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The effect of sodium chloride on thermodynamic parameters of ion association for cysteine and methionine with L-ascorbic acid in aqueous solution at different temperature

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Abstract. Molar conductivity for hydrogen bonds ion association of ascorbic acid (AA) with Cysteine (Cys) and Methionine (Met) at concentration ($2-5 \times 10^{-3}$, 2×10^{-3} and 2×10^{-3} M respectively) in water and in (2×10^{-3} M) sodium chloride (NaCl) solutions was examined at temperatures range (298 K to 313 K). The conductance equation of Shedlovsky was used to calculate the association constants (K_A) and the limiting molar conductance (Λ_o). The association heat, the Gibbs free energy, the change of entropy and the activation energy (ΔH° , ΔG° , ΔS° , and ΔE_S respectively) also calculated. All results are computed. The data showed increasing of Λ_o , with increases in temperature and the same trend in decreasing for K_A . The data showed negative values for ΔH° and ΔG° parameters. This refers to the exothermic association processes, and values increased with increasing in AA concentration, while ΔS° and ΔE_S parameters show a positive value related to the higher mobility of the ions and decreasing of ion association solvation in water and NaCl solutions. The ΔE_S and ΔH° of a different solution in present sodium chloride show decreasing in values as compared with water solution at different concentration of AA, while the opposite trend values appear for ΔG° and ΔS° with some exception.

1. Introduction

The human bodies do its essential functions by consuming food and converting it into usable energy. The efficiency of translation the energy of food to work energy through a physical activity is in the range of (2.6-6.5%), and this depends on a physical activity type. In spite of the high availability of energy (thermal and mechanical energy) through the physical activity and the low efficiency of this translation process, it is prepared a good form of the renewable energy [1].

Amino acids (AAs) are important and essential to all life. It exists in fruits and other foodstuffs. AAs are best known as the nutrient substrates for protein synthesis (protein play important roles in human systems); in addition, they can also participate as bioactive molecules in nutrition metabolism [2]. Sodium chloride is naturally present in all foods and may be added during food processing. It is found that sodium chloride increases the solubility of AAs, so "the solubility of AAs is highly important in determining biological action of bio-macromolecules such as proteins"[3]. The combustion of free AAs inside human body gives (1.3-6.5 Kcal/g) as heat of combustion (enthalpy change for AAs oxidation) [4].

In another side "ascorbic acid (Vitamin C, AA) is one of the most important vitamins with antioxidant properties that widely exist in various fruits and vegetables and widely used in pharmaceutical formulations and cosmetic applications. AA plays a crucial role in healthy and biological metabolism in



the body. It takes part in free radical scavenging, cancer prevention, collagen synthesis, iron absorption, wound healing, cholesterol metabolism, and immunity improvement. Deficiency of ascorbic acid in human may cause scurvy, anemia, gum problems, muscle degradation, and neurotic disorders"[5]. Beside that AA is effected on AAs absorption by increasing it from the oral given dose [6], (so "the energy content of a foodstuff is determined by multiple factors including the efficiency of intestinal absorption"[4]). Both AAs and AA (Figure 1) are water-soluble, and they are co-occurrence in some metabolic reactions, medication, food and cosmetic. Therefore, information on the interactions between AAs and AA is very important for their mention fields.

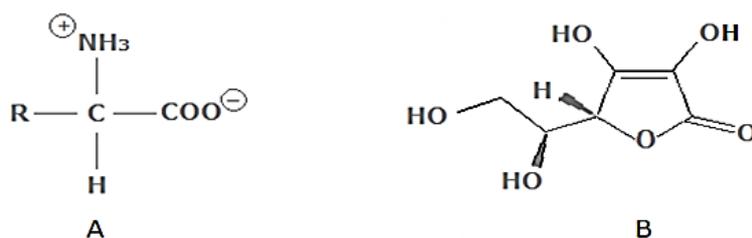


Figure 1. A: L- α -amino acid, B: L-Ascorbic acid.

The present work reports a study the effect of inorganic salt (NaCl) on thermodynamic parameters, the heat of association, the Gibbs free energy, the change of entropy and the activation energy (ΔH° , ΔG° , ΔS° , and ΔE_s consecutively) which calculated by the result of the molar conductance (Λ), limiting molar conductance (Λ_o), and ion association constant (K_A) of methionine (*Met*), cysteine amino acid (*Cys*) with AA in water and NaCl solutions at temperatures of ranges (298-313 K) as comparative study.

