Complexation of p*-Sulphonato-calix[6]arene by Glycine, Glycyl-glycine, and Glycyl-glycyl-glycine in Aqueous Solution*

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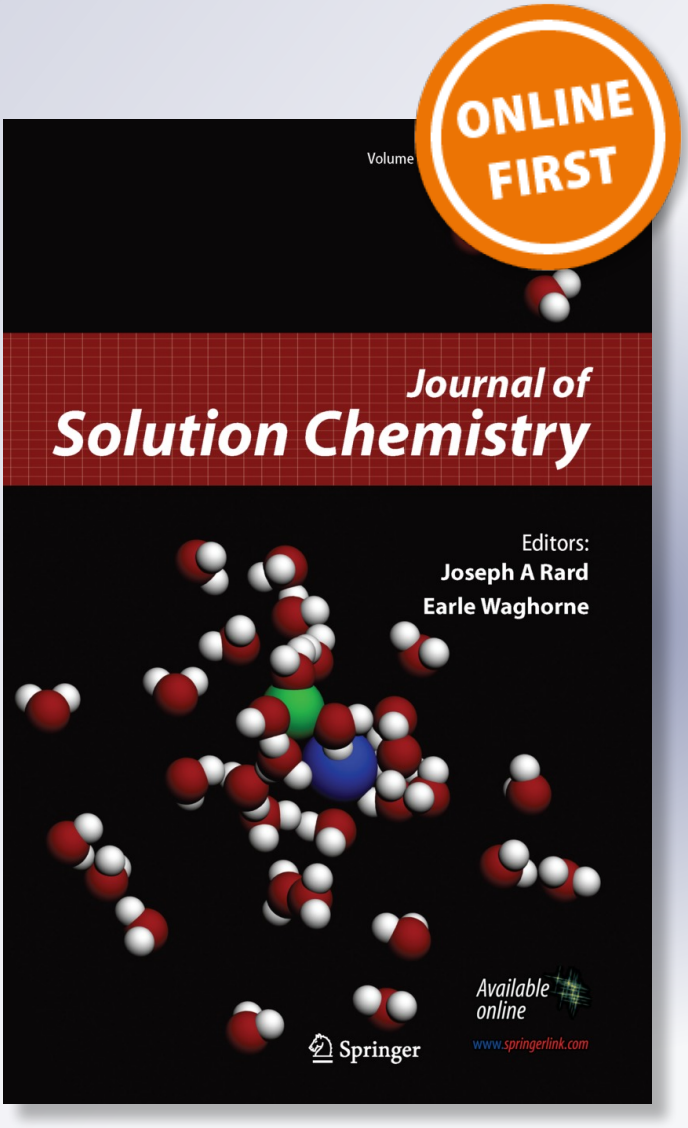

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Complexation of *p*-Sulphonato-calix[6]arene by Glycine, Glycyl-glycine, and Glycyl-glycyl-glycine in Aqueous Solution

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Abstract The complexing ability of *p*-sulphonato-calix[6]arene towards glycine, glycyl-glycine, and glycyl-glycyl-glycine has been evaluated at pH = 1.8 and 7.9 using UV–Vis spectrophotometry. At these pHs the guest molecules are in their cationic and zwitterionic forms, respectively. The results showed that the host is capable of complexing with the guests in 1:1 guest-to-host ratios. Formation constants of the systems have been determined at different temperatures (20 ± 0.1 to 40 ± 0.1 °C). Considering the formation constant values, the binding selectivity of the host towards the guests is in the order glycyl-glycyl-glycine > glycyl-glycine > glycine. The thermodynamic parameters have been evaluated and are interpreted in terms of the importance of the various interactions responsible for the complexation. A roughly linear relationship between ΔH^{o} and $T\Delta S^{o}$ has been observed for the studied systems and is discussed.

