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# Physico-chemical properties of the $C_{60}$ -arginine water solutions

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#### ABSTRACT

The paper presents experimental data on physico-chemical study of the  $C_{60}$ -arginine derivative water solutions. The data on temperature dependence of solubility in water, concentration dependence of density, specific conductivity, molar conductivity, dissociation constant, and dynamic light scattering are presented and characterized.

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## 1. Introduction

Water-soluble derivatives of fullerenes have great potential due to unique structural, physical, and chemical properties and especially due to possibilities of practical application in various fields of science and technology in particular in biology and medicine due to promising biological activity of such compounds [1–13].

The present paper is devoted to physico-chemical investigation of water solutions of the  $C_{60}\text{-L-arginine}$  derivative (see Fig. 1) -  $C_{60}(C_6H_{13}N_4O_2)_8H_8$  (i.e. isothermal and temperature dependences of densities, concentration dependence of the refractive index, specific and equivalent conductivities, nanoparticle size distribution in water as well as to investigation of solubility of the  $C_{60}$ -arginine derivative in water).

The actuality of the amino acid derivative investigation is mainly connected with the variety of application methods in biology and medicine. Let's dwell briefly on some typical results.

Kotelnikova et al. studied the modifying effects of the water-soluble  $C_{60}$  derivatives with DL-alanine and DL-alanyl-DL-alanine on the structure and permeability of the lipid bilayer of phosphatidylcholine liposomes

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[14]. It was found that synthesized derivatives are able to localize inside the artificial membrane, to penetrate into the liposomes through the lipid bilayer and to perform activated transmembrane transport of bivalent metal ions.

Hu et al. synthesized and characterized amphiphilic amino acid derivatives of  $C_{60}$  with  $\beta$ -alanine, cystine and arginine [15]. It was determined that these compounds are able to penetrate the cell membrane and reduce the accumulation of reactive oxygen species and cellular damage caused by hydrogen peroxide in PC12 cells. The authors of [15] concluded that the  $C_{60}$  amino acid derivatives have the potential to prevent oxidative stress-induced cell death without evident toxicity.

Kumar et al. synthesized and studied biological properties of the  $C_{60}$ -lysine derivative [16]. The synthesized compound exhibited high DNA cleavage efficiency upon visible light irradiation in the presence of NADH.

Jiang et al. synthesized novel water-soluble  $C_{60}$ -glycine derivative [17]. The cytotoxicity to cancer cell lines of the  $C_{60}$ -glycine derivatives was evaluated by MTT (3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyl tetrazolium bromide) and flow cytometry for the first time. The results show that  $C_{60}$ -glycine derivative exhibit mortality and apoptosis of the cells which increased with the increase of  $C_{60}$ -glycine derivative concentration.

Series of articles present the experimental data on neuroprotective action of hybrid structures based on fullerene  $C_{60}$  with attached proline

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**Fig. 1.** Adduct of the C<sub>60</sub> fullerene with μ-arginine.

amino acid (methyl ether of N-[( $\beta$ -alanyl-histidyl-ethyl) fullerenyl] proline, methyl ether of N-[(nitroxy-ethyl)fullerenyl] proline, methyl ether of N-[(2',3'-dinitroxy-propyl) fullerenyl] proline, methyl ether of N-[mono-hydro-fullerenyl] proline, carnosine) [18,19]. The authors determined that all studied compounds had antioxidant activity and inhibited glutamate induced Ca<sup>2+</sup> uptake into synaptosomes of the rat brain cortex.

Kotelnikova et al. studied the antioxidant properties of water-soluble amino acid derivatives of fullerene  $C_{60}$  with sodium salts of aminobutyric acid, aminocaproic acid as well as a hybrid structure based on fullerenylproline and carnosine [20]. Moreover a reliable correlation between the development of cytomegalovirus (CMV) infection and the process of lipid peroxidation in the cell culture was established. An efficient inhibitor of the CMV infection from the class of amino acid derivatives of fullerene with antioxidant activity was obtained.

