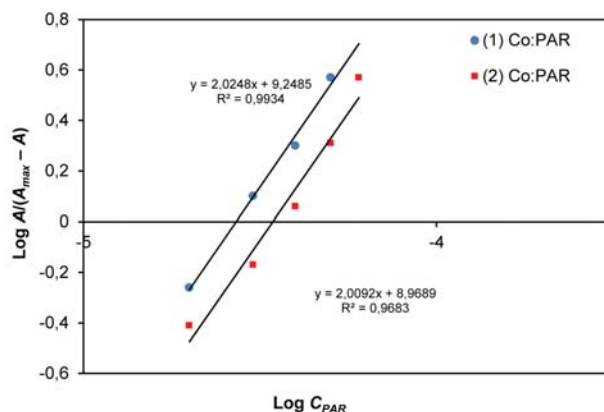**Figure 5.** Straitght lines by the Mobile Equilibrium method during the determination of Co-to-TS

Figure 6. Straitght lines by the Mobile Equilibrium method during the determination of Co-to-PAR (1) Co:PAR (Co-PAR-MTT); (2) Co:PAR (Co-PAR-TV)

Table 1. Characteristics concerning the application of the ternary IAC for extractive-spectrophotometric determination of Co(II)

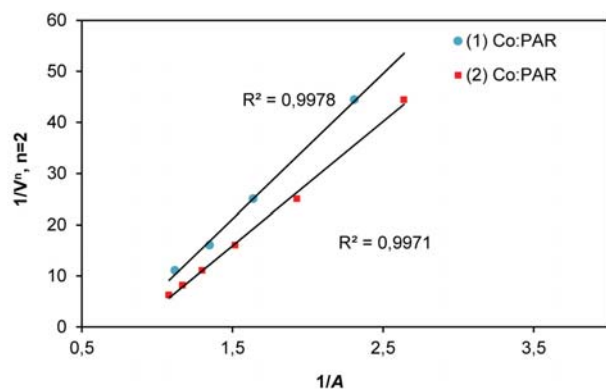| Analytical characteristics	Extraction system	
	Co–PAR–MTT–H$_2$O–CHCl$_3$	Co–PAR–TV–H$_2$O–CHCl$_3$
Apparent molar absorptivity (ε')	5.57×10^4 L mol^{-1} cm^{-1}	6.2×10^4 L mol^{-1} cm^{-1}
Sandell's sensitivity (SS)	1.06 ng cm^{-2}	0.95 ng cm^{-2}
Adherence to Beer's Law	up to 2.6 μg mL^{-1}	up to 2.8 μg mL^{-1}
Limit of detection (LOD)	0.14 μg mL^{-1}	0.07 μg mL^{-1}
Limit of quantification (LOQ)	0.46 μg mL^{-1}	0.25 μg mL^{-1}

Figure 7. Determination of the Co-to-PAR by the Method of Asmus: (1) Co:PAR = 1:2 (Co-PAR-MTT); (2) Co:PAR = 1:2 (Co-PAR-TV)
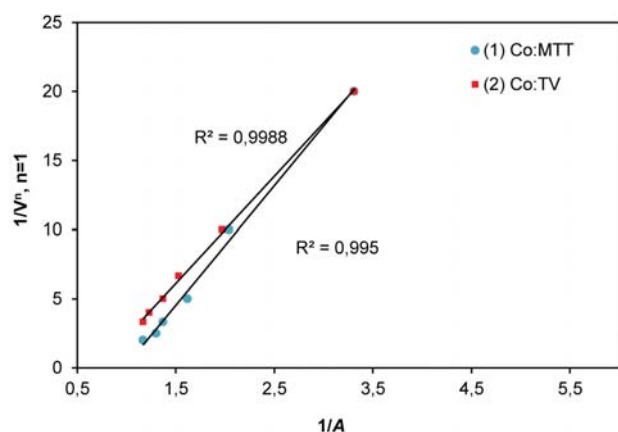

Figure 8. Determination of the Co-to-TS by the method of Asmus: Co:TS = 1:1

The values of correlation coefficient squared are presented in Table 2.