Keywords *p*-Sulphonato-calix[6]arene · Glycine · Glycyl-glycine · Glycyl-glycyl-glycine · Formation constant

1 Introduction

Biological activity studies concerning *p*-sulphonato-calix[*n*]arenes were started by the initial work of Atwood on the chloride ion-channel blocking properties of the sodium salt of *p*-sulphonato-calix[4]arene [1]. During the last two decades, the bio-activity of

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sulphonato-calix[*n*]arenes has attracted a lot of attention because of their potential applications in various fields. Several reports have been published on the use of this compound as molecular skeletons for the development of bio-mimetic systems [2–5], studying their interaction with some biological guests like amino acids [6], peptides [7], proteins [8, 9], DNA [10], etc. Controlling the size of the molecule (by changing the value of *n*) and introduction of various functional groups makes it useful for a variety of applications, such as: catalysts, slow releasers of drugs, anti-viral activity, enzyme inhibition property, etc. [11–14].

The dependence of formation constants and thermodynamic parameters ($\Delta H°$ and $\Delta S°$) on host and guest structures provides a useful tool to elucidate the factors governing the complexation. In fact, $\Delta H°$ and $\Delta S°$ values are interpreted in terms of importance of the various interactions responsible for complexation and analyzed in the context of the enthalpy-entropy compensation effect. However, the thermodynamic parameters and the nature of complex formation between *p*-sulphonato-calix[*n*]arenes and amino acids or peptides are usually determined by means of a calorimetric titration [7], but in the present work we measured them by spectrophotometric titration at different temperatures. The results of this work may contribute to the chiral separation studies of amino acids with *p*-sulphonato-calix[*n*]arenes.

In our previous report, various types of calix[4]arene molecules have been synthesized and tested as potential ligands for studies of complexation with different mono and divalent cations [15–23]. In the present work the complexation ability of *p*-sulphonato-calix[6]arene towards glycine (gly), glycyl-glycine (gly-gly), and glycyl-glycyl-glycine (gly-gly-gly) are reported in aqueous solution at two pH values (1.8 and 7.9) and different temperatures (20 ± 0.1 to 40 ± 0.1 °C).

2 Experimental

2.1 Chemicals

p-Sulphonato-calix[6]arene was received from the Louis Pasteur University, France, (gratefully acknowledged), while gly, gly-gly, and gly-gly-gly were purchased from Merck (p.a.) and used without further purification. All dilute solutions were prepared from double distilled water with a specific conductance equal to 1.2 ± 0.1 $\mu S \cdot cm^{-1}$.

2.2 Measurements

A Jenway research pH-meter (model 3520) was used for the pH measurements. The hydrogen ion concentration was measured with a Jenway combination electrode. The pH-meter was calibrated with Metrohm pH = 4.0 and 7.0 buffers leading to pH estimated errors of ±0.001 pH units.

Spectrophotometric measurements were performed using a UV–Vis Shimadzu 2100 scanning spectrophotometer with a Pentium 4 computer using 10 mm quartz cells. The system was thermostated at the desired temperature by circulating water from an isothermal bath. For each experiment, a 2 mL solution of SC6, (2.0–4.0) × 10^{-4} $mol \cdot dm^{-3}$, was titrated with stepwise addition of 2 mL of the guest solutions, (0.2–2.0) × 10^{-3} $mol \cdot dm^{-3}$, both of the same pH (1.8). The procedure was repeated at pH = 7.9 and at different temperatures (20 ± 0.1 to 40 ± 0.1 °C). The pH of solutions was controlled by adding an appropriate volume of a diluted hydrochloric acid or sodium hydroxide to the test solutions to acquire the desired pH. In the titration procedure after addition of a few drops of titrant, the absorbance

was measured in the range 250–350 nm (in the interval of 1 nm) and the procedure continued until the ratio of the guest to the host concentrations reached 2. The UV–Vis spectra of the mixtures undergo small changes but the measured absorbances are sufficient to allow the treatment of the data with the computer program. In all cases, the procedure was repeated at least three times, and the resulting average values and corresponding deviations from the average are shown in the text and Tables. To exclude carbon dioxide from the system, a stream of purified nitrogen was passed through a sodium hydroxide solution and then bubbled slowly through the reaction solution.