Additional group of articles are devoted to theoretical study of the amino acid derivatives of fullerenes in particular to calculation of: (i) ability of the individual  $C_{60}$  fullerene to interact with amino acids (the calculations were performed using DFT-B3LYP/3-21G theory). In particular authors of [21] revealed that the most favorable interactions of the fullerene is with arginine, leucine, and tryptophan which is related to the backbone structure of the corresponding amino acids. Hu [22] performed the same study referred to interaction of carboxyl-functionalized  $C_{60}$  cage ( $C_{60}C_3H_5$ COOH) with glycine; (ii) molecular structures of hybrid amino acid derivatives of  $C_{60}$  on the basis of quantum chemical methods [23]; (iii) dissociation constants ( $pK_a$ ) of amino acid derivatives of fullerenes. Luzhkov et al. calculated the  $pK_a$  values of the  $\alpha$ -COOH,  $\alpha$ -NH, and CH groups of the fullerene derivative L-Ala- $C_{60}$ H in water [24].

#### 2. Experimental

#### 2.1. Materials

Amino acid derivative of the  $C_{60}$  fullerene with L-arginine ( $C_{60}$ -Arg) —  $C_{60}(C_6H_{13}N_4O_2)_8H_8$  of mass fraction purity 99.5% was used for the physico-chemical investigation of water solutions. The reagent was produced in Ltd ZAO "ILIP" (St. Petersburg). Additionally we have carried out the identification of the purchased  $C_{60}$ -Arg using the complex of the physico-chemical methods such as IR and UV-spectroscopy, mass-spectrometry, elemental analysis.

## 2.2. Apparatus and procedures

The measurements of the concentration dependence of density of the  $C_{60}$ -Arg aqueous solutions were performed by the picnometer method. We used quartz picnometer, volume calibration was performed with distilled water, the accuracy of temperature control during the density measurement was  $\Delta T = \pm~0.1 \div~0.2$  K degrees, the accuracy of densities determination is equal to  $\Delta \rho = \pm~0.001$  g·cm $^{-3}$ . The series of the  $C_{60}$ -Arg water solutions were prepared by dilution of the basic solution at 298.15  $\pm~0.05$  K.

The concentration dependence of refractive index of the  $C_{60}$ -Arg derivative water solutions ( $n_D^{20}$ ) was measured by refractometry using Abbe refractometer IRF-454B2M (measurement limits in transmission light  $n_D^{25}=1.3-1.7$ ,  $\Delta n_D^{20}=\pm~0.0001$ , the accuracy of temperature control  $\Delta T=\pm~0.2$ K).

The temperature dependence of the C<sub>60</sub>-Arg derivative solubility in water in the temperature range 293.15–353.15 K was carried by the method of isothermal saturation in ampoules. The saturation time was equal to 8 h, temperature was maintained with accuracy equal to  $\pm 0.05$  K. For the C<sub>60</sub>-Arg water solutions saturation the thermostatic shaker (LAUDA ET 20) was used at a shaking frequency  $\omega \approx 80~\text{c}^{-1}$ , quantitative determination of the C<sub>60</sub>-Arg concentrations in water was performed using spectrophotometric technique at 330 nm (after the dilution and cooling of saturated solutions). The relative uncertainty of the solubility determination is equal to  $\pm 5\%$ . Relative air humidity was 40–50%.

For the thermogravimetric investigation of the  $C_{60}$ -Arg derivative we have used NETZSCH STA 449F3STA449F3A-0483-M apparatus, the temperature range was 293.15–903.15 K at the air, the heating rate was 5 K·min $^{-1}$ .

For determination of the specific conductivity of the C<sub>60</sub>-Arg water solutions the Cyber Scan PC-300 measuring device was used. The relative uncertainty of the specific conductivity determination is equal  $\pm\,1\%$ . The solutions used were saturated by atmospheric air

The measurements of the  $C_{60}$ -Arg nanoparticles size distribution in aqueous solutions of different concentrations carried out by the dynamic light scattering with the help of MalvernZetasizer 3000 (Great Britain) device.