3. 7. Suggested General Formula, Equilibrium Constants, Recovery Factors and Molar Absorptivities

Considering the studies published in the literature, there are two possible schemes of the reaction in the aqu-

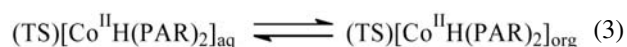eous phase and extraction of the resulting associate.[16,17,24,32, 35,38–50]

Scheme 1

Formation of ion-association complex in the aqueous phase:

$$Co^{2+} + 2\,H_2PAR \longrightarrow [Co^{II}H(PAR)_2]^- + 3\,H^+ \quad (1)$$

$$TS^+ + [Co^{II}H(PAR)_2]^- \rightleftharpoons (TS)[Co^{II}H(PAR)_2] \quad (2)$$

Distribution of the complex between the aqueous and the organic phases:

$$(TS)[Co^{II}H(PAR)_2]_{aq} \rightleftharpoons (TS)[Co^{II}H(PAR)_2]_{org} \quad (3)$$

Extraction from water into chloroform:

$$TS^+_{aq} + [Co^{II}H(PAR)_2]^-_{aq} \rightleftharpoons \atop (TS)[Co^{II}H(PAR)_2]_{org} \quad (4)$$
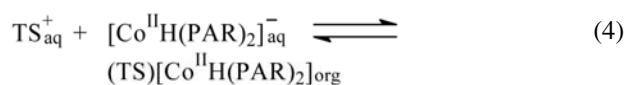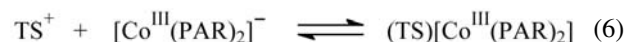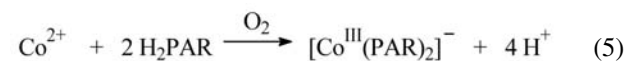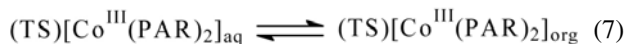
It is possible that in the formation of PAR cobalt chelate, an oxidation to Co(III) occurs, and this oxidation might consume one proton.

Scheme 2

Co(II) oxidation and formation of ion-association complex in the aqueous phase:

$$Co^{2+} + 2\,H_2PAR \xrightarrow{O_2} [Co^{III}(PAR)_2]^- + 4\,H^+ \quad (5)$$

$$TS^+ + [Co^{III}(PAR)_2]^- \rightleftharpoons (TS)[Co^{III}(PAR)_2] \quad (6)$$

Distribution of the complex between the aqueous and the organic phases:

$$(TS)[Co^{III}(PAR)_2]_{aq} \rightleftharpoons (TS)[Co^{III}(PAR)_2]_{org} \quad (7)$$

Extraction from water into chloroform:

$$TS^+_{aq} + [Co^{III}(PAR)_2]^-_{aq} \rightleftharpoons (TS)[Co^{III}(PAR)_2]_{org} \quad (8)$$

On the basis of published data and our unpublished studies we assume that in our conditions the interaction

Table 2. Determination of the Co-to-PAR and Co-to-TS molar ratios (n and m, respectively) by the Asmus method from the experimental data given in Figure 7 and Figure 8, respectively

Extraction system	Correlation coefficient squared values (CC^2) corresponding to molar ratios 1, 2 and 3	
	Co:PAR	**Co:TS**
Co–PAR–MTT–H_2O–$CHCl_3$	(n=1)···CC^2 = 0.9693	(m=1)···CC^2 = 0.9950
	(n=2)···CC^2 = 0.9978	(m=2)···CC^2 = 0.9495
	(n=3)···CC^2 = 0.8925	(m=3)···CC^2 = 0.8936
Co–PAR–TV–H_2O–$CHCl_3$	(n=1)···CC^2 = 0.9671	(m=1)···CC^2 = 0.9988
	(n=2)···CC^2 = 0.9971	(m=2)···CC^2 = 0.9764
	(n=3)···CC^2 = 0.9626	(m=3)···CC^2 = 0.9279

between PAR, cobalt and tetrazolium salts occurs in Scheme 1.