2. Materials and Methods

Methionine amino acid (*Met*), cysteine amino acid (*Cys*), ascorbic acid (AA), and sodium chloride (NaCl) were purchased from "Merck" (with purity at 99.9 %).

Water solvent (deionized) is used in different solutions with a specific conductance in the range of (40×10^{-6} S/cm). Solvent viscosities, η , relative permittivity, ϵ_r , densities, ρ_o , are characterized in Table 1. A digital conductivity bridges with a dip type immersion conductivity cell were used to measure a specific conductivities.

All the solutions of AA, *Cys* and *Met* are prepared at different concentrations ($2-5 \times 10^{-3}$ M) by dissolving the suitable quantity of the samples in water and NaCl solutions. Specific conductivity measurements were done at range (298-313 K) of the temperatures, under control by using a (GFL/D30938) water bath and thermometer device. The weight of samples was measured by using (Sartorius RC-210D) an analytical balance.

3. Results and discussion

3.1. Conductance of AA: AAs in water and NaCl solutions at different temperatures.

The hydrogen bond is "an attractive interaction between a hydrogen atom from a molecule or fragment ($X-H$) in which X is more electronegative than H , and an atom or a group of atoms A in the same or a different molecule, where there is evidence of bond formation. The acceptor may be an atom or an anion X , or a fragment or a molecule ($X-H$), where X is bonded to H . In any event, the acceptor is an electron-rich region such as a lone pair or a π cloud"[7]. The hydrogen bonding between molecules, as a result, is affected on molecules physical properties.

Amino acids are molecules containing at least one carboxyl group ($-COOH$) and at least one primary or secondary amino group ($-NH_2$). "The general formula of amino acids is then (H_2NCHR_1COOH). Due to the acidic-basic behavior, amino acids are considered as ampholytes, which means that they can with

the negatively charged ($-COO^-$) group or a positively charged ($-NH_3^+$) behave outwardly as acids or as a basis"[8]. Hydrophobic amino acid side chains can interfere in hydrogen bonding to water, or amino acid polar chains can participate with it. Neutral polar side chains contain ($-OH$, $-SH$, $-NH$, $-CO$) uncharged groups that can be bonded to water by hydrogen bond. Like cysteine and methionine amino acids that represented in Figure 2.



Figure 2. A: L- α -Cysteine amino acid (*Cys*), B: L- α -Methionine amino acid (*Met*).

Researchers proved that side chains of AAs in the heme groups interact with ketone and hydroxyl groups of AA by hydrogen bonds [9]. One of the hydrogen bond interactions between an amino acid and ascorbic acid in a solution can be represented in Figure 3. The interactions of AAs with AA have an important role in biochemical environments so it is evaluated that AA has an effect on increased absorption of some amino acids.

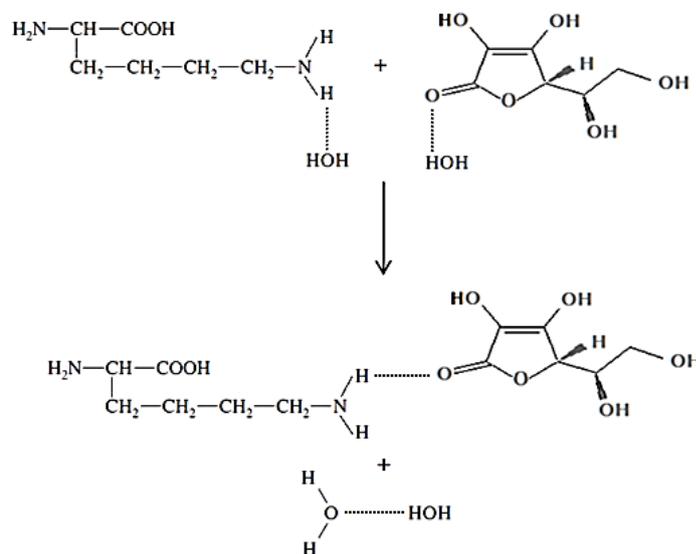


Figure 3. Some of hydrogen bond interactions between AAs and AA in a solution.