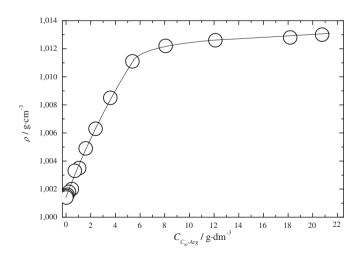
#### 3. Results and discussions

#### 3.1. Isothermal solution densities

Experimental data on isothermal water solution densities of the  $C_{60}$ -Arg derivative are presented in Fig. 2. Concentration dependence of average molar volume (V) of the solution components were calculated, according to classical thermodynamic Eq. (1):

$$\overline{V} = \frac{V}{n_{H_2O} + n_{C_{60}-Arg}},$$
 (1)

where: V is the volume of the  $C_{60}$ -Arg water solution;  $n_{H_2O}$  and  $n_{C_{60}}$ -Arg are the molar quantities of water and  $C_{60}$ -Arg derivative in 1 dm<sup>3</sup> of solution.



**Fig. 2.** Concentration dependence of the  $C_{60}$ -Arg water solution density ( $\rho$ ) at 298.15 K.  $C_{C_{50}-{\rm Arg}}$  is a volume concentration of the  $C_{60}$ -Arg derivative.

The partial molar volumes of the solution components ( $V_{H_2O}$  and  $V_{C_{60}-Arg}$ ) were calculated according to Eqs. (2), (3) [25,26]:

$$V_{H_2O} = \left(\frac{\partial V}{\partial n_{H_2O}}\right)_{T,P,n_{C_{60}-Arg}} V_{C_{60}-Arg} = \left(\frac{\partial V}{\partial n_{C_{60}-Arg}}\right)_{T,P,n_{H_2O}}, \tag{2}$$

$$V_{H_2O} = \overline{V} - x_{c_{60}-Arg} \left( \frac{\partial \overline{V}}{\partial x_{C_{60}-Arg}} \right)_{T,P} V_{c_{60}-Arg} = \overline{V} - x_{H_2O} \left( \frac{\partial \overline{V}}{\partial x_{H_2O}} \right)_{T,P}.$$
(3)

Concentration dependence of average molar volume  $(\overline{V})$  and partial molar volumes of the water solution components ( $V_{H_2O}$  and  $V_{C_{co}-Arg}$ ) are represented in Figs. 3. 4. One can see that in the area of diluted solutions the concentration dependence of the C<sub>60</sub>-Arg partial molar volume is rather complex. In the concentration range  $x \ge 0.0001$  the mole fraction dependence of the C<sub>60</sub>-Arg partial molar volume approaches to the molar volume of the solid  $C_{60}$ -Arg and in the mole fraction range  $10^{-4}$ – $10^{-6}$  the  $V_{C_{60}\text{-Arg}}(x_{C_{60}\text{-Arg}})$  dependence extremely decreases and even changes the sign (becomes negative). The latest fact reveals that addition of even small portions of the C<sub>60</sub>-Arg derivative extremely compact and structures the solution. Similar behavior of concentration dependences of partial molar volumes was previously observed in our scientific group for such water-soluble fullerene derivative as polyhydroxy- and carboxy-fullerenes [8,11,12]. It is noteworthy that crossing of the average molar volume through the minimum (Fig. 3) does not contradict to thermodynamic laws.

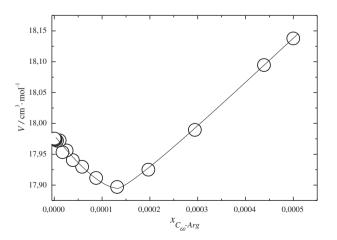
#### 3.2. Refraction of water solutions

Concentration dependence of the C<sub>60</sub>-Arg water solutions refraction index  $(n_D^{25})$  is presented in Fig. 5. Concentration dependence of specific and molar refractions of the C<sub>60</sub>-Arg aqueous solutions at 298 K was calculated using Eqs. (4), (5) [27,28].

