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A general approach to the interpretation of pH buffer catalyzed degradation profiles

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Abstract

A general equation for the pH-dependent general acid-base catalyzed degradation profiles of polybasic weak electrolytes is presented. The potential information which can be deduced from such profiles is discussed. The method is illustrated with a hypothetical example.

Key words: Degradation kinetics; Aqueous solution; Acid-base catalysis; Buffer effect; Catalysis; Weak electrolyte; Stability profile

1. Introduction

The combination of proton-catalyzed, solvent-catalyzed and hydroxyl-catalyzed degradation reactions of a substance, involved in n protolytic equilibria, results in complex pH-degradation profiles which can be described by means of $n+3$ macroreaction constants and n protolytic dissociation constants (Van der Houwen et al., 1988). If catalysis by buffer components or other weak electrolytes present in solution (which themselves are involved in protolytic equilibria) occurs, the pH-degradation profile becomes even more complicated. Such specific catalysis by buffer components or other weak electrolytes is important in the field of pharmacy as unstable pharmacologically active substances often have to be formulated as buffered solutions, containing several components. For the interpretation of the observed degradation, the profiles must be described in model equations appropriate for curve fitting. The curve fitting results in the calculation of a number of constants. The calculated constants are related to the rate constants of all individual reactions and to the protolytic dissociation constants. These relations can be described by a number of mathematical equations.

2. General mathematical approach

The weak electrolyte AH_n exists in solution in different species depending on the degree of de-

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protonation (and therefore on the pH). The stepwise dissociation of the substance is described by a series of equations with the general form of Eq. 1:

$$K_i = [\mathrm{H}^+] \cdot [\mathrm{AH}^{i-}_{n-i}] / [\mathrm{AH}^{1-i}_{n-i+1}] \tag{1}$$

assuming that AH_n itself is uncharged. K_i indicates the i-th protolytic dissociation constant of the solute.

The concentration of each species can be expressed as a function of the concentration of the totally protonated species by applying Eq. 1 to the successive deprotonation steps.

When the species are numbered from zero (AH_n) to n (A^{n-}), then the concentration of the i-th species is given by Eq. 2:

$$[\mathrm{AH}^{i-}_{n-\mathrm{i}}] = [\mathrm{AH}_n] \cdot \Pi^{i}_{h=0} K_h / [\mathrm{H}^+]^i \tag{2}$$

The fraction of the substance present as the i-th species is expressed by Eq. 3:

$$\text{fraction } i\text{-th species} = \frac{\Pi^{i}_{h=0} K_h / [\mathrm{H}^+]^i}{\sum\limits_{j=0}^{n} \left\{ \prod\limits_{h=0}^{j} K_h / [\mathrm{H}^+]^j \right\}} \tag{3}$$

To simplify the equations a non-existent constant K_0 is defined equal to 1.

In an analogous manner, the concentration of the i-th species of a buffering agent BH_m is given by Eq. 4:

$$\text{fraction } i\text{-th species} = \frac{\Pi^{i}_{h=0} K^{\mathrm{b}}_h / [\mathrm{H}^+]^i}{\sum\limits_{j=0}^{n} \left\{ \prod\limits_{h=0}^{j} K^{\mathrm{b}}_h / [\mathrm{H}^+]^j \right\}} \tag{4}$$

The i-th protolytic dissociation constant of the buffering agent is indicated by K^{b}_i.

Each of the $n + 1$ species of the substance can be involved in $m + 1$ degradation reactions, each catalyzed by one of the species of the buffering agent. The contribution of the reaction of the i-th species of the substance catalyzed by the j-th species of the buffering agent to the observed overall degradation rate constant k_{obs} is given by Eq. 5:

$$\frac{\Pi^{i}_{h=0} K_h / [\mathrm{H}^+]^i}{\sum\limits_{l=0}^{n} \{ \Pi^{l}_{h=0} K_h / [\mathrm{H}^+]^l \}} \cdot \frac{\Pi^{j}_{h=0} K^{\mathrm{b}}_h / [\mathrm{H}^+]^j}{\sum\limits_{l=0}^{m} \{ \Pi^{l}_{h=0} K^{\mathrm{b}}_h / [\mathrm{H}^+]^l \}} \cdot C_{\mathrm{buf}} \cdot k_{i,j} \tag{5}$$

in which $k_{\mathrm{i,j}}$ is the rate constant of the individual reaction involved and C_{buf} denotes the total concentration of the buffering agent.

