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Thermodynamics of the Second Dissociation of a Substituted Aminopropanesulfonic Acid (TAPSO) from 5 °C to 55 °C

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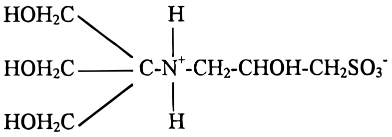
Electromotive force (emf) measurements of the cell without a liquid junction of the type Pt|H_2|TAPSO (m_1) + NaTAPSOate (m_2) + NaCl (m_3)|AgCl|Ag were made at intervals of 5 K from 5 °C to 55 °C, including 37 °C, with stoichiometric molalities $m_1 = m_2 = m_3$ ranging from 0.01 to 0.1 mol·kg^{-1}. At 25 °C, pK_2 is 7.635, and consequently buffer solutions of TAPSO and its salt (NaTAPSOate) are useful for pH control in the region pH 6 to 8. The second dissociation constant (pK_2) of TAPSO can be expressed by the equation 2254.0057/(T/K) − 4.9783 + 0.88672 ln(T/K), with a standard deviation of ±0.0008. The zwitterionic buffer TAPSO is represented as 3-[*N*-(tris(hydroxymethyl)methyl)amino]-2-hydroxypropanesulfonic acid, a derivative of the parent compound taurine.

Introduction

For useful study of buffers in the pH range 6−8, Good and co-workers (1966) recommended a series of zwitterionic buffer compounds compatible with most media of physiological and biochemical importance. These ampholytes were either amines or N-substituted amino acids. In a previous article (Roy et al., 1997), the thermodynamic dissociation constants and related thermodynamic quantities of two substituted aminoethanesulfonic acids ACES and CHES with zwitterionic character have been reported.

The search, since 1966 (Good et al., 1966), for better biological buffer compounds that can be used as either primary or secondary standards with respect to an isotonic saline solution (I = 0.16 mol·kg^{-1}), led to a discovery of seven new zwitterionic biochemical buffer substances (Ferguson et al., 1980). These compounds are aminoalkanesulfonates and exhibit improved properties in terms of the criteria (high solubility in water, stability in solution, minimum salt effects, etc.) for an excellent biological buffer. Although the preparation and properties of these seven new zwitterionic buffers were described (Ferguson et al., 1980), the pK_2 and pH values of these buffer substances have not, in general, been determined. Attention has been focused on one such compound, commonly known as TAPSO, for the isoelectric focusing method in analytical separation of protein over a pH gradient of 4−6. This ampholyte is named 3-[*N*-(tris(hydroxymethyl)methyl)amino]-2-hydroxypropanesulfonic acid and has the formula



To the authors' knowledge, no pK_2 data for this buffer substance are available in the literature. With a view to providing physiological pH buffer standards for clinical application at an ionic strength of 0.16 mol·kg^{-1} in the pH range 6−8, accurate knowledge of the second dissociation constant of TAPSO is a prerequisite.

Through our extended studies of the thermodynamics of the dissociation of zwitterionic compounds (Roy el al., 1977, 1979, 1973), we have now determined the pK_2 of TAPSO at 12 temperatures from (5 to 55) °C. From the data, the standard Gibbs energy ($\Delta G°$), enthalpy ($\Delta H°$), entropy ($\Delta S°$), and heat capacity ($\Delta C_p°$) changes for the dissociation process have been evaluated.

The emf cell, which had no liquid junction, can be represented by

$$\text{Pt}|\text{H}_2(\text{g, 1 atm})|\text{TAPSO }(m_1),\ \text{NaTAPSOate }(m_2),\ \text{NaCl }(m_3)|\text{AgCl, Ag} \quad \text{(A)}$$

The molalities m_1, m_2, and m_3 were equal in all of the buffer solutions.

