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Self-Associating Systems. I. Multinomial Theory for Ideal Systems*

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ABSTRACT: A general expression for the calculation of the equilibrium constants of self-associating ideal systems has been established. The derivations make use of the multinomial theorem. This general expression is applicable to associating systems of any degree of polymerization irrespective of whether the weight-, M_w or the number-average, M_n, molecular weight is used in the calculations. Using this expression, the equilibrium constants, from K_1 to K_4, for self-associating systems have been derived.

Many substances in solution behave as self-associating systems. The biological activity of some macromolecules, *e.g.*, subunit-type enzymes, appears to be closely related to their state of aggregation. The need to clarify some aspects of this relation in some of the systems currently under study in our laboratory was the main stimulus for this work. A theory will be presented that will allow by means of a single general expression the analysis of experiments that supply molecular weight averages, irrespective of whether these are either the weight average or the number average. A fundamental step in this work is the use of the multinomial theorem (see Parzen, 1960) and for this reason it is designated as the multinomial theory.

Theoretical

We are going to examine self-associating reactions of the type

$$nP_1 \rightleftarrows a_2P_2 + a_3P_3 + \ldots + a_mP_m, \text{ with } \sum_{i=2}^{m} ia_i = n \quad (1)$$

It is assumed that all species participating in the self-associating reaction have the same partial specific volume ($\bar{v}_1 = \bar{v}_2 = \ldots = \bar{v}$), the same refractive index increment $[(dn/dc_1)_{T,P} = (dn/dc_2)_{T,P} = \ldots = (dn/dc)_{T,P}]$, and that the activity coefficient of each associating species can be represented as

$$\ln y_i = iBM_1c + \text{higher powers in } c \quad (2)$$

where B is the virial coefficient, M_1 is the molecular weight of the monomer, and c is the total solute concentration. The higher powers in c are neglected. In the particular case $i = 1$, it is

$$\ln y_1 = BM_1c \quad (2a)$$

For ideal systems, $BM_1 = 0$. Using eq 2 and 2a and the nomenclature of Adams and Fujita (1963), the condition for chemical equilibrium can be stated in terms of the equilibrium constants, K_i, as

$$c_i = K_i\frac{y_1^{\,i}}{y_i}c_1^{\,i} = K_ic_1, \qquad i = 1, 2, \ldots \quad (3)$$

Consequently, $K_1 = 1$. Also, since

$$c = \sum_{i=1}^{m} c_i, \qquad i = 1, 2, \ldots \quad (4b)$$

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we can write

$$c = \sum_{i=1}^{m} K_i c_1^{\,i} \qquad (4a)$$

Differentiating eq 4a and rearranging we have

$$\frac{dc}{dc_1} = \sum_{i=1}^{m} i K_i c_1^{\,i-1} \qquad (4)$$

The weight-average molecular weight, M_{wc}, is now defined following a suggested nomenclature of Adams and Williams (1964); the subscript c is added to M_w (or M_n) to indicate a concentration-dependent quantity. Thus

$$M_{wc} = \frac{\sum_{i=1}^{m} c_i M_i}{\sum_{i=1}^{m} c_i} \qquad (5)$$

where M_i is the molecular weight of species i. Using eq 4a and rearranging, eq 5 becomes

$$\frac{M_1}{M_{wc}} = \frac{\sum_{i=1}^{m} K_i c_1^{\,i-1}}{\sum_{i=1}^{m} i K_i c_1^{\,i-1}} \qquad (5a)$$

Substituting eq 4 for the denominator of eq 5a and rearranging we have

$$\frac{M_1 dc}{M_{wc}} = \sum_{i=1}^{m} K_i c_1^{\,i-1} dc_1 \qquad (6)$$

which after integration becomes

$$\int_0^c \frac{M_1 dc}{M_{wc}} = \sum_{i=1}^{m} K_i \int_0^{c_1(c)} c_1^{\,i-1}\, dc_1 = \sum_{i=1}^{m} \frac{K_i}{i} c_1^{\,i} \qquad (7)$$