Combining the contributions of the possible reactions, Eq. 6 is obtained:

$$k_{\mathrm{obs}} = \frac{\sum\limits_{i=0}^{n} \left| \Pi^{i}_{h=0} K_h / [\mathrm{H}^+]^i \cdot \sum\limits_{j=0}^{m} \{ \Pi^{j}_{h=0} K^{\mathrm{b}}_h / [\mathrm{H}^+]^j \cdot k_{i,j} \} \right|}{\sum\limits_{i=0}^{n} \{ \Pi^{i}_{h=0} K_h / [\mathrm{H}^+]^i \} \cdot \sum\limits_{j=0}^{m} \{ \Pi^{j}_{h=0} K^{\mathrm{b}}_h / [\mathrm{H}^+]^j \}} \cdot C_{\mathrm{buf}} \tag{6}$$

If the numerator and the denominator are fully expanded and the resulting terms of the numerator and the denominator, containing the same power of $[\mathrm{H}^+]$, are combined, Eq. 6 can be transformed to the model Eq. 7. By doing so, the reaction constants of all individual degradation reactions, that are kinetically indistinguishable, are combined into one single 'macroreaction constant' M_i:

$$k_{\mathrm{obs}} = \frac{\sum\limits_{i=0}^{n+m} M_i / [\mathrm{H}^+]^i}{\sum\limits_{i=0}^{n+m} K^{\mathrm{c}}_i / [\mathrm{H}^+]^i} \cdot C_{\mathrm{buf}} \tag{7}$$

where M_i is defined by Eq. 8:

$$M_i = \sum_{j=0}^{i} \{ \Pi^{j}_{h=0} K_h \cdot \Pi^{i-j}_{h=0} K^{\mathrm{b}}_h \cdot k_{j,i-j} \} \tag{8}$$

and the combined protolytic dissociation constant K^{c}_i by Eq. 9:

$$K^{\mathrm{c}}_i = \sum_{j=0}^{i} \{ \Pi^{j}_{h=0} K_h \cdot \Pi^{i-j}_{h=0} K^{\mathrm{b}}_h \} \tag{9}$$

Terms containing subscripts outside the range $0 - n$ for the substance and $0 - m$ for the buffer in Eq. 8 and 9 refer to non-existing species. These terms are therefore to be neglected. The treatment of the hypothetical example illustrates how this is done (Eq. 11–22). Eq. 8 indicates that it is impossible to distinguish between the reactions corresponding with combinations of species of the substance and the buffer for which the sum of the charges of both species is equal.

The denominator of Eq. 7 indicates that the plot of the logarithms of the contribution of the macroreaction constants to the observed rate constant as a function of the pH can be approached by a combination of $m+n+1$ straight lines. The inflection points of the graph occur at pH values for which Eq. 10 is valid:

$$[\mathrm{H}^+] = K_i^c / K_{i-1}^c \tag{10}$$

The plots of the logarithms of the contribution of the individual reactions (Eq. 5) to the observed rate constants vs pH have analogous shapes, since the denominators of Eq. 5 and 7 are equal.

When the protolytic dissociation constants of the substance and the buffer all differ sufficiently in their order of magnitude, these inflection points occur at the pH values that correspond with the $\mathrm{p}K_a$ values of the substance and the buffer. This can be proven by expanding the constants K^c according to Eq. 9 and neglecting the parts that do not contribute significantly (according to their order of magnitude). If the actual values of the protolytic dissociation constants are known, the inflection points can be calculated with Eq. 10.