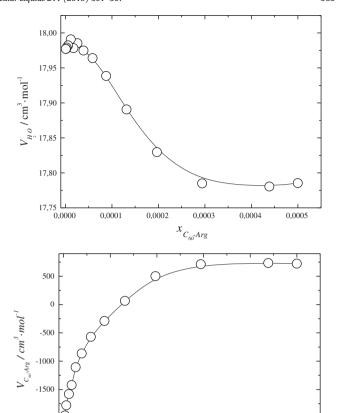
$$r = \left(\frac{n_D^{25^2} - 1}{n_D^{25^2} + 2}\right) \frac{1}{\rho} \tag{4}$$

$$R = \left(\frac{n_D^{25^2} - 1}{n_D^{25^2} + 2}\right) \frac{\overline{M}}{\rho},\tag{5}$$

where: r, R — are specific (cm $^3 \cdot g^{-1}$ ) and molar (cm $^3 \cdot mol^{-1}$ ) refractions,  $\overline{M}$  — average molecular weight of solution:  $\overline{M} = x_{H_2O} \cdot M_{H_2O} + x_{C_{00} - Arg} \cdot M_{C_{00} - Arg} (g \cdot mol^{-1})$ . Concentration dependencies of molar and



**Fig. 3.** Mole fraction  $(x_{C_{60}\text{-Arg}})$  dependence of the average molar volume (V) of the  $C_{60}$ -Arg water solution at 298.15 K.



**Fig. 4.** Mole fraction dependence of the partial volumes ( $V_{H_2O}$  and  $V_{C_{60}-Arg}$ ) of the solutions components (4.1, 4.2) at 298.15 K.

0.0002

0.0003

X C Arg

0.0004

0.0005

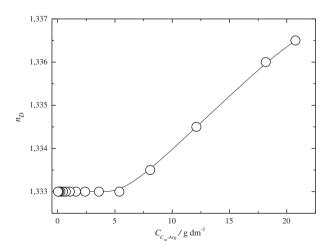
0.0001

0.0000

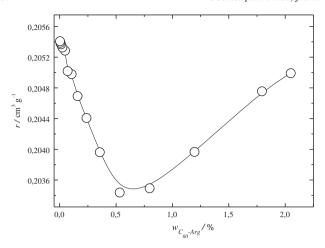
specific refractions are presented in Figs. 6, 7. According to the rule of additivity of the solution refractions we can obtain Eqs. (6.1), (6.2) [27,28]:

$$r = (r_{H_2O} \cdot w_{H_2O} + r_{C_{60}\text{-Arg}} \cdot w_{C_{60}\text{-Arg}}) \cdot \frac{1}{100}, \tag{6.1}$$

$$R = R_{H_2O} \cdot x_{H_2O} + R_{C_{60}\text{-Arg}} \cdot x_{C_{60}\text{-Arg}}$$
(6.2)



**Fig. 5.** Concentration dependence of the  $C_{60}$ -Arg water solution refraction index  $(n_D)$  at 208 K



**Fig. 6.** Mass fraction  $(w_{C_{60}}$ -Arg ) dependence of the  $C_{60}$ -Arg water solution specific refraction (r) at 298.15 K.

where:  $r_i$ ,  $R_i$  — specific and molar refraction of the i-th component,  $w_i$ ,  $x_i$  — mass and molar fractions of the i-th component. Thus, using the value of water refraction we can easily calculate molar and specific refractions of the C<sub>60</sub>-Arg derivative ( $r_{\text{C}_{60}\text{-Arg}}$ ,  $R_{\text{C}_{60\text{-Arg}}}$ ) (see Figs. 8, 9). One can see that both values of molar ( $R_{\text{C}_{60}\text{-Arg}}$ ) and specific ( $r_{\text{C}_{60}\text{-Arg}}$ ) refractions are constant. Taking into account the low accuracy of the refractions data at low concentration ( $x_{\text{C}_{60}\text{-Arg}} < 2 \cdot 10^{-4}$ ) we have not used them for the calculations.