The number-average molecular weight, M_{nc} is defined by

$$M_{nc} = \frac{\sum_{i=1}^{m} n_i M_i}{\sum_{i=1}^{m} n_i} = \frac{\sum_{i=1}^{m} c_i}{\sum_{i=1}^{m} \frac{c_i}{M_i}} = \frac{c}{\sum_{i=1}^{m} \frac{c_i}{M_i}} \qquad (8)$$

where n_i is the number of moles of species i. Equation 8 shows that M_n is a function of c and therefore of position in the cell only. Rearranging eq 8 and using eq 3 we have

$$\frac{cM_1}{M_{nc}} = \sum_{i=1}^{m} \frac{K_i c_1^{\,i}}{i} \qquad (9)$$

Comparing with eq 7, we have

$$F(c) = \frac{M_1 c}{M_{nc}} = \int_0^c \frac{M_1 dc}{M_{wc}} = \sum_{i=1}^{m} \frac{K_i}{i} c_1 \qquad (10)$$

Equation 10 shows that M_{nc} can be obtained by graphical integration from a plot of M_1/M_w *vs.* c (Adams, 1965a,b). Further, this equation can be used to derive a general analytical expression from which the values of K_i ($i = 1, 2, \ldots m$) can be obtained. Expanding $F(c)$ in powers of c (Maclaurin's theorem) and using eq 4a and 10, we have

$$F(c) = \sum_{r=0}^{\infty} \frac{1}{r!}\left(\frac{d^{(r)}F}{dc^r}\right)_{c=0} \left[\sum_{i=1}^{m} K_i c_1^{\,i}\right]^r = \sum_{i=1}^{m} \frac{K_i}{i} c_1^{\,i} \qquad (11)$$

The power term in brackets can be evaluated by making use of the multinomial theorem as follows

$$\left[\sum_{i=1}^{m} K_i c_1^{\,i}\right]^r = \sum_{\alpha_1=0}^{r} \sum_{\alpha_2=0}^{r} \cdots \sum_{\alpha_m=0}^{r} \binom{r}{\alpha_1, \alpha_2, \ldots, \alpha_m} \times K_1^{\alpha_1} K_2^{\alpha_2} \ldots K_m^{\alpha_m} c_1^{\alpha_1} c_1^{2\alpha_2} \ldots c_1^{m\alpha_m} = \sum_{\alpha_1=0}^{r} \sum_{\alpha_2=0}^{r} \cdots \sum_{\alpha_m=0}^{r} \binom{r}{\alpha_1, \alpha_2, \ldots, \alpha_m} \prod_{i=1}^{m} K_i^{\alpha_i} (c_1)^{\sum_{i=1}^{m} i\alpha_i} \qquad (12)$$

where $\alpha_1, \alpha_2, \ldots, \alpha_m$ take all positive integral values for which

$$\sum_{i=1}^{m} \alpha_i = r$$

Defining

$$\xi = \sum_{i=1}^{m} i\alpha_i = \alpha_1 + \sum_{i=2}^{m} i\alpha_i$$

or

$$\alpha_1 = \xi - \sum_{i=2}^{m} i\alpha_i$$

eq 12 can be rewritten

$$\left[\sum_{i=1}^{m} K_i c_1^{\,i}\right]^r = \sum_{\xi - \sum_{i=2}^{m} i\alpha_i = 0}^{r} \cdots \sum_{\alpha_m=0}^{r} \left(\left(\xi - \sum_{i=2} i\alpha_i\right), {}^{r}\alpha_2, \ldots, \alpha_m\right) \times K_1^{\xi - \sum_{i=2} i\alpha_i} \prod_{i=2}^{m} K_i^{\alpha_i} c_1^{\xi} \qquad (12a)$$

with the conditions

$$\xi - \sum_{i=2}^{m} i\alpha_i + \sum_{i=2}^{m} \alpha_i = \xi - \sum_{i=2}^{m} (i-1)\alpha_i = r$$