Theory and Equations

The second dissociation step involves the deprotonation of the substituted methylammonium of TAPSO, and can be represented by

$$T^{\pm} \overset{K_2}{\rightleftharpoons} H^+ + T^- \quad (1)$$

where the zwitterion $(CH_2OH)_3CN^+H_2-CH_2-CH(OH)CH_2SO_3^-$ is designated $T^{\pm}$, T^- is the TAPSOate ion, and K_2 is thermodynamic equilibrium constant. The "apparent" dissociation constant pK_2' can be obtained from emf of cell (A) and is given by the following equation:

$$\text{p}K_2' = \text{p}K_2 - \log_{10}\{\gamma_{T^\pm}\gamma_{Cl^-}\}/\gamma_{T^-} \quad (2)$$

$$= (E - E°)F/RT \ln 10 + \log_{10}(m_1 m_3/m_2) \quad (3)$$

where $\gamma_T{}^{\pm}$ and $\gamma_T{-}$ represent the activity coefficients of the ampholyte TAPSO and its anion TAPSOate, respectively, E is the corrected emf at a hydrogen partial pressure of 101.325 kPa, listed in Table 1, and $E°$ is the standard potential of the silver−silver chloride electrode given in by Roy et al. (1997, 1992).

The pH of all of the cell solutions of TAPSO and NaTAPSOate for cell (A) was so close to neutrality that the buffer ratio m_1/m_2 remained at unity; no correction for hydrolysis in eq 3 is needed, even in the most dilute buffers included. In addition, the molalities of TAPSO, NaTAPSOate, and NaCl were equal in all of the buffer solutions.

As in earlier studies (Roy et al., 1997, 1977), pK_2' in eq 3 was found to vary linearly with ionic strength I at each temperature, and the intercept (pK_2) at I = 0 was readily

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Table 1. Electromotive Force (V) of the Cell Pt; H_2(g, 1 atm)|TAPSO (m_1), NaTAPSOate (m_2), NaCl (m_3)|AgCl, Ag from (5 to 55) °C

$m_1 = m_2 = m_3$/mol·kg^{-1}	t/°C											
	5	10	15	20	25	30	35	37	40	45	50	55
0.01	0.792 96	0.793 17	0.793 25	0.793 18	0.792 91	0.792 63	0.792 11	0.791 91	0.791 53	0.790 78	0.789 96	0.788 98
0.01	0.792 99	0.793 21	0.793 27	0.793 09	0.792 94	0.792 51	0.792 19	0.791 99	0.791 35	0.790 68	0.789 85	0.788 90
0.02	0.776 79	0.776 44	0.776 32	0.776 02	0.775 43	0.774 91	0.774 20	0.773 81	0.773 23	0.772 22	0.771 17	0.769 90
0.02	0.776 70	0.776 60	0.776 43	0.775 97	0.775 54	0.774 82	0.774 13	0.773 80	0.773 14	0.772 11	0.771 01	0.769 78
0.03	0.767 41	0.767 00	0.766 66	0.766 15	0.765 47	0.764 69	0.763 85	0.763 46	0.762 71	0.761 56	0.760 27	0.758 93
0.03	0.767 35	0.767 08	0.766 78	0.766 09	0.765 55	0.764 60	0.763 75	0.763 37	0.762 61	0.761 45	0.760 18	0.758 83
0.04	0.760 84	0.760 40	0.760 04	0.759 28	0.758 57	0.757 51	0.756 65	0.756 21	0.755 39	0.754 14	0.752 74	0.751 26
0.04	0.760 90	0.760 38	0.759 90	0.759 19	0.758 57	0.757 58	0.756 53	0.756 10	0.755 35	0.754 01	0.752 62	0.751 22
0.06	0.755 85	0.755 31	0.754 81	0.753 93	0.753 18	0.752 16	0.751 00	0.750 53	0.749 74	0.748 37	0.746 97	0.745 48
0.06	0.755 94	0.755 29	0.754 75	0.754 05	0.753 17	0.752 06	0.751 14	0.750 67	0.749 65	0.748 32	0.746 86	0.745 34
0.08	0.751 77	0.751 13	0.750 53	0.749 61	0.748 78	0.747 70	0.746 67	0.746 17	0.745 26	0.743 89	0.742 24	0.740 83
0.08	0.751 91	0.751 23	0.750 63	0.749 75	0.748 90	0.747 79	0.746 57	0.746 08	0.745 16	0.743 74	0.742 34	0.740 69
0.09	0.745 65	0.744 87	0.744 14	0.743 14	0.742 18	0.740 98	0.739 73	0.739 19	0.738 23	0.736 90	0.735 23	0.733 69
0.09	0.745 74	0.744 97	0.744 28	0.743 23	0.742 17	0.741 07	0.739 85	0.739 31	0.738 32	0.736 80	0.735 13	0.733 44
0.10	0.741 11	0.740 21	0.739 37	0.738 27	0.737 33	0.735 92	0.734 65	0.734 07	0.733 03	0.731 69	0.729 93	0.728 21
0.10	0.741 23	0.740 22	0.739 47	0.738 38	0.737 26	0.736 06	0.734 74	0.734 17	0.733 14	0.731 60	0.729 94	0.728 21