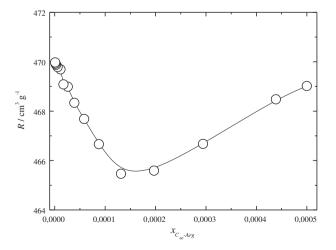
We can also calculate the  $C_{60}$ -Arg molar refraction according to another additivity rule [28] using the atomic refractions —  $R_{i(j)}$  of i-th atom in j-th functional group:

$$\begin{split} R^{add} \approx & 108R_c + 8R_{O(-OH)} + 8R_{O(=CO)} + 8R_{N(-NH_2)} + 24R_{N(-NH-)} \\ & + 112R_H \approx & 470 \pm 25 \text{ cm}^3 \cdot \text{mol}^{-1}. \end{split} \tag{7}$$

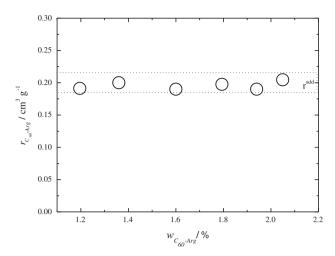
The discrepancy in the molar refraction calculation is connected with the choice of the spectral lines ( $H_{\alpha}[\lambda=658.3(\text{nm})]$  and  $H_{\gamma}[\lambda=436.1(\text{nm})]$ ). Alternative calculation of the C<sub>60</sub>-Arg molar refraction can be performed using the refractions of functional groups  $-R_{i}$ , (according to Fogel [28]):

$$R^{\text{add}} \approx 60R_{\text{c}} + 8R_{\text{-COOH}} + 8R_{-NH_2} + 24R_{-NH-} + 8R_{-CH-}$$

$$+ 24R_{-CH_3} \approx 462 \pm 25 \text{ cm}^3 \cdot \text{mol}^{-1}.$$
(8)



**Fig. 7.** Mole fraction  $(x_{C_{60}\text{-Arg}})$  dependence of the  $C_{60}$ -Arg water solution molar refraction (R) at 298.15 K.



**Fig. 8.** Specific refraction of  $C_{60}$ -Arg  $(r_{C_{60}}$ -Arg) in water solutions at 298.15 K.

The specific refraction of the  $C_{60}$ -Arg can be calculated using Eq. (9):

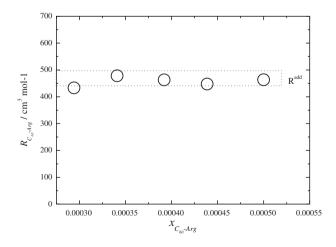
$$r^{add} = \frac{R^{add}}{M(C_{60} - Arg)} \approx 0.200 \div 0.215 \text{ cm}^3 \cdot \text{g}^{-1}.$$
 (9)

The value of the  $C_{60}$ -Arg specific refraction is very close to the specific refraction of water

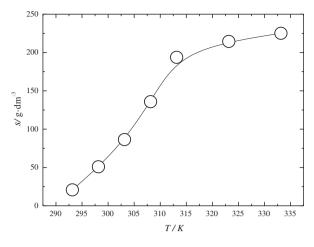
$$(r^{add}(HOH) \approx 0.207 \pm 0.003 \text{ cm}^3 \cdot \text{g}^{-1}).$$

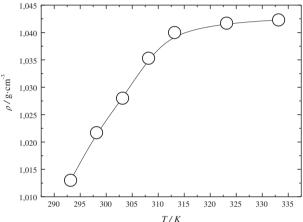
3.3. Temperature dependence of the  $C_{60}$ -Arg solubility in water and water solution densities

Temperature dependences of the  $C_{60}$ -Arg solubility and density in water are presented in Fig. 10. One can see the following: (i) the solubility values of  $C_{60}$ -Arg are very high (tens and hundreds  $g \cdot dm^{-3}$ ), such values correspond to the solubility of well-soluble phases as fullerenol-d or, for example halite — NaCl [29–31]; (ii) the solubility diagram consists of only one branch, corresponding to crystallization of non-solvated  $C_{60}$ -Arg; (iii) mono-variant branch of the  $C_{60}$ -Arg crystallization has a sigmoidal form. Such dependences are typical for the fullerene-solvent binary systems. For example the temperature dependences of solubility in binary  $C_{70}$ -o-xylene system or the branches of



**Fig. 9.** Molar refraction ( $R_{C_{60}\text{-Arg}}$ ) of the  $C_{60}\text{-Arg}$  derivative in water solutions at 298.15 K.