In another side, sodium chloride (*NaCl*) is a famous inorganic salt, was one of the first nutrients to be identified as necessary to life. "Sodium is absorbed in the small intestine, often in association with the absorption of other substances (amino acids and glucose)"[10]. Searchers results refer that sodium chloride increase the solubility of amino acids, and other physical properties [3]. Therefore, sodium chloride molecules are considered as contributors in the interaction and involvement in the ion association constant equation and the choice of (0.002 M) because it is in the concentration range of the biological environment [11].

The transport properties of "mixtures of ionic liquids (*conductance, viscosity, and transference numbers*) are important because the values provide useful and sensitive information about ion-solvent interaction, ionic association, and solvent structure" [12]. In most chemical interactions a suitable solvent is very important. Water is the famous solvent in inorganic chemistry [13], has a vital role in biological processes, and it's mainly to all life. Table 1 [12] detailed the physical properties of water, so the densities, viscosities, and relative permittivity are used in ion association constant expression.

Table-1: Densities, ρ , viscosities, η , and relative permittivity, ϵ_r , of conductivity water at different temperatures.

T/K	$\rho / g m^{-3}$	$\eta / g m^{-1} s^{-1}$	ϵ_r
298	0.9971	0.8903	78.30
303	0.9957	0.7974	76.55
308	0.9941	0.7194	74.83
313	0.9923	0.6531	73.15

The molar concentration (C) is used to convert the specific conductance (κ) for a solution to the corresponding molar conductance (Λ) by equation 1.

$$\Lambda = \frac{1000 K}{c} \quad (1)$$

Chemical association (ex. hydrogen bond) is an aggregation of molecules or atoms into bigger units connected together by a weaker force than a chemical bond that atoms bond to a molecule. Hydrogen bond increase in more number of molecules, as a result, to increase in concentration. So the conductivity data were analyzed at ($2.5 \times 10^{-3} M$), ($2 \times 10^{-3} M$) and ($2 \times 10^{-3} M$) for AA, AAs, and NaCl in water respectively. Association constant is evaluated by uses a set of equations. K_A is calculated from conductance values using Shedlovsky equation (Eq. 2) [12].

$$\frac{1}{\Lambda S(Z)} = \frac{1}{\Lambda_o} + \frac{K_A C \Lambda f_{\pm}^2 S(Z)}{\Lambda_o^2} \quad (2)$$

Where Λ , is the molar conductance, C , the molar concentration of a solution, Λ_o , the limiting molar conductance, K_A , the association constant, f_{\pm} , free ions activity coefficient and will be calculated by using the expression (Eq. 3):

$$-\log f_{\pm} = \frac{A\sqrt{I}}{1+B R\sqrt{I}} \quad (3)$$

$$A = \frac{1.8247 \times 10^6}{(DT)^{3/2}}; \quad B = \frac{0.5209 \times 10^{10}}{\sqrt{DT}}$$

Where: R , is the ion association distance, T , the temperature, D , the dielectric constant, and η , the viscosity.

$$S(Z) = \left(\frac{Z}{2} + \sqrt{1 + \left(\frac{Z}{2} \right)^2} \right)^2 \quad (4)$$

$$Z = \left[\frac{S}{\Lambda_o^{3/2}} \right] \sqrt{C\Lambda}; \quad S = \alpha\Lambda_o + \beta; \quad \beta = \frac{82.501}{\eta\sqrt{DT}}; \quad \alpha = \frac{0.8204 \times 10^6}{(DT)^{3/2}}$$

As shown in Figure 1, AAs usually at neutral pH have both α -ammonium (+ve) and carboxylate (-ve) groups, with a small value in conductivity, and as a reason we take the conductivity of AA solution to comparative it with the conductivity of AAs: AA ion association in water and in sodium chloride solutions.

The molar conductivity, Λ , of AA to AAs in water and NaCl solution at different temperature are detailed in Table 2.

Table 2. The molar conductivity, $\Lambda/S\text{cm}^2\text{mol}^{-1}$, the association constant, $K_A/\text{dm}^3\text{mol}^{-1}$ and the limiting molar conductivity, $\Lambda_0/S\text{cm}^2\text{mol}^{-1}$, of different solutions at different temperature relative to AA concentration.