3 Results and Discussion

Assuming that the absorbance of sulphonato-calix[6]arene would change upon complexation with a guest molecule, we performed spectrophotometric measurements. The complex $SC6_pL_q$ formed is characterized by its stoichiometry, p and q, where L represents the amino acid or peptide. To determine the formation constant of complexation, K, Eq. 1 is defined,

$$pSC6 + qL \rightleftarrows SC6_pL_q \qquad K = \left[SC6_pL_q\right]/[SC6]^p[L]^q \tag{1}$$

Determination of the formation constant was carried out using the method described previously [23]. Absorbance, A, was measured after successive additions of the amino acid or peptide solution to the sulphonato-calix[6]arene solution. The absorption bands of sulphonato-calix[6]arene decreased upon addition of the amino acid or peptide in all cases. The changes of the absorbance are the result of dilution due to the titration procedure and complex formation because the extinction coefficient of the complex is different from that of the sulphonato-calix[6]arene. Treatment of the spectrophotometric data obtained during the titrations was conducted with the computer program Star [24].

Considering Eq. 1, different models including SC6L, $SC6_2L$, and $SC6L_2$ species were tested by the program. Taking into account $SC6_2L$ alone or together with SC6L does not improve the quality of the fit and leads to the rejection of the model. The model finally chosen, formed by SC6L, resulted in a satisfactory numerical and graphical fitting for the studied systems. The formation constant of the 1:1 complex species are listed in Table 1 at different temperatures and pHs.

The interesting curves resulting from the spectrophotometric titration of SC6 with the amino acid or each peptide at pH = 1.8 are shown in Fig. 1. The curves show an almost sharp break point when the ratio of the concentration of L to the SC6 reaches unity, indicating the formation of a stable complex species for gly-gly-gly. However, the same titrations for gly-gly and gly show absorbance increases with a small and more continuous variation with concentration ratios in the complexation curves, indicating a lower stability constant. This behavior is typical for less stable complexes than that found for the former species. In these cases extrapolating the slopes at high and low L to SC6 ratios corresponds to 1:1 complex stoichiometry at the point of intersection. The same trend for the amino acid and the peptides was observed at pH = 7.9 (not shown).

The dependence of formation constant and thermodynamic parameters (ΔH° and ΔS°) on the host and guest structures provides a tool to elucidate the factors governing the complexation.

It is seen from Table 1 that SC6 forms relatively strong complexes with the guest molecules in this work. The formation constants of the complexations increase from gly to gly-gly-gly. This could be due to the size of the guest molecules. It can thus be concluded

Table 1 Average values of $\log_{10} K$ for *p*-sulphonato-calix[6]arene-gly, gly-gly, and gly-gly-gly at different temperatures together with formation constants of some other amino acids or peptides with SC6 reported in the literature for comparison

Temperature/°C[a]	Guest molecule	$\log_{10} K$ pH = 1.8	$\log_{10} K$ pH = 7.9	References
20	Gly	2.15 ± 0.05	1.75 ± 0.04	This work
25	Gly	2.11 ± 0.04	1.72 ± 0.03	This work
30	Gly	2.04 ± 0.06	1.67 ± 0.06	This work
35	Gly	1.99 ± 0.05	1.59 ± 0.06	This work
40	Gly	1.96 ± 0.08	1.54 ± 0.09	This work
20	Gly-gly	2.43 ± 0.04	1.92 ± 0.03	This work
25	Gly-gly	2.39 ± 0.05	1.87 ± 0.03	This work
30	Gly-gly	2.31 ± 0.06	1.83 ± 0.05	This work
35	Gly-gly	2.25 ± 0.05	1.74 ± 0.06	This work
40	Gly-gly	2.22 ± 0.09	1.69 ± 0.08	This work
20	Gly-gly-gly	3.15 ± 0.02	2.92 ± 0.03	This work
25	Gly-gly-gly	3.09 ± 0.04	2.83 ± 0.03	This work
30	Gly-gly-gly	3.04 ± 0.03	2.76 ± 0.06	This work
35	Gly-gly-gly	2.91 ± 0.04	2.69 ± 0.05	This work
40	Gly-gly-gly	2.90 ± 0.10	2.61 ± 0.08	This work
	Arg	1.65 (pH = 1.0)		[25]
	Lys	1.25 (pH = 1.0)		[25]
	Gly-ala	3.21		[7]
	Gly-val	3.24		[7]
	Gly-leu	3.22		[7]
	Leu-ala	3.23		[7]
	Gly-ph-ala	3.23		[7]
	Arg	2.27 (pH = 8.0)		[25]
	Lys	1.97 (pH = 8.0)		[25]