The constants needed for the quantitative assessment of the extraction equilibrium were calculated as follows:

The association constants β and the true molar absorptivities ε by the method of Komar – Tolmachev.[37] (Figure 9);

The distribution constants K_D and the recovery factors $R\%$:

$$K_D = A_1/(A_3 - A_1); \qquad R\% = 100\, K_D/(K_D + 1)$$

where A_1 is the absorption for a single extraction, A_3 – the absorption for a triple extraction;

The extraction constants K_{ex}:

$$K_{ex} = K_D \times \beta; \qquad \text{Log } K_{ex} = \text{Log } K_D + \text{Log } \beta$$

The values obtaining are presented in Table 3.

4. Conclusions

The formation of the ion-association complexes of cobalt with 4-(2-pyridylazo)resorcinol (PAR) and tetrazolium salts (TS) – 3-(4,5-dimethylThiazol-2-yl)-2,5-diphenyl-2H-tetrazolium bromide (thiazolyl blue, MTT) and 3-(2-naphtyl)-2,5-diphenyl-2H-tetrazolium chloride (tetrazolium violete, TV) – was proved by the present study. The bulky molecules of the compounds obtained contributed to their poor solubility in water and good solubility in organic solvents. The optimum conditions for the quanti-
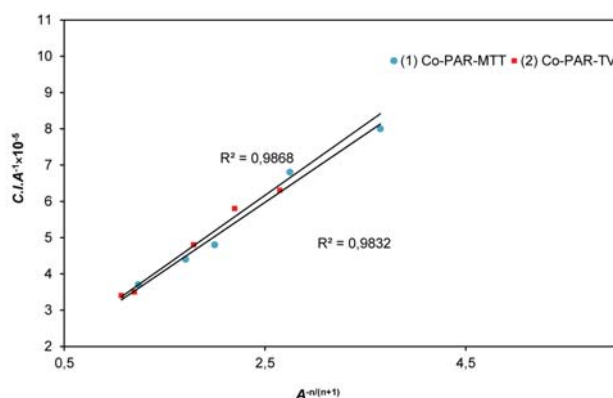
tative liquid-liquid extraction of the ion-associates were found. The equilibrium constants and analytical characteristics, needed for the quantitative assessment of the extraction equilibrium were calculated i.e. constant of association (β), constant of distribution (K_D), constant of extraction (K_{ex}), recovery factors (R), apparent molar absorptivity (ε'), true molar absorptivity (ε), limit of detection (LOD), limit of quantification (LOQ) and Sandell's sensitivity (SS). The results presented in this study will give possibility to develop a method for the analysis of cobalt in suitable objects.

5. Reference

1. A. Alexandrov, O. Budevsky, A. Dimitrov, *J. Radioanal. Chem.* **1976**, *29*, 243–250.
2. A. Dimitrov, A. Alexandrov, A. Hrdlička, M. Vrhlabsky, *Scripta Fac. Sci. Nat. Univ. Purk. Brun.* **1984**, *14*, 91–102.
3. A. Dimitrov, S. Kostova, A. Alexandrov, *Anal. Lab.* **1998**, *7*, 20–23.
4. A. Dimitrov, S. Kostova, J. Pancheva, V. Lekova, *Bulg. Chem. and Ind.* **2002**, *73*, 20–23.
5. K. B. Gavazov, A. N. Dimitrov, V. D. Lekova, *Russ. Chem. Rev.* **2007**, *7*, 169–179.
6. A. N. Dimitrov, V. D. Lekova, K. B. Gavazov, B. S. Boyanov, *J. Anal. Chem.* **2007**, *62*, 122–125.
7. P. Racheva, K. Gavazov, V. Lekova, A. Dimitrov, *J. Anal. Chem.* **2010**, *65*, 21–25.
8. K. Stojnova, K. Gavazov, V. Lekova, *Acta Chim. Slov.* **2013**, *60*, 390–396.
9. K. Gavazov, V. Lekova, G.Patronov, *Acta Chim. Slov.* **2006**, *53*, 506–511.
10. V. A. Lemos, E. S. Santos, M. S. Santos, R. T. Yamaki, *Microchim. Acta* **2007**, *158*, 189 204.
11. R. G. Anderson, G. Nickless, *Analyst* **1967**, *92*, 207–238.
12. P. X. Baliza, S. L. C. Ferreira, L. S. G. Teixeira, *Talanta* **2009**, *79*, 2–9.
13. A. V. Ivanov, *Russ. Chem. Rev.* **1976**, *45*, 213–226.
14. F. Karipcin, E. Kabalcilar, S. Ilican, Y. Caglar, M. Caglar, *Spectrochim. Acta A* **2009**, *73*, 174–180.
15. F. I. Lobanov, G. K. Nurtaeva, E. E. Ergozhin: Extraction of complex metal ions with piridine and oxyazocompounds, Alma-Ata, Nauka, Kazakhstan, **1983**, pp. 94.
16. K. Ueda, *Anal. Lett.* **1978**, *11*, 1009–1021.

