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# THE SOLUBILITY OF THALLOUS CHLORIDE IN THE PRESENCE OF EDESTIN NITRATE[1]

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One may well begin a discussion of the interaction of salts and proteins in solution by recalling the fact that thermodynamic reasoning cannot decide from measurements of ion activities and concentrations whether there is combination between salt and protein, or whether a decreased activity is due to interionic attraction. We shall set $\gamma = a/m$, where $a$ stands for activity and $m$ for molality, without making any assumption as to how changes in $a$ are brought about. Some particular hypothesis may, however, appear more probable in the light of other physical theories or by analogy with simpler systems.

It will be shown later that edestin changes the mean activity coefficient of thallous chloride

$$\gamma_{\pm} = \sqrt{\frac{a_{Tl^+} a_{Cl^-}}{m^2}} = \sqrt{\gamma_{Tl^+} \gamma_{Cl^-}}$$

by about the same amount as an equal edestin concentration changes $\gamma_{Cl^-}$. From this we may conclude that $\gamma_{Tl^+}$ is affected by the addition of edestin equally with $\gamma_{Cl^-}$. An equal binding of $Cl^-$ and $Tl^+$ in spite of considerable hydrogen ion changes seems unlikely. In opposition to the above may be cited the findings of Northrop and Kunitz,[2] who observed no influence of gelatin on the activity of $Li^+$, $Na^+$, $K^+$, $NO_3^-$ or $SO_4^=$ although these ions should be sensitive to changes in ion atmosphere or dielectric constant if the chloride activity variations be due to these factors.

## Experimental

The thallous chloride in this work was the same preparation used by the author in a previous investigation,[3] from which paper values for the solubility in 0.025 molal nitric acid and 0.050 molal nitric acid are taken. The edestin, extracted from hemp seed with 10% sodium chloride, was recrystallized five times from 10% sodium chloride by diluting with three volumes of water at 50° and allowing to cool, then recrystallized twice from 7.5% potassium nitrate by the same method. It was washed free from nitrate by suspending five times in water. Experiments with sodium chloride had shown that this would reduce the salt content to a point where its contribution to the ionic strength could be neglected. The preparation was chloride free.

[1] This work was aided by a grant from The Chemical Foundation.

[2] Northrop and Kunitz, *J. Gen. Physiol.*, **11**, 481 (1928).

[3] Failey, THIS JOURNAL, **54**, 576 (1932).

Determinations of the solubility at 25.00 ± 0.01° were carried out as described in reference 3 except for the method of analysis. The following procedure gave maximum errors of 0.3% on protein solutions of known chloride content. To weighed samples of approximately 100 g. a 50% excess of silver nitrate was added, then 100 ml. of concentrated chloride-free nitric acid. The flasks were digested for twenty-four hours on the steam-bath with the addition of a few drops of caprylic alcohol to prevent foaming. After cooling the silver chloride was washed thoroughly by decantation and weighed in Gooch crucibles with porous porcelain bottoms. Thallous chloride was shown by analysis to be the solid phase in the most concentrated protein solution.

## Discussion

The results are tabulated in Table I where the first column indicates grams of edestin per 1000 g. of water; the second, molality of nitric acid; the third, thallous chloride solubility in moles per 1000 g. of water; the fourth, $-\log \gamma_{\pm}$; and the fifth, $\gamma_{\pm}$.

The activity coefficients were obtained by means of the equation $-\log \gamma_{\pm} = \log (1/m_{\pm}^{\circ}) - \log (1/m_{\pm})$ on the assumption that $\log (1/m_{\pm}^{\circ}) = 1.8630$.[4]

TABLE I

| Edestin, g. | $HNO_3$ | $S \times 10^3$ | $-\text{Log } \gamma_{\pm}$ | $\gamma_{\pm}$ |
|---|---|---|---|---|
| 0 | 0.025 | 1734 | 0.102 | 0.791 |
| 4.5 | .025 | 1763 | .109 | .778 |
| 13.4 | .025 | 1802 | .119 | .761 |
| 13.8 | .025 | 1827 | .125 | .733 |
| 30.1 | .025 | 1924 | .148 | .710 |
| 34.7 | .025 | 1948 | .153 | .703 |
| 48.8 | .025 | 2043 | .173 | .691 |
| 0 | .050 | 1821 | .123 | .753 |
| 5.2 | .050 | 1842 | .128 | .745 |
| 10.1 | .050 | 1861 | .133 | .736 |
| 24.2 | .050 | 1953 | .154 | .702 |
| 28.1 | .050 | 1971 | .158 | .695 |
| 45.9 | .050 | 2131 | .192 | .643 |

Figure 1 is a plot of Table I with $-\log \gamma_{\pm}$ as ordinate and g. of edestin per 1000 g. of water as abscissa. It may be seen that the points for each acid concentration fall approximately on a straight line. Roughly we may set $-\Delta \log \gamma_{\pm}/g = 1.5 \times 10^{-3}$ where $g$ is grams of edestin per 1000 g. of water. The addition of 10.7 g. of edestin has the same effect on $\gamma_{\pm}$ TlCl as 0.1 mole = 7.5 g. of glycine.[3]

Pauli and Stenzinger[5] have measured the solubility of calcium sulfate in protein solutions. Values of $-\Delta \log \gamma_{\pm}/g$ calculated from their data are shown in Table II.