AA/water $C \times 10^{-3} \text{ M}$	T/K							
	298		303		308		313	
	Λ	K_A	Λ	K_A	Λ	K_A	Λ	K_A
2	186	430	189	415	191	410	193	404
3	157	516	160	496	163	479	165	469
4	141	538	144	516	146	505	148	495
5	130	544	132	531	135	508	137	497
Λ_0	289		291		293		295	
<i>AA: Cys/water</i>								
2	178	513	180	498	182	490	185	474
3	148	627	150	607	153	584	155	573
4	129	701	131	678	133	663	135	647
5	115	767	117	740	119	720	122	686
Λ_0	290		291		293		295	
<i>AA: Met/water</i>								
2	164	477	168	445	171	444	173	438
3	140	544	142	532	145	523	147	512
4	122	616	124	601	126	599	129	572
5	109	675	111	656	112	666	114	648
Λ_0	262		264		268		270	
<i>AA/ NaCl</i>								
2	301	628	304	615	306	610	308	604
3	230	950	232	938	235	914	237	902
4	198	1068	201	1037	203	1020	206	927
5	174	1189	177	1150	179	1128	181	1108
Λ_0	520		522		524		526	
<i>AA: Cys/ NaCl</i>								
2	287	655	290	641	292	639	294	633
3	219	987	221	973	224	953	226	940
4	183	1195	185	1174	187	1160	190	1124
5	159	1362	161	1334	163	1313	165	1268
Λ_0	502		504		507		509	
<i>AA: Met/ NaCl</i>								
2	275	628	277	622	280	608	282	602
3	213	914	215	901	217	888	220	865
4	176	1146	178	1125	179	1121	181	1101
5	151	1351	154	1299	156	1272	159	1225
Λ_0	475		477		479		481	

The limiting molar conductivity, Λ_0 , can be obtained by linear extrapolation from the plot of the square root of the molar concentration, $C^{1/2}$ versus the molar conductivity, Λ , [15] by used equation-5 as shown in Figure 4.

$$\Lambda = -k\sqrt{C} + \Lambda_0 \quad (5)$$

Where, k is a constant.

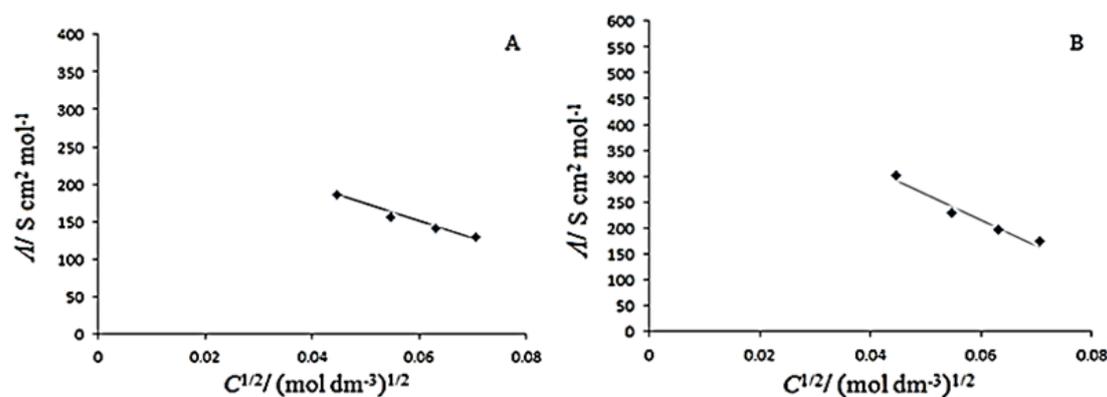


Figure 4. Dependence the limiting molar conductance (Λ) on the square root concentration ($C^{1/2}$) of AA, A- in water, B- in 0.002 M NaCl solution at 298 K.