condition 1a

or

$$\xi - r = \sum_{i=2}^{m} (i-1)\alpha_i \qquad \text{condition 1}$$

and

$$\xi - \sum_{i=2}^{m} i\alpha_i \geqslant 0 \qquad \text{condition 2}$$

and

$$\sum_{i=1}^{m} \alpha_i = r \qquad \text{condition 3}$$

However, since by definition $K_1 = 1$, the factor

$$K_1^{\xi - \sum_{i=1}^{m} i\alpha_i}$$

can be eliminated from eq 12a. In addition, examination of eq 12a and its limiting conditions indicates (a) that the smallest value ξ can take is that resulting when

$$\sum_{i=2}^{m} (i - 1)\alpha_i = 0$$

since in any other case it would be greater than r; (b) the largest value ξ can take is mr. Equation 12a can then be written as

$$\left[\sum_{i=1}^{m} K_i c_1^{\,i}\right]^r = \sum_{\xi=r}^{mr} \left\{ \sum_{\alpha_2=0}^{r} \sum_{\alpha_m=0}^{r} \binom{r}{\left(\xi - \sum_{i=2}^{m} i\alpha_i\right), \alpha_2, \alpha_3, \ldots, \alpha_m} \times \prod_{i=2}^{m} K_i^{\alpha_i} \right\} c_1^{\xi} \quad (12b)$$

Introducing eq 12b into eq 11 we have

$$F(c) = \sum_{r=0}^{\infty} \frac{1}{r!}\left(\frac{d^{(r)}F}{dc^r}\right)_{c=0} \sum_{\xi=r}^{mr} G(\xi, r, K_i) c_1^{\xi} = \sum_{i=1}^{m} \frac{K_i}{i} c_1^{\,i} \quad (11a)$$

where

$$\sum_{\alpha_2=0}^{r} \cdots \sum_{\alpha_m=0}^{r} \binom{r}{\left(\xi - \sum_{i=2}^{m} i\alpha_i\right), \alpha_2, \ldots, \alpha_m} \times \prod_{i=2}^{m} K_i^{\alpha_i} = G(\xi, r, K_i)$$

and with conditions 1a, 2, and 3 applicable as in eq 12a. Equation 11a can be rewritten as

$$\sum_{r=0}^{\infty} \sum_{\xi=r}^{mr} \frac{1}{r!}\left(\frac{d^{(r)}F}{dc^r}\right)_{c=0} G(\xi, r, K_i) c_1^{\xi} = \sum_{\xi=1}^{m} \frac{K_\xi}{\xi} c_1^{\xi} \quad (11b)$$

Comparing the powers of c_1 on both sides of the equal sign in eq 11b, it can be seen that there will be nonvanishing terms only for values of r such that $1 \leqslant r \leqslant m$ and for values of ξ between r and m. Equation 11b can then be rewritten as

$$\sum_{r=1}^{m} \sum_{\xi=r}^{m} \frac{1}{r!}\left(\frac{d^{(r)}F}{dc^r}\right)_{c=0} G(\xi, r, K_i) c_1^{\xi} = \sum_{\xi=1}^{m} \frac{K_\xi}{\xi} c_1^{\xi} \quad (11c)$$

Further, upon changing the order of summation in the double sum of eq 11c we find the formal equivalence

$$\sum_{r=1}^{m} \sum_{\xi=r}^{m} = \sum_{\xi=1}^{m} \sum_{r=1}^{\xi}$$

Then, we can rewrite eq 11d as

$$\sum_{\xi=1}^{m} \sum_{r=1}^{\xi} \frac{1}{r!}\left(\frac{d^{(r)}F}{dc^r}\right)_{c=0} G(\xi, r, K_i) c_1^{\xi} = \sum_{\xi=1}^{m} \frac{K_\xi}{\xi} c_1^{\xi} \quad (11d)$$