[4] "International Critical Tables," Vol. VII, p. 319.

[5] Pauli and Stenzinger, *Biochem. Z.*, **205**, 71 (1929).

TABLE II

| Protein | Salt or ion | $\frac{-\Delta \log \gamma_{\pm}}{g} \times 10^3$ | Reference |
|---|---|---|---|
| Edestin | TiCl | 1.5 | This paper |
| Serum albumin | $CaSO_4$ | 1.5 | 5 |
| Pseudoglobulin | $CaSO_4$ | 3.9 | 5 |
| Hemoglobin | $CaSO_4$ | 4.6 | 5 |
| Edestin | $Cl^-$ | 1.7 | 6 |
| Gelatin | $Cl^-$ | 0.9 | 6 |
| Gelatin | $Cl^-$ | 1.3 | 2 |

The e. m. f. measurements of Hitchcock[6] gave results with which the above may be compared. Hitchcock stated that his experiments agree with the assumption that in 0.1 molal hydrochloric acid 1 g. of edestin

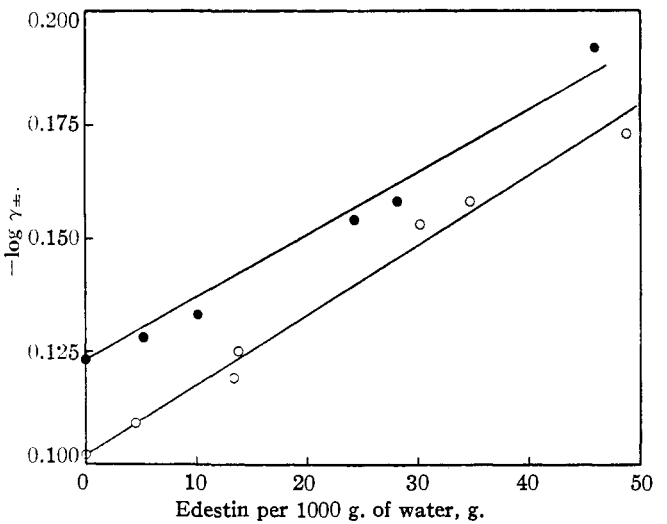


Fig. 1.—○, edestin + $HNO_3$, $M/40$; ●, edestin + $HNO_3$, $M/20$.

combines with a maximum of $3.9 \times 10^{-4}$ mole of $Cl^-$ and 1 g. of gelatin combines with $2.0 \times 10^{-4}$ mole of $Cl^-$. This would make $a_{Cl^-}$ and also $\gamma_{Cl^-}$ a linear function of $g$. For small changes in $\gamma_{Cl^-}$ it would be true that log $\gamma_{Cl^-}$ is also linear. Then for edestin we may set

$$\frac{-\Delta \log \gamma_{Cl^-}}{g} = \frac{\log 0.1 - \log (0.1 - 3.9 \times 10^{-4} g)}{g} = 1.7 \times 10^{-3}$$

For gelatin the value is $0.9 \times 10^{-3}$.

Northrop and Kunitz[2] concluded from measurements of membrane equilibria that gelatin may bind approximately $3 \times 10^{-4}$ mole of $Cl^-$ in $CuCl_2$, per g. of protein. Then $-\Delta \log \gamma_{Cl^-}/g = 1.3 \times 10^{-3}$.

The data of Stadie and Sunderman[7] on the freezing point of solutions

[6] Hitchcock, *J. Gen. Physiol.*, **14**, 99 (1930); *ibid.*, **15**, 125 (1931).

[7] Stadie and Sunderman, *J. Biol. Chem.*, **91**, 227 (1931).

of hemoglobin to which sodium hydroxide has been added offer an opportunity for calculating changes in $\gamma_{Na^+}$ if one makes the necessary assumption that the authors have been able to "correct for ΔHb." This assumption postulates that in the four term Duhem equation

$$d \ln a_1 = -\frac{m_2}{55.51} d \ln \gamma_2 m_2 - \frac{m_3}{55.51} d \ln \gamma_3 m_3 - \frac{m_4}{55.51} d \ln \gamma_4 m_4$$