Figure 9. Straight lines obtained by the Komar- Tolmachev Method during the determination of the constants of association (β) and the molar absorptivities (ε): Co:TS = 1:1; n=1

Table 3. Values of the constants characterizing the extraction of Cobalt

Extraction system	Log β	Log K_D	Log K_{ex}	R, %	$\varepsilon \times 10^4$, Lmol^{-1}cm^{-1}
Co–PAR–MTT–H$_2$O–CHCl$_3$	4.8 ± 0.3	0.85 ± 0.09	5.6 ± 0.5	91.3 ± 0.2	(5.7 ± 0.2)
Co–PAR–TV–H$_2$O–CHCl$_3$	5.0 ± 0.4	1.03 ± 0.04	6.0 ± 0.4	93.9 ± 0.4	(6.1 ± 0.2)

17. K. Ueda, _Bull. Chem. Soc. Jpn._ **1979**, _52_, 1215–1216.

18. T. Yotsuyanagi, R. Yamashita, K. Aomura, _Jpn. Anal._ **1970**, _19_, 981–982.

19. R. Yamashita, T. Yotsuyanagi, K. Aomura, _Jpn. Anal._ **1971**, _20_, 1282–1288.

20. S. N. Bhadani, M. Tewari, A. Agrawal, C. Sekhar, _J. Indian Chem. Soc._ **1998**, _75_, 176–177.

21. S. G. Mamuliya, I. V. Pyatnitskii, L. L. Kolomiets, K. I. Grigalashvili, _Zh. Anal. Khim._ **1980**, _35_, 1306–1309.

22. A. G. Gaikwad, H. Noguchi, M. Yoshio, _Anal. Lett._ **1991**, _24_, 1625–1641.

23. P. V. Racheva, K. B. Gavazov, V. D. Lekova, A. N. Dimitrov, _J. Mater._ **2013**, _2013_, 7pages, http://dx.doi.org/10.1155/2013/897343

24. L. Marić, M. Široki, Z. Štefanac, M. J. Herak, _J. Inorg. Nucl. Chem._ **1981**, _43_, 3357–3361.

25. O. Navratil, R. W. Frei, _Anal. Chim. Acta_ **1970**, _52_, 221–227.

26. V. V. Divarova, K. B. Gavazov, V. D. Lekova, A. N. Dimitrov, _Chemija_ **2013**, _24_, 81–87.

27. V. Lekova, P. Racheva, K. Stojnova, A. Dimitrov, K. Gavazov, _Chemija_ **2010**, _21_, 106–111.

28. K. B. Gavazov, K. T. Stojnova, T. S. Stefanova, G. K. Toncheva, V. D. Lekova, A. N. Dimitrov, _Chemija_ **2012**, _23_, 278–285.

29. I. V. Pyatnitskii (ed): Analytical Chemistry of Cobalt, Nauka, Moscow, **1965**, pp. 147–149.

30. N. T. Sizonenko, L. V. Gudzenko, _Zav. Lab._**1985**, _51_, 9–10.

31. S. G. Mamuliya, I. V. Pyatnitskii, L. L. Kolomiets, K. I. Grigalashvili, _Zh. Anal. Khim._ **1980**, _35_, 1306–1309.