The results of such calculations are given for the hypothetical example.

Table 1
(A) Logarithms of the micro reaction constants, (B) logarithms of the macro reaction constants and (C) values of the combined protolytic dissociation constants

(A) Solute species	Buffer species			
	BH_3	BH_2^-	BH^{2-}	B^{3-}
AH^2	−1.0 (1) [a]	−3.0 (2) [a]	−2.0 (4) [a]	−1.0 (9) [a]
AH^-	−2.0 (3)	−7.1 (5)	−6.0 (7)	−4.0 (11)
A^{2-}	−0.0 (6)	−5.0 (8)	−5.1 (10)	−3.0 (12)

(B) Logarithms of the macro reaction constants M_i

M_i	Value
M_0	−1.00
M_1	−4.00
M_2	−10.00
M_3	−18.00
M_4	−27.09
M_5	−38.00

(C) Values of the combined protolytic dissociation K_i^c

K_i^c	Value
K_0^c	1
K_1^c	10^{-1}
K_2^c	10^{-5}
K_3^c	10^{-12}
K_4^c	10^{-22}
K_5^c	10^{-35}

[a] Numbers in parentheses refer to the curves in Fig. 3.

3. A hypothetical example

The generalized mathematical treatment is illustrated with the hypothetical example of substance AH_2 with $\mathrm{p}K_a$ values of 4 and 10 and a buffering agent BH_3 with $\mathrm{p}K_a$ values of 1, 7 and 13, respectively. The values of the rate constants of the individual degradation reactions are given in Table 1. Fig. 1 shows the fraction of the substance AH_2 present in its respective species as a function of the pH. Fig. 2 gives analogous plots for the buffer. The pH corresponding with the respective $\mathrm{p}K_a$ values is indicated with vertical lines.

In Fig. 3, the contribution of the individual rate constants to the observed degradation constant k_{obs} is plotted as a function of pH. From these plots it is obvious that these reactions can be divided into six groups, containing, respectively, one, two, three, three, two, and one micro reactions. The plots of the reactions within a group are parallel. It is therefore impossible to decide from kinetic measurements to which extent the individual reactions within a group contribute to k_{obs}. This means that these reactions are kinetically indistinguishable. For the interpretation of degradation measurements, the kinetically indistinguishable reactions must be combined into a single macro reaction constant.

Applying Eq. 8 these macro reaction constants M_0–M_5 can be written as Eq. 11–16:

$$M_0 = \Pi_{i=0}^{0} K_i \cdot \Pi_{j=0}^{0} K_j^b \cdot k_{0,0} = k_{0,0} \tag{11}$$

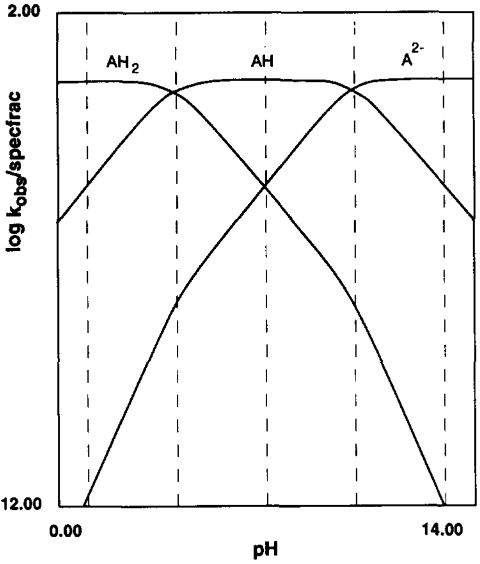

Fig. 1. Logarithms of the fraction of each of the species resulting from AH_2, present in the solution as a function of the pH.