where the subscript 1 refers to $H_2O$, 2 to $Na^+$, 3 to $OH^-$ and 4 to Hb, the $m_3$ term is negligible and the $m_4$ term is unaffected by changes in $\gamma_4$. The authors find on adding sodium hydroxide to Hb and also on diluting a solution of NaHb of given $Na^+/Hb^-$ ratio that the observed freezing point depression ascribed to $Na^+$ is equal to 0.75 the theoretical within the limits of error, or $\varphi_{Na} = 0.75$. On the assumptions made as to $a_{Hb}$ and $a_{OH}$ the Duhem equation reduces to

$$-d \ln a_1 = \frac{m_2}{55.51} d \ln \gamma_2 m_2$$

Following the treatment of Lewis and Randall,[8] observing that $j = 1 - \varphi_{Na^+} = 0.25$ and that $dj = 0$, we have for these small freezing point depressions

$$d \ln \frac{a_2}{m_2} = -j \, d \ln m_2 = -0.25 \, d \ln m_2$$

or, changing to the base 10 and integrating between $m = 0.01$ and any given molality

$$\log \frac{\gamma_2}{\gamma_2^{\#}} = -0.25 (\log m_2 - \log 0.01)$$

where $\gamma_2^{\#}$ is the value of the activity coefficient when $m_2 = 0.01$. If we assume $\gamma_2^{\#} = 0.90$, which is an average value for $\gamma_{\pm}$ in 0.01 molal salt solutions, the following values are obtained for $Na^+$ in the presence of $Hb^-$.

TABLE III

| $m_2$ | $\gamma_2/\gamma_2^{\#}$ | $\gamma^2$ |
|---|---|---|
| 0.01 | 1.00 | 0.900 |
| .02 | .841 | .757 |
| .03 | .760 | .684 |
| .05 | .669 | .602 |
| .08 | .593 | .535 |
| .10 | .558 | .502 |

TABLE IV

| C | $\gamma_{Cl^-}$ | $\gamma_{Na^+}$ |
|---|---|---|
| 0.0125 | 0.86 | 0.87 |
| .0128 | .89 | .87 |
| .0178 | .80 | .78 |
| .0361 | .62 | .66 |
| .0404 | .65 | .64 |
| .0405 | .59 | .64 |

These may be compared with the results of Adair[9] on solutions of edestin chloride. The first column given salt concentration; the second, Adair's values for $\gamma_{Cl^-}$; and the third, $\gamma_{Na^+}$ interpolated from Table III.

Tables III and IV are plotted in Fig. 2 together with values of $\gamma_{\pm}$ in sodium chloride solution for comparison.[10] The calculations show that

[8] Lewis and Randall, "Thermodynamics," 1923, p. 285.

[9] Adair, *Proc. Roy. Soc.* (London), **A120**, 573 (1928).

[10] Taylor, "Treatise on Physical Chemistry," 1931, Vol. I, p. 772.

with a constant $\varphi_{Na^+}$ there is associated a considerable change in $\gamma_{Na^+}$ as the molality is varied. Figure 2 indicates the agreement between the work of Adair and that of Stadie and Sunderman, although the latter authors, assuming that constant $\varphi$ implied constant $\gamma$, found contradictions. It may finally be pointed out that Adair's values would give a much greater $-\Delta\log \gamma_{Cl^-}/g$ than any shown in Table II.

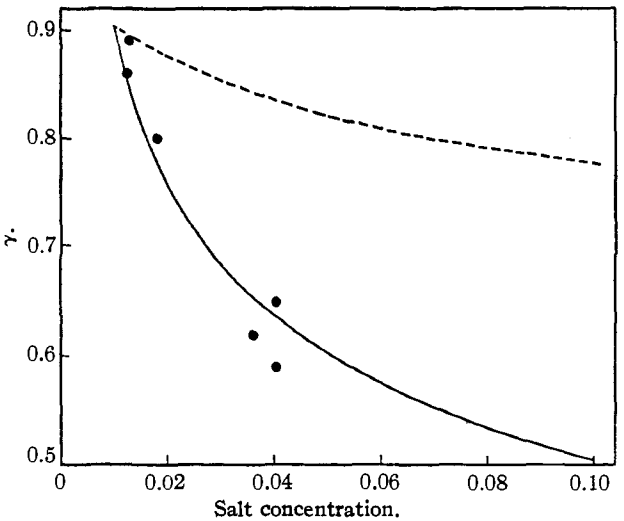


Fig. 2.—The points are Adair's values for $\gamma_{Cl^-}$ in edestin chloride. The full line represents $\gamma_{Na^+}$ in the presence of hemoglobin assuming $\varphi_{Na^+} = 0.75$ and that $\gamma_{Na^+} = 0.90$ when $c = 0.01$. For comparison the broken line gives $\gamma_{\pm}$ in aqueous sodium chloride solutions.

## Summary

The solubility of thallous chloride in solutions of edestin nitrate has been determined. The negative logarithm of the activity coefficient of thallous chloride is approximately a linear function of grams of edestin per 1000 g. of water. Some existing data on the activity of salts in the presence of protein have been compared.

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