Table 2. Second Dissociation Constants of TAPSO from (5 to 55) °C: Slope Parameters β and the Standard Deviation of the Intercept $\sigma(\mathrm{p}K_2)$ and Slope Parameters $\sigma(\beta)$

t/°C	pK_2	σ(pK_2)	β/kg·mol^{-1}	$\sigma(\beta)$/kg·mol^{-1}
5	8.1159	0.0005	0.337	0.005
10	7.9882	0.0005	0.322	0.005
15	7.8664	0.0005	0.325	0.005
20	7.7479	0.0005	0.319	0.004
25	7.6347	0.0005	0.328	0.004
30	7.5244	0.0005	0.330	0.004
35	7.4188	0.0004	0.334	0.004
37	7.3757	0.0004	0.336	0.004
40	7.3148	0.0005	0.336	0.004
45	7.2159	0.0005	0.355	0.004
50	7.1193	0.0005	0.357	0.004
55	7.0291	0.0005	0.374	0.005

obtained by linear regression analysis:

$$\mathrm{p}K_2' = \mathrm{p}K_2 + \beta I \qquad (4)$$

where β is the slope parameter and I is given by

$$I = m_2 + m_3 \qquad (5)$$

The simplest form of eq 3 results from the assumption that the activity coefficient term $\log(\gamma_{\mathrm{T}^{\pm}}\gamma_{\mathrm{Cl}^-}/\gamma_{\mathrm{T}^-})$ will be small in very dilute solutions and will vary linearly with ionic strength at high concentrations. The values of pK_2 and β together with their standard deviations for TAPSO are summarized in Table 2.

Experimental Section

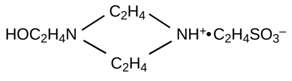
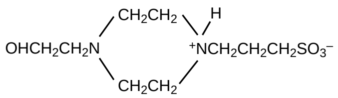
A commercial sample of TAPSO was obtained from Research Organics Inc. (Cleveland, OH) and was purified by recrystallization from 70 mass % ethanol. The product assayed 99.97% (std dev = 0.01) when titrated with a standard solution of sodium hydroxide under carbon dioxide-free conditions. For the determination of pK_2, 16 buffer solutions containing a 1:1 molar ratio of TAPSO to its salt NaTAPSOate, were prepared by mixing accurately known masses of TAPSO, a standard solution of carbon dioxide-free sodium hydroxide to make NaTAPSOate, recrystallized sodium chloride, and carbon dioxide-free double-distilled water. Vacuum corrections were applied to all weighings.

The emf method used is essentially that of Harned and Ehlers (1932), and the procedures were the same in almost all respects as in our previous studies (Roy et al., 1997, 1977, 1973). The emf measurements of cell A were made at intervals of 5 K from 5 °C to 55 °C using a Keithley Model 191 digital voltmeter recording to the nearest 0.01 mV. The design of all-glass cells (Gary et al., 1964) and the preparation of the hydrogen electrodes and the silver−

Table 3. Thermodynamic Quantities with Uncertainties for the Dissociation of TAPSO at (5, 25, 37, and 55) °C

	t/°C			
	5	25	37	55
$\Delta G°$/J·mol^{-1}	43 218 ± 4	43 574 ± 3	43 809 ± 4	44 151 ± 4
$\Delta H°$/J·mol^{-1}	38 430 ± 117	39 091 ± 38	37 887 ± 40	37 582 ± 124
$\Delta S°$/J·mol^{-1}·K^{-1}	−17.89 ± 0.39	−19.12 ± 0.13	−19.82 ± 0.13	−20.81 ± 0.38
$\Delta C_p°$/J·mol^{-1}·K^{-1}	−17 ± 4.0	−17 ± 4	−17 ± 4	−17 ± 4