**Fig. 10.** Temperature dependence of solubility (S) (1) and density ( $\rho$ ) (2) of the C<sub>60</sub>-Arg water solutions.

crystallization of the sesqui-solvated fullerenes in the  $C_{60}$  ( $C_{70}$ )- $\alpha$ -chloronaphthalene ( $\alpha$ -bromonaphthalene), systems are characterized by sigmoid shape of the polytherms of solubility [32,33].

## 3.4. Conductivity of water solutions of C<sub>60</sub>-Arg

Concentration dependence of specific electric conductivity of the  $C_{60}$ -Arg water solutions at 298.15 K was investigated carrying out the measurement of the specific resistance of the solutions  $\rho$  ( $\Omega \cdot cm$ ) [34]:

$$\kappa = \frac{1}{\rho}.\tag{10}$$

Specific electric conductivity corresponds to conductivity of the unit volume of solution located between two parallel planar electrodes (at the distance of 1 cm) with the surface equal to 1 cm. One can see (Table 1) that the dependence  $\kappa$  ( $C_M$ ) ( $C_M$  is molarity of the  $C_{60}$ -Arg water solution) increases with increasing of concentration.

Molar electric conductivity  $(\lambda - S \cdot cm^2 \cdot mol^{-1})$ , i.e. conductivity of the electrolyte volume containing 1 mol of the solute was calculated using Eq. (11) [34]:

$$\lambda = \frac{1000\kappa}{C_{\text{M}}},\tag{11}$$

where:  $C_M$  is molarity of solution (mol·dm<sup>-3</sup>). Experimental data on molar conductivity are presented in Table 1. For the molar electric conductivity determination in the infinitely diluted solutions ( $\lambda_0$ ) we have extrapolated the  $\lambda$  ( $C_M^{1/2}$ ) dependence to  $C_M^{1/2}=0$ , according to Onsager equation [34]:

$$\lambda = \lambda_0 - A \cdot C_M^{\frac{1}{2}},\tag{12}$$

where: A is a constant under conditions of experiment.

Apparent degree of dissociation  $\alpha$  was calculated, according to Eq. (13) [34]:

$$\alpha = \frac{\lambda}{\lambda_0},\tag{13}$$

The calculated values of the apparent degree of dissociation are presented in Table 1 ( $\lambda_0 \approx 3 \cdot 10^4 \, \text{S} \cdot \text{cm}^2 \cdot \text{mol}^{-1}$ ). We propose the protonic mechanism of the C<sub>60</sub>-Arg derivative dissociation according to the following scheme:

$$C_{60}(C_5H_{12}N_4COOH)_8H_8 \leftrightarrow C_{60}(C_5H_{12}N_4COOH)_7H_8C_5H_{12}N_4COO^- + H^+. \eqno(14)$$

The concentration dependence of concentration dissociation constant —  $K_D$  (see Table 1) was calculated according to the "Ostwald dilution law" (neglecting by activity coefficients of the ions and non-dissociated molecular —  $\gamma_i = \gamma_\pm = 1$ ) [34]:

$$K_d = \frac{C_M \alpha^2}{(1 - \alpha)}. (15)$$

Thermodynamic dissociation constant  $-K_D^{therm}$  was calculated by extrapolation of  $K_D$  ( $C_M$ ) dependence to the region of infinitely diluted solution:

$$K_D^{therm} = \lim_{C_M \to 0} (K_D), pK_D = -\lg K_D.$$
 (16)

According to our calculation  $pK_D^{therm} = 7.2$ .