[a] Uncertainties in controlling the temperatures is 0.1 °C

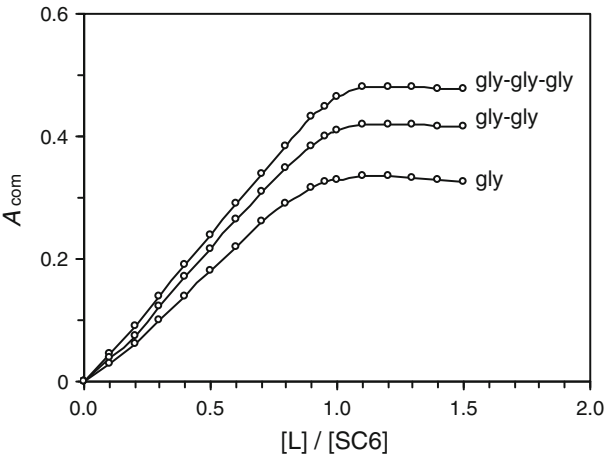

Fig. 1 Spectrophotometric titration plots, $A_{complex}$ versus the mole ratios of the guests to *p*-sulphonato-calix[6]arene at 25 ± 0.1 °C, 290 nm, and pH = 1.8

that gly-gly-gly is included in the calixarene cavity in contrast to gly which remains outside the cavity. As can be seen in Table 2, changing the pH from 7.9 to 1.8 causes more stable complexes to form between SC6 and the guest molecules. This is possibly due to disappearance of the repulsion between the sulphonato groups of SC6 and the carboxy group of the amino acid and the peptides at pH = 7.9, when the medium becomes acidic. It is well known that the following species of the amino acid and the peptides exist in solution at different pH, L^-, $HL^{\pm}$, and H_2L^+, where L^- represents the fully dissociated form of gly and each peptide. At pH = 1.8 and 7.9 the guest molecules have their cationic and zwitterionic forms, respectively. So, favorable interactions between the positively charged amino acid and peptides and the negatively charged SC6 are involved. In fact, in the absence of this particular repulsion the guest can penetrate more deeply into the calixarene cavity and so give stronger complexes.

Table 1 shows that the host–guest formation constant decreases on increasing the temperature (from 20 ± 0.1 to 40 ± 0.1 °C) in all cases. This implies a negative value of $\Delta H°$. The values of $\Delta H°$ and $\Delta S°$ determined from the slope and the intercept of the straight line of $\log_{10}K$ versus $1/T$, and are listed in Table 2 both at pH = 1.8 and 7.9. These findings suggest that the binding of SC6 by the guest molecules in water is enthalpy-driven. In similar works the same type of results were obtained in complexation with some other amino acids or peptides [7, 25–27]. It can thus be concluded that larger guest molecules are included in the calixarene cavity in contrast to the smaller molecules which remains outside the cavity and bind with the SO_3^- groups by ionic interactions at the upper rim of SC6. A literature survey shows that many complexation processes involving amino acids or peptides show similar thermodynamic behavior towards sulphonato-calixarenes [7, 25–27]. Further, according to Smithrud et al. [28], a large part of the favorable enthalpy change results from solvent-specific contributions. Their calorimetric study in 12 different solvents with various polarities shows that water is not special in providing an enthalpic driving force for apolar complexation. Their results suggest that the enthalpic driving force for tight apolar inclusion increases with increasing polarity, becoming strongest in polar protic solvents, and ultimately in water.