The study of electrolytes transport properties in different solvents is particularly important to obtain information about the solvation and association activities of ions in solutions. The molar conductivity of electrolytes in solvents primarily depends on the viscosity of the solvent and the electrolyte concentration. Conductivity measurements are used usually in various environmental and industrial fields to measure the ionic content in a solution. Table 2 showed that the molar conductivity and the association constant of different solutions in presence of sodium chloride increasing in values as compared with water solutions at a different concentration of AA and at different temperatures. The conductivity of Cys and Met with AA in water and sodium chloride solutions appears a decrease in molar conductivity relative to the conductivity of the AA solution alone at a different concentration of AA and at different temperatures and this refers to the effect of hydrogen bond in the ion association. From Table 2, it can be seen that Λ_o , values increase for ion association as the temperature rise, and the same tendency is seen for K_A , values but in a decrease. "The increase of Λ_o , with an increase of temperature, is due to the decrease of solvent viscosity" [16], and the "higher mobility or a reduced amount of solvation of the ions in aqueous solution. This is owing to the fact that the growth thermal energy effects in greater bond breaking and also lead to higher mobility and higher frequency of ions because of a variant in translational, rotational, and vibrational energy of molecules" [17].

The values of K_A , for Cys and Met with AA in water and NaCl solutions appear increasing in values relative to the association constant of the AA solution alone at different concentration and at different temperatures.

3.2. Measurements of AA: AAs Thermodynamic functions in water and NaCl solutions:

A thermodynamic parameter is a measure of the valuable work in a system (chemical reaction); it is related to the system's temperature, enthalpy, and entropy. The reaction rate depends on collisions frequency, a factor of probability and activation energy.

The activation energy "(which is defined in a chemical system as the amount of energy available for a reaction to take place) can be calculated from the equation of S. A. Arrhenius" [17] (Eq. 6).

$$\log \Lambda_o = - \left(\frac{\Delta E_s}{2.303R} \right) \left(\frac{1}{T} \right) + \log A \quad (6)$$

Where: ΔE_s , is activation energy, A , is the factor of Arrhenius, T , is the temperature, and R , is the constant of an ideal gas. A graph plot between $(1/T)$ and $(\log A)$, to obtain a straight line as displayed in 'Figure 5', with a slope equal to $(-\Delta E_s/2.303R)$, so ΔE_s value can be calculated.

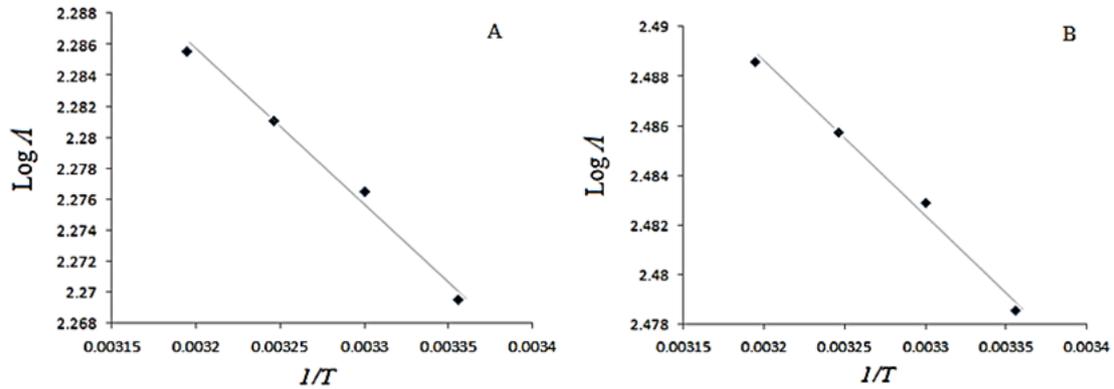


Figure 5. The plot of $\log A$ as a function of $1/T$ for $0.002 M AA$, A- in water, B- in $0.002 M NaCl$.

The standard enthalpy ΔH° , (or the formation heat association is the enthalpy change when a compound is formed from one mole of its elements) calculated by using Van't Hoff's equation (Eq. 7) [17].

$$\log K_A = -\left(\frac{\Delta H^\circ}{2.303R}\right)\left(\frac{1}{T}\right) + \left(\frac{\Delta S^\circ}{2.303R}\right) \quad (7)$$

So the ΔH° can be considered as a thermodynamic potential. It is an extensive quantity and a state function. It is referred to the heat released (or absorbed) and also to the difference in energies of the bond between the product and reactant in a chemical reaction.

The ΔH° value can be obtained from $(-\Delta H^\circ/2.303R)$, the slope that results from a plot of the graph between $1/T$ and a $\log K_A$, as shown in Figure 6.