Comparing the coefficients of the same power of c on both sides of eq 11d we have

$$\sum_{r=1}^{\xi} \frac{1}{r!}\left(\frac{d^{(r)}F}{dc^r}\right)_{c=0} G(\xi, r, K_i) = \frac{K_\xi}{\xi}, \xi = 1, 2, \ldots, m \quad (13a)$$

or

$$\sum_{r=1}^{\xi} \frac{1}{r!}\left(\frac{d^{(r)}F}{dc^r}\right)_{c=0} \sum_{\alpha_2=0}^{r} \cdots \sum_{\alpha_m=0}^{r} \times \binom{r}{\left(\xi - \sum_{i=2}^{m} i\alpha_i\right), \alpha_2, \ldots, \alpha_m} \prod_{i=2}^{m} K_i^{\alpha_i} = \frac{K_\xi}{\xi} (\xi = 1, 2, \ldots, m) \quad (13)$$

with conditions 1, 2, and 3 applicable as before. Expression 13 represents a set of equations from which the equilibrium constants K_1, K_2, $\ldots$, K_m can be determined.

Application of the Theory to Calculation of Equilibrium Constants. The general expression 13 allows the derivation of explicit equations for the calculation of the equilibrium constants K_ξ, $\xi = 1, 2, \ldots, m$, of ideal self-associating systems of the type defined by eq 1. In addition to the calculation of these particular cases, a derivation will be made in some detail as an illustration of the procedure, for the cases K_1, K_2, K_3, and K_4. The calculation of explicit expressions for any desired equilibrium constant or constants will then be readily obtainable. Further, the final equations presented here will appear exclusviely in terms of the experimentally available quantities M_{wc} or M_{nc}. Examining eq 13 we can see that $r!$ is simultaneously multiplying all summation terms (since it is in the multinomial coefficient) and dividing these terms as well ($1/r!$ precedes the derivative); the number $r!$ can be deleted from eq 13. Also, noting that

$$\left(\frac{d^{(r)}F}{dc^r}\right)_{c=0} = \left(\frac{d^{(r-1)}\left(\dfrac{M_1}{M_w}\right)}{dc^{r-1}}\right)_{c=0} = \left(\frac{r d^{(r-1)}\left(\dfrac{M_1}{M_n}\right)}{dc^{r-1}}\right)_{c=0} \quad (14)$$

one can substitute either the second or the third form of eq 14 for the first into eq 13. Thus eq 13 becomes

$$\sum_{r=1}^{\xi}\left(\frac{\mathrm{d}^{(r-1)}\left(\frac{M_1}{M_w}\right)}{\mathrm{d}c^{r-1}}\right)_{c=0}\left[\sum_{\alpha_2=0}^{r}\cdots\sum_{\alpha_m=0}^{r}\times\left(\frac{1}{\left(\xi-\sum_{i=2}^{m}i\alpha_i\right),\alpha_2,\ldots,\alpha_m}\right)\prod_{i=2}^{m}K_i^{\alpha_i}\right]=\frac{K_\xi}{\xi}\qquad(15a)$$

or

$$\sum_{r=1}^{\xi}\left(\frac{r\mathrm{d}^{(r-1)}\left(\frac{M_1}{M_n}\right)}{\mathrm{d}c^{r-1}}\right)_{c=0}\left[\sum_{\alpha_2=0}^{r}\cdots\sum_{\alpha_m=0}^{r}\times\left(\frac{1}{\left(\xi-\sum_{i=2}^{m}i\alpha_s\right),\alpha_2,\ldots,\alpha_m}\right)\prod_{i=2}^{m}K_i^{\alpha_i}\right]=\frac{K_\xi}{\xi}\qquad(15b)$$

Then, eq 15a and 15b can be used immediately with the experimental quantity indicated in the differential (M_w or M_n, respectively) as supplied by the experimental method used.

Using eq 15a and 15b it is possible to derive explicit equations for the equilibrium constants, K_ξ, with $\xi = 1, 2, \ldots, m$ for any ideal self-associating system.