Table 2 Thermodynamic parameters ($\Delta G°$, $\Delta H°$, $\Delta S°$) for the binding of SC6 with gly, gly-gly, and gly-gly-gly at 25 ± 0.1 °C together with some values reported in the literature

Guest molecule	$-\Delta G°$/kJ·mol^{-1}		$-\Delta H°$/kJ·mol^{-1}		$-T\Delta S°$/kJ·mol^{-1}		References
	pH = 1.8	pH = 7.9	pH = 1.8	pH = 7.9	pH = 1.8	pH = 7.9	
Gly	12.0 ± 0.2	9.7 ± 0.1	17.6 ± 0.2	19.3 ± 0.1	5.6 ± 0.1	9.5 ± 0.3	This work
Gly-gly	13.6 ± 0.1	10.7 ± 0.1	19.7 ± 0.3	20.7 ± 0.1	6.2 ± 0.2	10.1 ± 0.3	This work
Gly-gly-gly	17.6 ± 0.3	16.2 ± 0.2	23.9 ± 0.2	26.7 ± 0.3	6.3 ± 0.2	10.5 ± 0.2	This work
Arg			48.2 (pH = 1.0)		38.7 (pH = 1.0)		[25]
Lys			27.1 (pH = 1.0)		19.8 (pH = 1.0)		[25]
Gly-ala			38.2		18.3		[7]
Gly-val			40.7		22.3		[7]
Gly-leu			60.6		42.3		[7]
Leu-ala			52.7		34.6		[7]
Gly-ph-ala			46.9		28.1		[7]
Arg			41.2 (pH = 8.0)		28.2 (pH = 8.0)		[25]
Lys			21.8 (pH = 8.0)		10.4 (pH = 8.0)		[25]

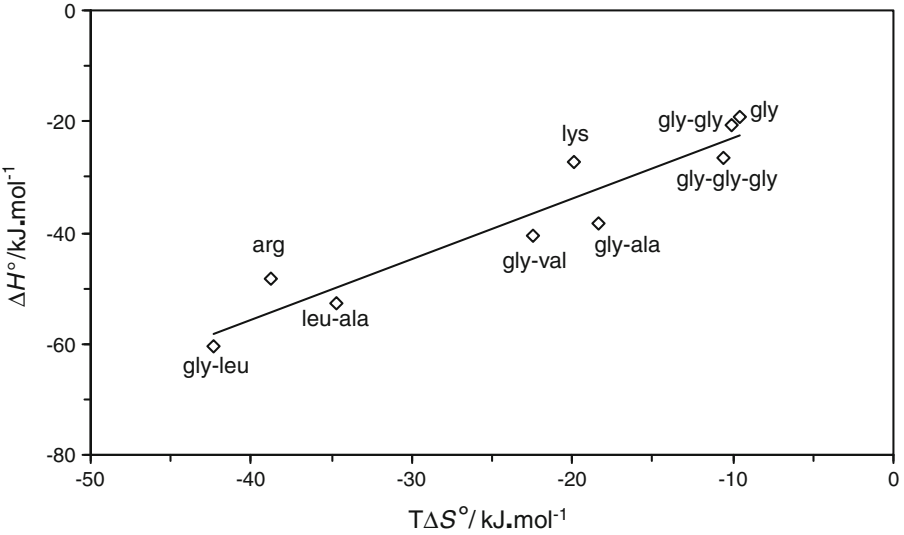

Fig. 2 The plot of ΔH° versus $T\Delta S^\circ$ for the binding of *p*-sulphonato-calix[6]arene with some guest molecules

One of the most important parameters showing the nature of the intermolecular host–guest interaction is the enthalpy change of a reaction. The enthalpy change results from several factors including hydrogen bonding, electrostatic interactions, and van der Waals forces. The entropy change also consists of several factors. In an entropy-driven reaction, the entropy gain is possibly due to the loss of the arrangement of water molecules originally surrounding the organic molecules that are in a highly ordered state. However, in an enthalpy-driven reaction, the entropy loss is possibly due to the freezing of motional freedom of the guest molecule as a result of association with the host species. Table 2 shows that the binding of SC6 by gly-gly-gly is more exothermic than gly and the entropy change decreases from gly to gly-gly-gly, indicating more stable complexes between SC6 and the larger guest molecule, in accordance with the previous discussion. Further, the ΔG° of complexations between SC6 and gly, gly-gly, and gly-gly-gly are not very different from each other, Table 2. This indicates that the factors governing the complexation of the guests are possibly the same.