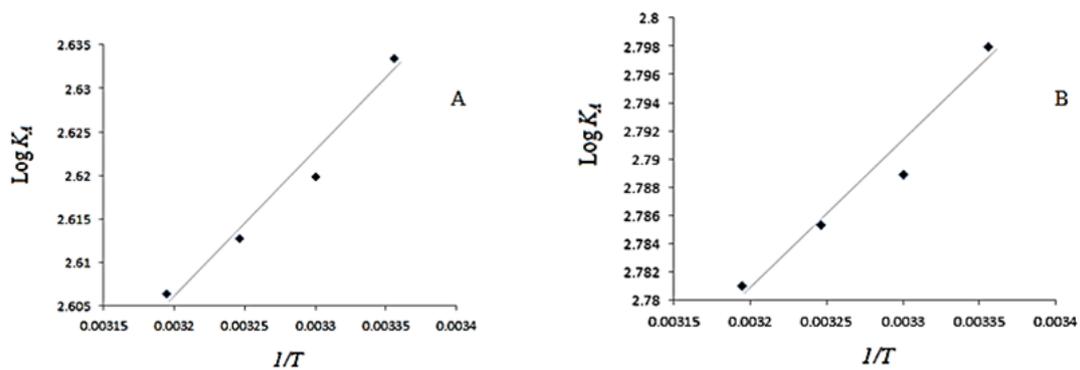


Figure 6. The plot of $\log K_A$ as a function of $1/T$ for $0.002 M AA$, A- in water, B- in $0.002 M NaCl$.

The measure of a usable energy throw association process can be obtained from the change in Gibbs free energy ΔG° (Eq. 8) [17].

$$\Delta G^\circ = -RT \ln K_A \quad (8)$$

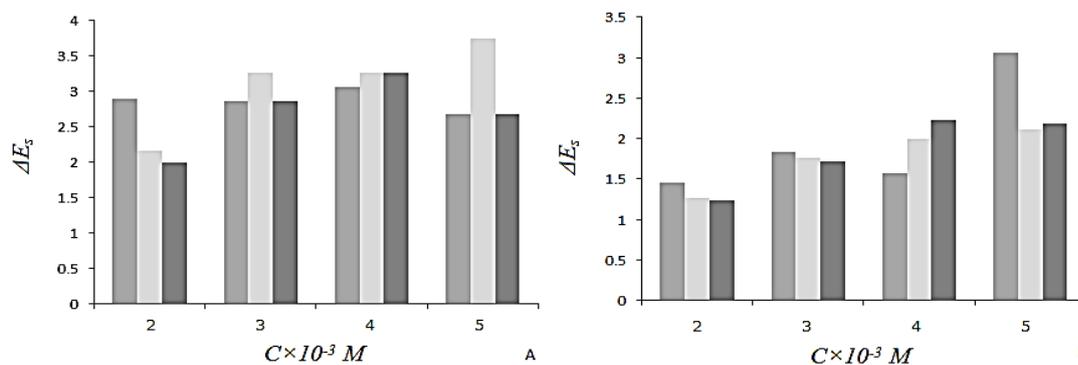
A measure of randomness or disorder can give by the state function, ΔS° (the change of entropy) from the Gibbs-Helmholtz equation (Eq. 9) [17].

$$\Delta G^{\circ} = \Delta H^{\circ} - T\Delta S^{\circ} \quad (9)$$

The data obtained from equations (6-9) can be detailed in Table 3.

Table 3. Thermodynamic parameters: ΔG° (KJ mol⁻¹), ΔS° (J K⁻¹mol⁻¹), ΔH° (KJ mol⁻¹), and ΔE_s (KJ mol⁻¹), of different solutions relative to AA concentration.