Derivation of K_1. The only case is $\xi = 1$, $r = 1$. Condition 1, $\alpha_2 + 2\alpha_3 + 3\alpha_4 + \ldots = \xi - r = 0$, is satisfied only for $\alpha_i = 0$, all i. Conditions 2 and 3 are only satisfied for these values of α_i, with $\alpha_1 = 1$. Substituting these results into eq 15a we have

$$\left(\frac{M_1}{M_w}\right)_{c=0}\frac{1}{1!0!0!\ldots0!}K_1K_2^0K_3^0\ldots = \left(\frac{M_1}{M_w}\right)_{c=0} = \frac{K_1}{1}\qquad(16)$$

Since by definition $K_1 = 1$, it follows that at $c = 0$, $M_1 = M_w$.

Derivation of K_2. *Summation terms in eq 15a.* First term, for $\xi = 2$, $r = 1$. Condition 1, $\alpha_2 + 2\alpha_3 + 3\alpha_4 + \ldots = \xi - r = 1$, is satisfied only for $\alpha_2 = 1$, $\alpha_i = 0$, and $i > 2$. This result satisfies also conditions 2 and 3, with $\alpha_1 = 0$, only. Using these results and eq 16 the first summation term in eq 15a becomes

$$\left(\frac{M_1}{M_w}\right)_{c=0}\frac{1}{0!1!0!\ldots0!}K_2^1K_3^0K_4^0\ldots = K_2\qquad(17c)$$

Second summation term (for $\xi = 2$, $r = 2$). Condition 1, $\alpha_2+2\alpha_3+3\alpha_4+\ldots = \xi - r = 0$, is satisfied only if $\alpha_i = 0$, all i; this result satisfies conditions 2 and 3 as well, for $\alpha_1 = 2$, only. With these results we can write a second summation term, to complete eq 15a, as follows

$$\left(\frac{\mathrm{d}\left(\frac{M_1}{M_w}\right)}{\mathrm{d}c}\right)_{c=0}\frac{1}{2!0!\ldots0!}K_2^0K_3^0\ldots = \frac{1}{2}\left(\frac{\mathrm{d}\left(\frac{M_1}{M_w}\right)}{\mathrm{d}c}\right)_{c=0}\qquad(17b)$$

Adding together the two summation terms obtained for $\xi = 2$, eq 15a can be written as

$$K_2 + \frac{1}{2}\left(\frac{\mathrm{d}\left(\frac{M_1}{M_w}\right)}{\mathrm{d}c}\right)_{c=0} = \frac{K_2}{2}\qquad(17a)$$

and solving for K, we have

$$K_2 = -\left(\frac{\mathrm{d}\left(\frac{M_1}{M_w}\right)}{\mathrm{d}c}\right)_{c=0}\qquad(17)$$

Derivation of K_3. *Summation terms for* $\xi = 3$. The first term is for $r = 1$. Condition 1, $\alpha_2 + 2\alpha_3 + 3\alpha_4 + \ldots = \xi - r = 2$, is satisfied if (a) $\alpha_3 = 1$, $\alpha_i = 0$, $i \neq 3$, or (b) $\alpha_2 = 2$, $\alpha_i = 0$, $i > 2$. However, conditions 2 and 3 are obeyed only in case a, with $\alpha_1 = 0$, thus excluding case b. Using these results and eq 16 the first summation term of eq 15a for $\epsilon = 3$ becomes

$$\left(\frac{M_1}{M_w}\right)_{c=0}\frac{1}{0!0!1!0!\ldots0!}K_2^0K_3^1K_4^0K_5^0\ldots = K_3\qquad(18d)$$