In Fig. 2, we have plotted ΔH° against $T\Delta S^\circ$ for complexation of two families of guest molecules (amino acids and peptides) with the host molecule SC6. A roughly linear relationship is observed between the values obtained in this work and those reported in the literature using different methods to determine the formation constants and the thermodynamics parameters. Such a linear relationship implies that the change in $T\Delta S^\circ$ is proportional to the corresponding change in ΔH° [24]. Although the origin of this compensatory effect is not entirely clear, it is not unreasonable to think that, as the host–guest interactions become stronger, the degrees of freedom of the resulting complex will be significantly reduced due to the increased rigidity of the system. As a result, part of the enthalpic gain will be cancelled by an entropic loss. Similarly, as the host–guest interactions become weaker, the corresponding enthalpic loss will be partially compensated by a simultaneous entropic gain due to the greater degrees of freedom of the resulting complex.

References

1. Atwood, J.L., Bridges, R.J., Juneja, R.K., Singh, A.K.: Calixarene chloride-channel blockers. US Patent Number 5,489,612 (1996)
2. Lin, Q., Park, H.S., Hamuro, Y., Lee, C.S., Hamilton, A.D.: Protein surface recognition by synthetic agents: Design and structural requirements of a family of artificial receptors that bind to cytochrome c. Biopolymers **47**, 285–297 (1998)
3. Park, H.S., Lin, Q., Hamilton, A.D.: Protein surface recognition by synthetic receptors: A route to novel submicromolar inhibitors for α-chymotrypsin. J. Am. Chem. Soc. **121**, 8–13 (1999)
4. Hamuro, Y., Calama, M.C., Park, H.S., Hamilton, A.D.: Calixarene with four peptide loops: An antibody mimic for recognition of protein surfaces. Angew. Chem. **36**, 2680–2683 (1997)
5. Da Silva, E., Nouar, R., Coleman, A.W., Rather, B., Zaworotko, M.J.: Solution and solid-state interactions between para-sulphonato-calix[4]arene and some common organic biological buffers. J. Incl. Phenom. Macrocycl. Chem. **46**, 161–166 (2003)
6. Kalchenko, O.I., Da Silva, E., Coleman, A.W.: Determination of the stability constants of inclusion complexes of *p*-H-37-(2-carboxy-methyloxy)-calix-[6]-arene and *p*-sulphonato-37-(2-carboxy-methyloxy)-calix-[6]-arene with 15 amino acids by RP-HPLC. J. Incl. Phenom. Macrocycl. Chem. **43**, 305–310 (2002)
7. Buschmann, H.J., Mutihac, L., Schollmeyer, E.: Complexation of some amino acids and peptides by *p*-sulphonato-calix[4]arene and hexasodium *p*-sulphonato-calix[6]arene in aqueous solution. J. Incl. Phenom. Macrocycl. Chem. **46**, 133–137 (2003)
8. Aime, S., Barge, A., Botta, M., Casnati, A., Fragai, M., Luchinat, C., Ungaro, R.: A calix[4]arene GdIII complex endowed with high stability, relaxivity, and binding affinity to serum albumin. Angew. Chem. **40**, 4737–4739 (2001)
9. Memmi, L., Lazar, A., Brioude, A., Ball, V., Coleman, A.W.: Protein–calixarene interactions: Complexation of bovine serum albumin by sulfonatocalix[n]arenes. J. Chem. Soc. Chem. Commun. **23**, 2474–2475 (2001)
10. Shi, Y., Schneider, H.J.: Interaction between aminocalixarenes and nucleotides or nucleic acids. J. Chem. Soc. Perkin Trans. **2**, 1797–1803 (1999)
11. Hwang, K.M., Qi, Y.M., Liu, S.Y., Lee, T.C., Choy, W., Chen, J.: Antithrombotic treatment with calix(n)arene compounds. US Patent Number 5,409,959 (1995)
12. Hwang, K.M., Qi, Y.M., Liu, S.Y., Lee, T.C., Choy, W., Chen, J.: Macrocyclic anti-viral compound and method. US Patent Number 5,196,452 (1991)
13. Harris, S.J.: Preparation of calixarenes as anti-viral compounds. PCT Int. Appl. WO 0244121 (2002)
14. Hulmes, D.J.S., Aubert-Foucher, E., Coleman, A.W.: Use of calix(n)arenes for treating fibrotic diseases. French Patent 98.10074 (1998)
15. Gharib, F., Zare, K., Taghvaei, S., Monajjemi, M., Amiri, A.: Complexation of tri-*o*-propoyl-*p*-t-butyl calix[4]arene with alkali metal cations in carbon tetrachloride solvent. Main Group Met. Chem. **26**, 255–258 (2003)
16. Gharib, F., Zare, K., Amiri, A.: Spectrophotometric studies of alkali metal ion complexes of *p*-tetrabutyl-calix[4]arenes. Russ. J. Inorg. Chem. **49**, 1511–1514 (2004)
17. Gharib, F., Taghvaei-Ganjali, S., Eslamipanah, M., Mazooji, R., Ebrahimi, S.: Complexation of di and tetra-benzyloxy ether derivatives of calix[4]arene with alkali metal cations. Acta Chim. Slov. **53**, 424–427 (2006)
18. Gharib, F., Taghvaei, S., Ghazi, S.: Complexation of di-benzyloxy ether derivatives of calix[4]arene with alkali earth metal cations. Russ. J. Inorg. Chem. **52**, 1915–1917 (2007)
19. Gharib, F., Shakarami, M., Taghvaei-Ganjali, S.: Synthesis and complexive abilities in cone conformation of lower rim functionalized calix[4]arene with alkali metal cations. Acta Chim. Slov. **55**, 570–574 (2008)
20. Gharib, F., Osouleddini, N., Zare, K., Taghvaei-Ganjali, S.: Spectrophotometric studies of alkali and alkali earth metal ions complexes of mono amino derivative of calix[4]arene. Russ. J. Inorg. Chem. **55**, 434–437 (2010)
21. Gharib, F., Taghvaei, S., Eslamipanah, M.: Complexation of di-benzyloxy ether calix[4]arene with alkali metal cations. Main Group Met. Chem. **33**, 315–320 (2010)
22. Gharib, F., Zare, K., Osouleddini, N.: Synthesis and complexive abilities in cone conformation of upper rim functionalized of calix[4]arene with alkali and alkali earth cations. Main Group Met. Chem. **34**, 47–51 (2011)
23. Gharib, F., Hajmalek, M., Ahmadi Alamoti, R., Farajtabar, A.: Complexation of dioxovanadium(V) and dioxouranium(VI) by *p*-sulphonato-calix[4]arene in aqueous solution. J. Mol. Liq. **159**, 161–164 (2011)