$$\begin{aligned} M_1 &= \Pi^0_{i=0} K_i \cdot \Pi^1_{j=0} K^b_j \cdot k_{0,1} \\ &\quad + \Pi^1_{i=0} K_i \cdot \Pi^0_{j=0} K^b_j \cdot k_{1,0} \\ &= K_0 \cdot k_{0,1} + K^b_0 \cdot k_{1,0} \end{aligned} \tag{12}$$

$$\begin{aligned} M_2 &= \Pi^0_{i=0} K_i \cdot \Pi^2_{j=0} K^b_j \cdot k_{0,2} \\ &\quad + \Pi^1_{i=0} K_i \cdot \Pi^1_{j=0} K^b_j \cdot k_{1,1} \\ &\quad + \Pi^2_{i=0} K_i \cdot \Pi^0_{j=0} K^b_j \cdot k_{2,0} \\ &= K^b_1 \cdot K^b_2 \cdot k_{0,2} + K_1 \cdot K^b_1 \cdot k_{1,1} + K_1 \cdot K_2 \cdot k_{2,0} \end{aligned} \tag{13}$$

$$\begin{aligned} M_3 &= \Pi^0_{i=0} K_i \cdot \Pi^3_{j=0} K^b_j \cdot k_{0,3} \\ &\quad + \Pi^1_{i=0} K_i \cdot \Pi^2_{j=0} K^b_j \cdot k_{1,2} \\ &\quad + \Pi^2_{i=0} K_i \cdot \Pi^1_{j=0} K^b_j \cdot k_{2,1} \\ &= K^b_1 \cdot K^b_2 \cdot K^b_3 \cdot k_{0,3} + K_1 \cdot K^b_1 \cdot K^b_2 \cdot k_{1,2} \\ &\quad + K_1 \cdot K_2 \cdot K^b_1 \cdot k_{2,1} \end{aligned} \tag{14}$$

$$\begin{aligned} M_4 &= \Pi^1_{i=0} K_i \cdot \Pi^3_{j=0} K^b_j \cdot k_{1,3} \\ &\quad + \Pi^2_{i=0} K_i \cdot \Pi^2_{j=0} K^b_j \cdot k_{2,2} \\ &= K_1 \cdot K^b_1 \cdot K^b_2 \cdot K^b_3 \cdot k_{1,3} \\ &\quad + K_1 \cdot K_2 \cdot K^b_1 \cdot K^b_2 \cdot k_{2,2} \end{aligned} \tag{15}$$

$$\begin{aligned} M_5 &= \Pi^2_{i=0} K_i \cdot \Pi^3_{j=0} K^b_j \cdot k_{2,3} \\ &= K_1 \cdot K_2 \cdot K^b_1 \cdot K^b_2 \cdot K^b_3 \cdot k_{2,3} \end{aligned} \tag{16}$$

The macro reaction constants are calculated by substituting the protolytic dissociation constants and the individual reaction constants in Eq. 11–16. The results are given in Table 1.

The constants K^c_0–K^c_5 are given by Eq. 17–22.

$$K^c_0 = \Pi^0_{i=0} K_i \cdot \Pi^0_{j=0} K^b_j = 1 \tag{17}$$

$$\begin{aligned} K^c_1 &= \Pi^0_{i=0} K_i \cdot \Pi^1_{j=0} K^b_j + \Pi^1_{i=0} K_i \cdot \Pi^0_{j=0} K^b_j \\ &= K_0 + K^b_0 \end{aligned} \tag{18}$$

$$\begin{aligned} K^c_2 &= \Pi^0_{i=0} K_i \cdot \Pi^2_{j=0} K^b_j + \Pi^1_{i=0} K_i \cdot \Pi^1_{j=0} K^b_j \\ &\quad + \Pi^2_{i=0} K_i \cdot \Pi^0_{j=0} K^b_j \\ &= K^b_1 \cdot K^b_2 + K_1 \cdot K^b_1 + K_1 \cdot K_2 \end{aligned} \tag{19}$$