Second summation term (for $\xi = 3$, $r = 2$*).* Condition 1, $\alpha_2 + 2\alpha_3 + 3\alpha_4 + \ldots = 1$, is satisfied only for $\alpha_2 = 1$, $\alpha_i = 0$, $i > 2$. Conditions 2 and 3 are obeyed for this result, with $\alpha_1 = 1$, only. Then, the second summation term for $\xi = 3$ is

$$\left(\frac{\mathrm{d}\left(\frac{M_1}{M_w}\right)}{\mathrm{d}c}\right)_{c=0}\frac{1}{1!1!0!0!\ldots0!}K_2^1K_3^0K_4^0\ldots = \left(\frac{\mathrm{d}\left(\frac{M_1}{M_w}\right)}{\mathrm{d}c}\right)_{c=0}K_2\qquad(18c)$$

Third summation term (for $\xi = 3$, $r = 3$. Condition 1 is obeyed for $\alpha_i = 0$, all i, only. In this case, conditions 2 and 3 are satisfied as well, with $\alpha_1 = 3$, only. Using these results, the third summation term for $\xi = 3$ becomes

$$\left(\frac{\mathrm{d}^{(2)}\left(\frac{M_1}{M_w}\right)}{\mathrm{d}c^2}\right)_{c=0}\frac{1}{3!0!0!\ldots0!}K_2^0K_3^0K_4^0\ldots = \frac{1}{6}\left(\frac{\mathrm{d}^{(2)}\left(\frac{M_1}{M_w}\right)}{\mathrm{d}c^2}\right)_{c=0}\qquad(18b)$$

Adding together all summation terms obtained for $\xi = 3$, eq 15a is now complete and can be written as

$$\frac{K_3}{3} = K_3 + \left(\frac{\mathrm{d}\left(\frac{M_1}{M_w}\right)}{\mathrm{d}c}\right)_{c=0}K_2 + \frac{1}{6}\left(\frac{\mathrm{d}^{(2)}\left(\frac{M_1}{M_w}\right)}{\mathrm{d}c^2}\right)_{c=0}\qquad(18a)$$

and using eq 17 and solving for K_3, we have

$$K_3 = \frac{3}{2}\left(\frac{\mathrm{d}\left(\dfrac{M_1}{M_w}\right)}{\mathrm{d}c}\right)^2_{c=0} - \frac{1}{4}\left(\frac{\mathrm{d}^{(2)}\left(\dfrac{M_1}{M_w}\right)}{\mathrm{d}c^2}\right)_{c=0} \quad (18)$$

Derivation of K_4. First summation term (for $\xi = 4$, $r = 1$). Condition 1 is satisfied in three cases: (a) $\alpha_2 = 1, \alpha_3 = 1, \alpha_i = 0, i > 3$; (b) $\alpha_2 = 3, \alpha_i = 0, i > 2$; and (c) $\alpha_4 = 1, \alpha_i = 0, i \neq 4$.

However, conditions 2 and 3 are obeyed in case c only, with $\alpha_1 = 0$, thus excluding cases a and b. With these results the first summation term in eq 15a becomes

$$\left(\frac{M_1}{M_w}\right)_{c=0}\frac{1}{0!0!0!1!0!\ldots0!}K_2^0K_3^0K_4^1K_5^0 \ldots = K_4 \quad (19f)$$

Summation terms for $\xi = 4$, $r = 2$. In this case, conditions 1, 2, and 3 are satisfied in two cases: (a) $\alpha_3 = 1$, $\alpha_i = 0, i \neq 3$; and (b) $\alpha_2 = 2, \alpha_i = 0, i > 2$, with $\alpha_1 = 1$ and $\alpha_1 = 0$, respectively. Thus, two summation terms are generated for $\xi = 4$, $r = 2$, as

$$\text{(a)}\quad \left(\frac{\mathrm{d}\left(\dfrac{M_1}{M_w}\right)}{\mathrm{d}c}\right)_{c=0}\frac{1}{1!0!1!0!0!\ldots0!}K_2^0K_3^1K_4^0K_5^0 \ldots = \left(\frac{\mathrm{d}\left(\dfrac{M_1}{M_w}\right)}{\mathrm{d}c}\right)_{c=0}K_3 \quad (19e)$$

$$\text{(b)}\quad \left(\frac{\mathrm{d}\left(\dfrac{M_1}{M_w}\right)}{\mathrm{d}c}\right)_{c=0}\frac{1}{0!2!0!\ldots0!}K_2^2K_3^0K_4^0 \ldots = \frac{1}{2}\left(\frac{\mathrm{d}\left(\dfrac{M_1}{M_w}\right)}{\mathrm{d}c}\right)_{c=0}K_2^2 \quad (19d)$$