24. Beltran, J.L., Codony, R., Prat, M.D.: Evaluation of stability constants from multi-wavelength absorbance data program Star. Anal. Chim. Acta **276**, 441–454 (1993)
25. Douteau-Guevel, N., Coleman, A.W., Morel, J.P., Morel-Desrosiers, N.: Complexation of the basic amino acids lysine and arginine by three sulphonato-calix[*n*]arenes ($n = 4$, 6, and 8) in water: microcalorimetric determination of the Gibbs energies, enthalpies and entropies of complexation. J. Chem. Soc. Perkin Trans. **2**, 629–633 (1999)
26. Da Silva, E., Coleman, A.W.: Synthesis and complexation properties towards amino acids of mono-substituted *p*-sulphonato-calix[*n*]arenes. Tetrahedron **59**, 7357–7364 (2003)
27. Mutihac, L., Buschmann, H.J., Mutihac, R.C., Schollmeyer, E.: Complexation and separation of amines, amino acids, and peptides by functionalized calix[*n*]arenes. J. Incl. Phenom. Macrocycl. Chem. **51**, 1–10 (2005)
28. Smithrud, D., Wyman, T.B., Diederich, F.: Enthalpically driven cyclophane–arene inclusion complexation: Solvent-dependent calorimetric studies. J. Am. Chem. Soc. **113**, 5420–5426 (1991)