AA/water		Thermodynamic parameters		
$C \times 10^{-3} \text{ M}$	ΔE_s	ΔH°	ΔG°	ΔS°
2	1.99	-3.25	-15.02	39.50
3	2.87	-4.98	-15.48	35.23
4	3.26	-4.21	-15.58	38.15
5	2.68	-5.36	-15.61	34.40
<i>AA: Cys/water</i>				
2	2.17	-4.34	-15.50	37.45
3	3.26	-4.98	-15.96	36.85
4	3.26	-4.60	-16.23	39.03
5	3.75	-6.13	-16.46	34.66
<i>AA:Met/water</i>				
2	2.89	-4.70	-15.30	35.60
3	2.87	-3.06	-15.61	42.11
4	3.06	-4.21	-15.91	39.26
5	2.68	-2.30	-16.14	49.43
<i>AA/ NaCl</i>				
2	1.23	-2.17	-16.00	44.60
3	1.72	-2.68	-16.99	48.02
4	2.22	-7.66	-17.28	32.28
5	2.18	-3.83	-17.54	46.01
<i>AA: Met/ NaCl</i>				
2	1.26	-1.70	-16.10	48.32
3	1.76	-2.11	-17.08	50.23
4	1.99	-3.45	-17.56	47.35
5	2.11	-3.87	-17.88	47.01
<i>AA: Met/ NaCl</i>				
2	1.45	-2.30	-16.00	46.00
3	1.84	-3.06	-16.89	46.41
4	1.57	-2.30	-17.45	50.84
5	3.06	-4.98	-17.86	43.22