Summation term for $\xi = 4$, $r = 3$. Conditions 1, 2, and 3 are satisfied for $\alpha_1 = 2$, $\alpha_2 = 1$, $\alpha_i = 0$, $i > 2$, only. Then another summation term can be written as

$$\left(\frac{\mathrm{d}^{(2)}\left(\dfrac{M_1}{M_w}\right)}{\mathrm{d}c^2}\right)_{c=0}\frac{1}{2!1!0!0!\ldots0!}K_2^1K_3^0K_4^0 \ldots = {}^1\!/_2K_2\left(\frac{\mathrm{d}^{(2)}\left(\dfrac{M_1}{M_w}\right)}{\mathrm{d}c^2}\right)_{c=0} \quad (19c)$$

Summation term for $\xi = 4$, $r = 4$. In this case, conditions 1, 2, and 3 are satisfied only for $\alpha_1 = 4$, $\alpha_i = 0$, $i \neq 1$. Thus, a final summation term is generated for $\xi = 4$

$$\left(\frac{\mathrm{d}^{(3)}\left(\dfrac{M_1}{M_w}\right)}{\mathrm{d}c^3}\right)_{c=0}\frac{1}{4!0!0!\ldots0!}k_2^0k_3^0k_4^0 \ldots = \frac{1}{24}\left(\frac{\mathrm{d}^{(3)}\left(\dfrac{M_1}{M_w}\right)}{\mathrm{d}c^3}\right)_{c=0} \quad (19b)$$

Adding together all summation terms obtained for $\xi = 4$, eq 15a can be written

$$\frac{K_4}{4} = K_4 + \left(\frac{\mathrm{d}\left(\dfrac{M_1}{M_w}\right)}{\mathrm{d}c}\right)_{c=0}(K_3 + {}^1\!/_2K_2^2) + {}^1\!/_2K_2\left(\frac{\mathrm{d}^{(2)}\left(\dfrac{M_1}{M_w}\right)}{\mathrm{d}c^2}\right)_{c=0} + \frac{1}{24}\left(\frac{\mathrm{d}^{(3)}\left(\dfrac{M_1}{M_w}\right)}{\mathrm{d}c^3}\right)_{c=0} \quad (19a)$$

Solving for K_4, and using eq 17 and 18, eq 19a becomes

$$K_4 = -\frac{8}{3}\left(\frac{\mathrm{d}\left(\dfrac{M_1}{M_w}\right)}{\mathrm{d}c}\right)^3_{c=0} + \left(\frac{\mathrm{d}\left(\dfrac{M_1}{M_w}\right)}{\mathrm{d}c}\right)_{c=0}\left(\frac{\mathrm{d}^{(2)}\left(\dfrac{M_1}{M_w}\right)}{\mathrm{d}c^2}\right)_{c=0} - \frac{1}{18}\left(\frac{\mathrm{d}^{(3)}\left(\dfrac{M_1}{M_w}\right)}{\mathrm{d}c^3}\right)_{c=0} \quad (19)$$