Table 4. Thermodynamic Quantities at 298.15 K for the Dissociation of a Series of Structurally Related Compounds in Water

structure	common name	pK_2	$\Delta H°_{\mathrm{m}}$/J·mol^{-1}	$\Delta S°_{\mathrm{m}}$/J·K^{-1}·mol^{-1}	$\Delta C°_{p,\mathrm{m}}$/J·K^{-1}·mol^{-1}	ref
$O_3^-S(H_2C)_2NH_3^+$	TAURINE	9.061	41 860	−33.05	−33	King, 1951
$O_3^-S(H_2C)_2{\cdot}NH^+(C_2H_4OH)_2$	BES	7.187	24 184	−56.48	4	Roy et al., 1977
		7.187	24 393	−55.65	0	Vega and Bates, 1976
	HEPES	7.565	20 376	−76.57	46	Feng et al., 1989
		7.562	20 041	−77.49	25	Vega and Bates, 1976
	HEPPS	7.957	21 168	−84.50	48	Roy et al., submitted 1997
$(HOCH_2)_3C^+NH_2CH_2CH(OH)CH_2SO_3^-$	TAPSO	7.635	38 091	−19.12	−17	this work

silver chloride electrodes (thermal electrolytic type) have been described elsewhere (Bates, 1973). The bias potentials of the silver−silver chloride electrodes were always within ±0.05 mV. The emf readings at 25 °C were recorded at the beginning, in the middle, and sometimes at the end of the temperature series. These measurements agreed well, within 0.04 mV; thus the cells showed excellent stability. The temperature was regulated within ±0.005 °C with a digital thermometer (Guideline Model 9540).

Results and Discussion

Values of pK_2 and β listed in Table 2 at 12 temperatures were found by the method of linear regression analysis using the relation set forth in eq 4. The experimental values of pK_2 from Table 2 at the 12 temperatures fitted well by the method of least squares to an equation of the form recommended by Ives and Moseley (1976):

$$pK_2 = 2254.0057/(T/\mathrm{K}) - 4.9783 + 0.88672 \ln(T/\mathrm{K}) \quad (6)$$

where T, the thermodynamic temperature, lies between (278.15 and 328.15) K. The standard deviation of regression was 0.0008. The standard thermodynamic functions, standard Gibbs energy ($\Delta G°$), enthalpy ($\Delta H°$), entropy ($\Delta S°$), and heat capacity ($\Delta C_p°$) changes, characterizing the dissociation of the zwitterionic TAPSO were calculated by the usual thermodynamic formulas (Roy et al., 1997) from the constants of eq 6. These values are given in Table 3 at (5, 25, 37, and 55) °C. The estimates for the uncertainties of the standard deviations are also listed in Table 3 and are calculated by the method of propagation of error. A comparison of the values of the thermodynamic quantities of some commonly used structurally related zwitterionic buffers such as taurine and glycine (King, 1951), BES (Roy et al., 1977; Vega and Bates, 1976), HEPPS (Roy et al., submitted 1977), and HEPES (Feng et al., 1989; Vega and Bates, 1976) is shown in Table 4.

It is evident from a comparison of pK_2 values of TAPSO at 25 °C with those of the parent compound taurine and glycine (King, 1951) and HEPPS (Roy et al., submitted) that substitution of three hydroxymethyl groups lowers pK_2. Thus, the acidity (removal of H^+ from the nitrogen atom) of TAPSO is enhanced possibly due to (i) steric effects since the substituent is present adjacent to the protonated nitrogen but relatively remote from the sulfonic group and (ii) the inductive effect of the oxygen atom in the hydroxymethyl group. The lowering of pK_2 is paralleled by the decrease of $\Delta G°$ and $\Delta H°$ for the isoelectric dissociation process. Another probable explanation for this decrease in $\Delta H°$ is that these relatively bulky hydrophilic groups (hydroxymethyl or hydroxyethyl) partially shield the basic nitrogen atom; hence interactions with hydrogen ions in N-substituted compounds become more difficult. The negative values of $\Delta S°$ may indicate stabilization of the solvent structure (increased order) in the vicinity of the molecules of uncharged base (the zwitterion TAPSO is assumed to behave as an uncharged molecule). The small negative value of $\Delta C_p°$ may signify some change in the solvation pattern for the dissociation process, as is also the case for tricine (Roy et al., 1973).

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