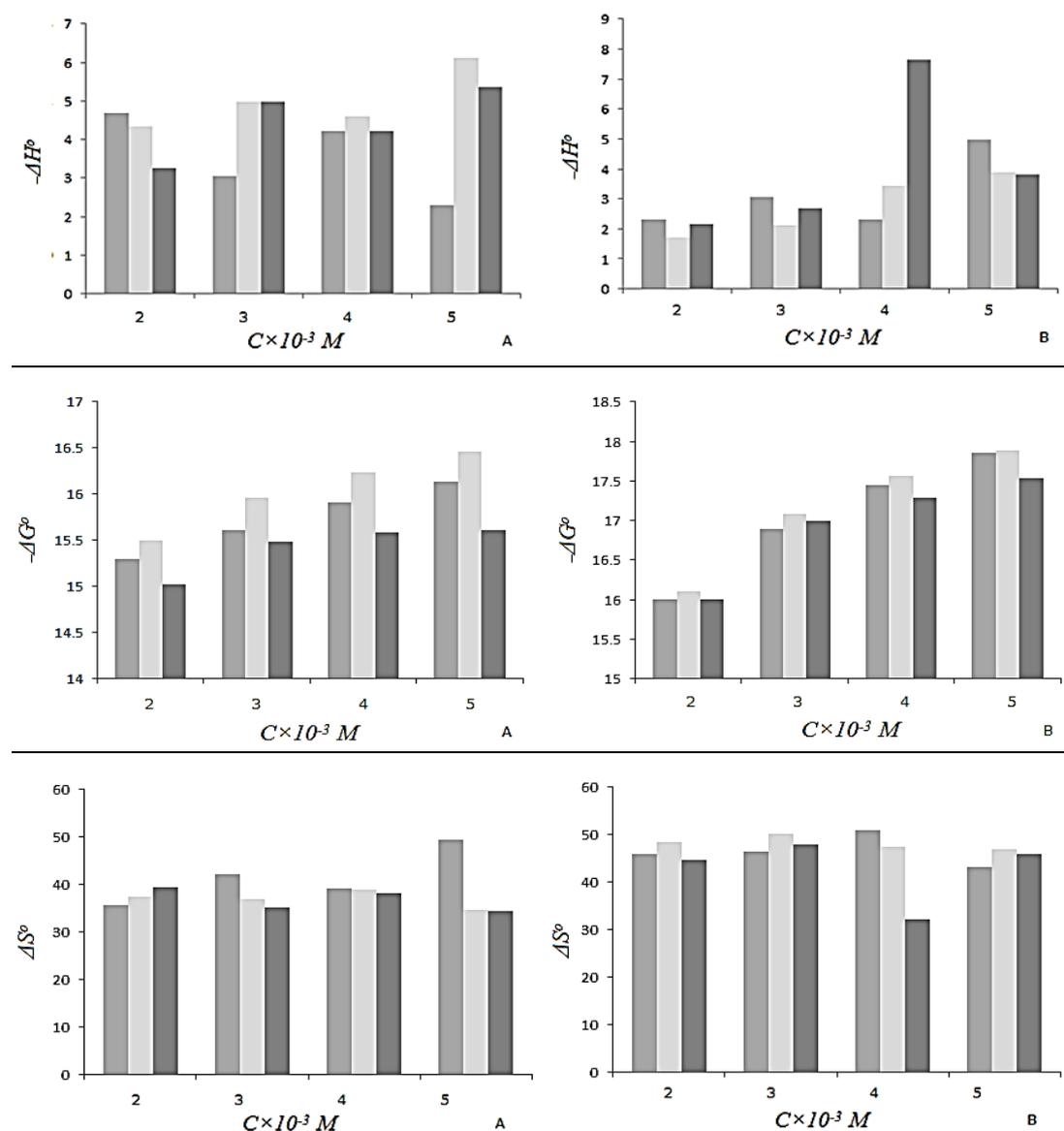


Figure 7. ΔE_s , ΔH° , ΔG° and ΔS° as a function of AA concentration: A- \blacksquare In water, \square in water + 0.002 M Cys, \blacksquare in water + 0.002 M Met B- \blacksquare In 0.002 M NaCl, \square in 0.002 M NaCl + 0.002 M Cys, \blacksquare in 0.002 M NaCl + 0.002 M Met.

The results data from Table 3 and Figure 7 show that:

The ΔE_s and ΔH° of different solutions with sodium chloride show a decrease in values as compared with water solution at different concentration of AA, while the opposite trend values appear for ΔG° and ΔS° with some exception. The activation energy ΔE_s , is a positive value for the ion association by hydrogen bond in water and in sodium chloride solutions and this representative an increase in mobility of the ions and, therefore, an increasing in Λ_o , values. The ΔG° , are appeared in negative values for the association process in solutions, this signifying that the associations between molecules are favorite rather than the dissociation process in solutions. The ΔH° , function shown negative results in water and NaCl solutions and this refers to exothermic association processes for the interaction between molecules ions. A positive value for ΔS° parameter indicates increasing in disorder of ions in different solutions, because of reduction in the solvation of ion association related to that of the free ion. This can be

accustomed to an increase in the degree of freedom through association processes, and this related to the solvent molecules freedom [12,16 and 17]. The results showed decrease ΔE_s and ΔH° while increase ΔG° and ΔS° in NaCl solutions as compared to water solutions for the materials under study, the reason accustomed that the materials in NaCl solution have bigger molar conductivity and association constants relative to the same concentration of materials in water solutions.

4. Conclusions

The search study the relation between AAs (Cys and Met) with AA through thermodynamic properties for ion association represented by hydrogen bonds in water and in NaCl solutions. The molar conductivity of AA with Cys and Met in different solutions have been recorded at $T = 298-313$ K. The association constant K_A , and the limiting molar conductivity Λ_o , are calculated using the Shedlovsky equation. The data showed that the values of Λ_o , increasing for hydrogen bonds ion association as the temperature increases, and the opposite trend is for K_A values. The rise of Λ_o , with a rise of temperature, accustomed to the decreasing in viscosity of the solvent [16] and reducing the ions solvation in different solutions. "This is owing to the fact that the growth thermal energy effects in greater bond breaking and also lead to higher mobility and higher frequency of ions because of a variant in translational, rotational, and vibrational energy of molecules" [12]. ΔH° , ΔG° , ΔS° , and ΔE_s , also calculated. ΔE_s , is a positive result for the ion association in different solutions and this representing that in different solutions, the ions own higher mobility. The ΔG° show negative values indicate that the ion association process favorite rather than the dissociation in different solutions. The ΔH° , shown negative values refer to the exothermic association in different solutions, and this can be caused by the interaction between molecule ions. A positive ΔS° , values refer a rising in disorders of ions in different solutions, because of reduction in the ion association solvation related to that of the free ion, and this because of the free solvent molecules [12, 16 and 17]. The results showed decrease ΔE_s and ΔH° while increase ΔG° and ΔS° in NaCl solutions as compared to water solutions for the materials under study, the reason accustomed that the materials in NaCl solution have bigger molar conductivity and association constants relative to the same concentration of materials in water solutions. The actual driving force for a chemical reaction is a combination of enthalpy change and entropy change, so the interaction happened at favorable conditions because of $\Delta H < 0$ and $\Delta S > 0$. The above information is expected to be useful for studying the interactions of AAs and AA with other materials because the release energies as a result for hydrogen bond interactions, and so the role of hydrogen bond in molecular optical materials and drugs designing [18, 19], since the addition of inorganic salt (NaCl) to AAs:AA drug assist hydrogen bond interactions to form in a good manner and release energies, as a result, for the increase of AAs solubility and absorptivity.

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