Derivation of Equations to Determine Equilibrium Constants from Eq 15b. The approach and derivations necessary to use eq 15b are essentially the same as with eq 15a. The only difference is the experimental information supplied, M_{nc}, obtained from osmotic pressure measurements or calculated by any other means, *e.g.*, by means of eq 10. Therefore, it would be redundant to repeat here the derivations used in the preceding sections. Only final equations are, therefore, presented, as

$$K_1 = \left(\frac{M_1}{M_n}\right)_{c=0} \quad (20)$$

$$K_2 = -2\left(\frac{\mathrm{d}\left(\dfrac{M_1}{M_n}\right)}{\mathrm{d}c}\right)_{c=0} \quad (21)$$

$$K_3 = 6\left(\frac{\mathrm{d}\left(\dfrac{M_1}{M_n}\right)}{\mathrm{d}c}\right)^2_{c=0} - \frac{3}{4}\left(\frac{\mathrm{d}^{(2)}\left(\dfrac{M_1}{M_n}\right)}{\mathrm{d}c^2}\right)_{c=0} \quad (22)$$

$$K_4 = -\frac{64}{3}\left(\frac{\mathrm{d}\left(\dfrac{M_1}{M_n}\right)}{\mathrm{d}c}\right)^3_{c=0} + 6\left(\frac{\mathrm{d}\left(\dfrac{M_1}{M_n}\right)}{\mathrm{d}c}\right)_{c=0}\left(\frac{\mathrm{d}^{(2)}\left(\dfrac{M_1}{M_n}\right)}{\mathrm{d}c^2}\right)_{c=0} - \frac{2}{9}\left(\frac{\mathrm{d}^{(3)}\left(\dfrac{M_1}{M_n}\right)}{\mathrm{d}c^3}\right)_{c=0} \quad (23)$$

Discussion

Steiner (1952, 1954) derived equations for the analysis of ideal associating systems. His derivations make use of the number fraction weight of monomer and the definitions of either M_n or M_w, which are not presented as functionally related. Thus, two independent methods were presented for computing the equilibrium constants from osmotic pressure and light-scattering experiments, respectively. Adams (1965a,b) and Adams and Lewis (1968) derived expressions for the analysis of nonideal systems. They also used the number fraction weight of monomer (Steiner, 1952, 1954). In addition, Adams (1965a,b) established a functional relation between M_n and M_w. This relation has been used in this work as $F(c)$ (eq 10). However, this work differs from those mentioned above in several respects: (1) the use of a power expansion to express $F(c)$, (2) the replacement of c in this power expansion by its value in terms of the equilibrium constants, and (3) the use of the multinomial theorem. The present method requires nothing more than M_{nc} or M_{wc}, for which the same general equation is applicable. Furthermore, this general expression is valid irrespective of whether these are either the weight average or the number average. Also, this equation is valid for self-associating reactions of any degree of polymerization that behave ideally. The evaluation of the derivative of M_1/M_w (or M_1/M_n) in the vicinity of $c = 0$ for the associating solute may generate some uncertainty in the values of the equilibrium constants. This uncertainty principally arises from the steepness frequently encountered in the limiting slopes of the plots of M_1/M_w (or M_1/M_n) *vs.* c (*cf.* Adams and Filmer, 1966; Adams and Lewis, 1968). This uncertainty should be reduced as the sensitivity and precision in the determination of M_{wc} (or M_{nc}) is increased particularly in the region near $c = 0$. Some recent advances in technique are promising here. One of these is the introduction of the photoelectric scanner in the absorption optical system (Hanlon *et al.*, 1962; Schachman *et al.*, 1962) for ultracentrifuge work at very low solute concentrations. Also for use with interference optics, a method[1] that uses a mesh of fine wires interposed before the photographic plate to follow fringe displacement offers increased sensitivity and precision in sedimentation equilibrium measurements. Therefore, firstly, the availability of more precise experimental data, and secondly, the fact that the intercept of the plot of M_1/M_{wc} or (M_1/M_{nc}) with the ordinate at $c = 0$ is always precisely known ($M_1/M_{wc} = M_1/M_{nc} = 1$), together with, thirdly, the application of numerical analysis to obtain the curve of best fit in this region should provide more precise expressions of its derivatives. Work is in progress for the application of the multinomial theory to nonideal systems.

[1] M. Derechin (1968), manuscript submitted for publication.

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