32. M. Široki, L. Marić, Z. Štefanac, M. J. Herak, _Anal. Chim. Acta_ **1975**, _75_, 101–109.

33. A. I. Busev, V. M. Ivanov, _Zh. Anal. Khim._ **1963**, _18_, 208–215.

34. V. M. Ivanov, N. I. Ershova, V. N. Figurovskaya, A. V. Ivanov, _J. Anal. Chem_ **2001**, _56_, 143–148.

35. F. H. Pollard, P. Hanson, W. J. Geary, _Anal. Chim. Acta_ **1959**, _20_, 26–31.

36. T. Yotsuyanagi, R. Yamashita, K. Aomura, _Anal. Chem._ **1972**, _44_, 1091–1093.

37. M. I. Bulatov, I. P. Kalinkin: Practical Handbook on Photometric Methods of Analysis, Khimiya, Leningrad, **1986**, pp. 239–253.

38. V. Cucinotta, R. Caruzo, A. Giuffrida, M. Messina, G. Maccarrone, A. Torrisi, _J. Chromatogf. A_ **2008**, _1179_, 17–23.

39. T. Okutani, A. Sakuragawa, M. Murakami, _Anal. Sci._ **1991**, _7_, 109–112.

40. J. B. Noffsinger, N. D. Danielson, _J. Liq. Chromatogr. Relat. Technol._ **1986**, _9_, 2165–2183.

41. C. Baiocchi, E. Cantone, A. Marcheto, M. Gennaro, E. Mentasti, C. Carzanini, _Chromatographia_ **1987**, _23_, 736–746.

42. A. G. Gaikwad, H. Noguchi, M. Yoshio, _Anal. Lett._ **1991**, _24_, 1625–1641.

43. M. S. Sastary, R. Ghose, _Bull. Chem. Soc. Ethiopia_ **1990**, _4_, 61.

44. A. Corsini, Q. Fernando, H. Freiser, _Inorg. Chem._ **1963**, _2_, 224–226.

45. N. A. Nassir, _Ir. Natl J. Chem._ **2007**, _26_, 263–269.

46. J. Dolezal, L. Sommer, _Collect. Czech. Commun._ **1994**, _59_, 2209–2226.

47. F. Karipcin, E. Kabalcilar, _Acta Chim. Slov._ **2007**, _54_, 242–247.

48. B. D. Öztürk, H. Filik, E. Tütem, R. Apak, _Talanta_ **2000**, _53_, 263–269.

49. M. I. Toral, P. Richter, L. Silva, _Talanta_ **1993**, _40_, 1405–1409.

50. Z. Sosa, J. P. Trujillo, J. J. Arias, F. G. Montelongo, _Polyhedron_ **1987**, _6_, 1993–1997.

51. E. Asmus, _Fresen. J. Anal. Chem._ **1960**, _178_, 104–116.

Povzetek

Proučevana je bila tvorba in ekstrakcija ionskih asociatnih kompleksov med Co(II), 4-(2-piridilazo)resorcinolom (PAR) in tetrazolijevimi solmi (TS): (3-(4,5-dimetilthiazol-2-il)-2,5-difenil-2_H_-tetrazolijev bromid (MTT) oziroma 3-(2-naftil)-2,5-difenil-2_H_-tetrazolijev klorid (TV). Izvedena je bila optimizacija pogojev za ekstrakcijo Co(II). Na podlagi eksperimentalnih podatkov in njihove matematične obdelave so bili kvantitativno proučeni procesi asociacije v vodni fazi ter ekstrakcijski procesi. Množinska razmerja komponent v asociatu so bila določena. Na podlagi rezultatov smo dokazali, da ima ekstrahirana zvrst splošno formulo (TS)[CoH(PAR)₂]. Sledeče konstante so bile določene: konstanta asociacije, konstanta distribucije, konstanta ekstrakcije in druge. Preverjena je bila veljavnost Beerovega zakona in izračunane nekatere analizne karakteristike.