**Table 1** Experimental data on electric conductivity of the  $C_{60}$ -Arg water solutions.

C/g⋅dm <sup>-3</sup>	$M/\text{mol}\cdot\text{dm}^{-3}$	α	κ/S⋅cm <sup>-1</sup>	$\lambda/S \cdot cm^2 \cdot mol^{-1}$	$pK_D$
0	0	1.00 (extrapolation)	0.00	2.98 · 10 <sup>4</sup> (extrapolation)	7.2 (extrapolation)
0.003125	0.00000095	0.31	$8.87 \cdot 10^{-6}$	9340	7.2
0.00625	0.0000019	0.16	$9.04 \cdot 10^{-6}$	4760	7.2
0.0125	0.0000038	0.10	$1.15 \cdot 10^{-5}$	3030	7.4
0.025	0.0000075	0.075	$1.68 \cdot 10^{-5}$	2240	7.3
0.05	0.000015	0.061	$2.73 \cdot 10^{-5}$	1820	7.2
0.0625	0.00003	0.033	$2.94 \cdot 10^{-5}$	980	7.5
0.125	0.00006	0.029	$5.13 \cdot 10^{-5}$	855	7.3
0.25	0.00012	0.026	$9.12 \cdot 10^{-5}$	760	7.1
0.5	0.000235	0.024	$1.70 \cdot 10^{-4}$	723	6.8
1	0.00047	0.023	$3.26 \cdot 10^{-4}$	693	6.6

### 3.5. Size distribution of the $C_{60}$ -Arg associates in water solutions

The C<sub>60</sub>-Arg water solutions were analyzed by dynamic light scattering (typical examples are presented in supporting information). We can point out the following regularities: (i) we have not detected monomer molecules (with linear dimension equal to  $\delta_0 \approx 2$  nm) in all investigated solutions even in very diluted; (ii) the presence of the «first type associates» with linear dimension  $\delta_1 \approx 40$ –50 nm was observed in comparatively dilute solutions ( $C_{C_{60}-Arg} > 0.0125 \, g \cdot dm^{-3}$ ), in more concentrated solutions ( $C_{C_{60}-Arg} > 0.05 \text{ g} \cdot \text{dm}^{-3}$ ) we detected the presence of the «second type associates» with linear dimension  $\delta_2 \approx 200$ –400 nm and «third type associates» ( $\delta_3 \approx 1000 \pm 100$  nm) in the concentration range ( $C_{C_{60}-Arg} > 0.5~g\cdot dm^{-3}$ ). The formation of the associates with such gigantic dimensions (in the latest case) proves the microheterogeneous nature of the C<sub>60</sub>-Arg water solutions; (iii) investigation of the size distribution of the C<sub>60</sub>-Arg associates in more concentrated solutions (10–30 g $\cdot$ dm<sup>-3</sup>) reveals the global association processes with formation of viscous and gelatinous solutions including "huge associates" with linear dimension  $\delta_4 \approx n1 \div n10 \, \mu m$  (units or tens of μm). In this case the destroying of stable colloid solution takes place; (iv) for the description of such experimental facts in the association process the model of consequent hierarchical association model was used [11,12]. We consider that monomer spherical molecular form first type spherical associates, than first type spherical associates form second type spherical associates (first and second type associates form nano-colloid solutions), than second type spherical associates form third type spherical associates (the third type associates corresponds to the colloid heterogeneous system), than third type spherical associates form forth type spherical associates (the forth type associates corresponds to destroying colloid system). The obtained experimental data are summarized in Table 2.

## 3.6. Complex thermal analysis of the C<sub>60</sub>-Arg derivative

The result of complex thermal analysis of the  $C_{60}$ -Arg derivative is presented in Fig. 11. Analogous behavior with multistage dehydration and decarboxylation was previously observed for carboxylated fuller-ene- $C_{60}[(C(COOH)_2]_3 \ [11]]$ . Analysis of Fig. 11 reveals the following: (i) the absence of crystallohydrates in the solid phase; (ii) in the temperature range 373–813 K the multistage processes of destroying of the functional "arginine groups" in the presence of  $O_2$  (from air) is realized. The processes of dehydration ( $-nH_2O$ ), soft decarboxylation (-nCO), diazotizing ( $-N_2$ ) and dehydrogenation ( $-H_2$ ) are accompanied by oxidation. According to TG-curve (see Fig. 11) the arginine groups were decomposed in the temperature range 373–813 K. The complete decomposition of the arginine groups is equivalent to the mass loss equal to 69%; (iii) in the temperature range 813–923 K the oxidation of the fullerene core ( $C_{60}$ ) takes place.

Table 2

Average size of the  $C_{60}$ -Arg associates in water solutions.  $C_{C_{00}}$ -Arg-volume concentration of the  $C_{60}$ -Arg derivatives;  $\delta$  is a average diameter of the  $C_{60}$ -Arg associate;  $N_i$  is a average number of the  $C_{60}$ -Arg molecules in the associate (the calculations were performed according to equation:  $N = \left[\frac{\delta}{\delta_0}\right]^3 \left[K_{packing}^{spheroid}\right]^W$ , where  $K_{packing}^{spheroid}$  characterize the ratio between the volume occupied by the  $C_{60}$ -Arg monomer molecules and the total volume of the associate (in spheroid approximation),  $K_{packing}^{spheroid} = \frac{1}{2} \left[8,11,12\right]$ ); W is a order of the  $C_{60}$ -Arg associate;  $C_{I}$ -is a number of the  $W_I$  order associates per liter of solution ( $C_I = \frac{C_{C_{00}-Arg}N_I}{M_{C_{00}-Arg}N_I}$ , where  $N_A$ -Avogadro constant);  $V_I$  is a volume of associates referred to 1 dm³ of solution ( $V_I = \frac{1}{6}$ ,  $C_I \pi \delta^3$ ,  $\pi = 3.14$ ).

Nº	$C_{C_{60}-Arg}/g\cdot l^{-1}$	δ/nm	$N_i$	$C_i$ /particles·l <sup>-1</sup>	W	V <sub>i</sub> /nm <sup>3</sup>
1	<10 <sup>-3</sup>	$2 \pm 0.2$	$\approx 1 \cdot 10^0$	<3 · 10 <sup>17</sup>	0	<1 · 10 <sup>18</sup>
2	0.01	$45 \pm 5$	$\approx 6 \cdot 10^3$	$\approx$ 5 · 10 <sup>14</sup>	1	$\approx 2 \cdot 10^{19}$
3	0.1			$\approx 1 \cdot 10^{13}$	2	$\approx 3 \cdot 10^{20}$
4	3	$1000\pm200$	$\approx 3 \cdot 10^8$	$\approx 2 \cdot 10^{12}$	3	$\approx 1 \cdot 10^{21}$

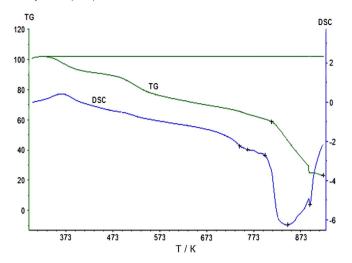


Fig. 11. Thermal analysis of the C<sub>60</sub>-Arg derivative.

#### 4. Conclusions

Novel physico-chemical properties of the  $C_{60}$ -Arg water solutions were investigated. The concentration dependence of density was investigated by the pycnometer method; the average molar and partial volumes of the solution components were calculated at 298.15 K. By the isothermal saturation method the  $C_{60}$ -Arg solubility in the distilled water in the temperature range 293.15 to 333.15 K was studied. The specific and molar conductivities, dissociation constant and apparent degree of dissociation were obtained for the  $C_{60}$ -Arg water solutions. By the dynamic light scattering method, the average size of the  $C_{60}$ -Arg associates was determined. The actuality of such investigation is closely connected with possibilities of application of the obtained results for the development and optimization of the water-soluble derivatives of fullerenes with amino acids application in biology and medicine.

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