$$\begin{aligned} K^c_3 &= \Pi^0_{i=0} K_i \cdot \Pi^3_{i=0} K^b_j + \Pi^1_{i=0} K_i \cdot \Pi^2_{j=0} K^b_j \\ &\quad + \Pi^2_{i=0} K_i \cdot \Pi^1_{j=0} K^b_j \\ &= K^b_1 \cdot K^b_2 \cdot K^b_3 + K_1 \cdot K^b_1 \cdot K^b_2 + K_1 \cdot K_2 \cdot K^b_1 \end{aligned} \tag{20}$$

$$\begin{aligned} K^c_4 &= \Pi^1_{i=0} K_i \cdot \Pi^3_{j=0} K^b_j + \Pi^2_{i=0} K_i \cdot \Pi^2_{j=0} K^b_j \\ &= K_1 \cdot K^b_1 \cdot K^b_2 \cdot K^b_3 + K_1 \cdot K_2 \cdot K^b_1 \cdot K^b_2 \end{aligned} \tag{21}$$

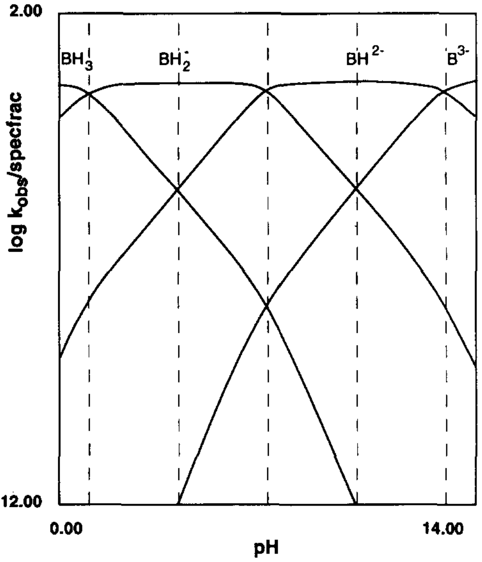

Fig. 2. Logarithms of the fraction of each of the species resulting from BH_3, present in the solution as a function of the pH.

$$K_5^c = \Pi_{i=0}^{2} K_i \cdot \Pi_{j=0}^{3} K_j^b = K_1 \cdot K_2 \cdot K_1^b \cdot K_2^b \cdot K_3^b \qquad (22)$$

The values for K_i^c are calculated by substitution of the protolytic dissociation constants in Eq. 17–22. The results are given in Table 1.

The total observed degradation profile, and the contribution of each of the macroreaction constants to it, is given in Fig. 4 as a function of the pH.

When the calculated constants K_i^c are substituted in Eq. 10, the pH values of 1.0, 4.0, 7.0, 10.0 and 13.0 are obtained for the inflection points in the graphs in Fig. 3 and 4. These values correspond with the pK_a values of the solute and the buffer.

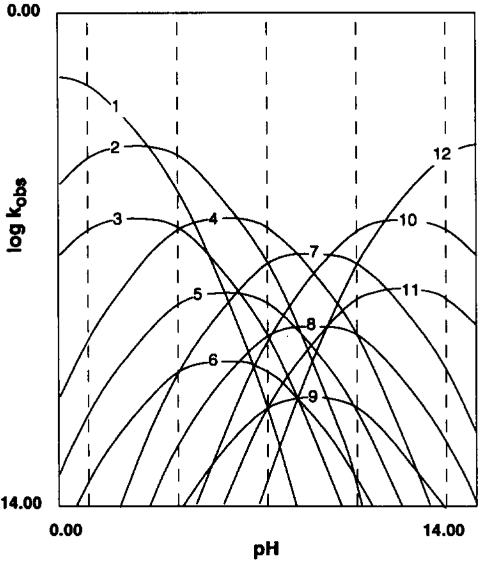

Fig. 3. Logarithm of the contribution of all reactions of the different species resulting from AH$_2$ to k_{obs} as a function of pH. The numbers of the graphs correspond with the numbers within parentheses in Table 1.

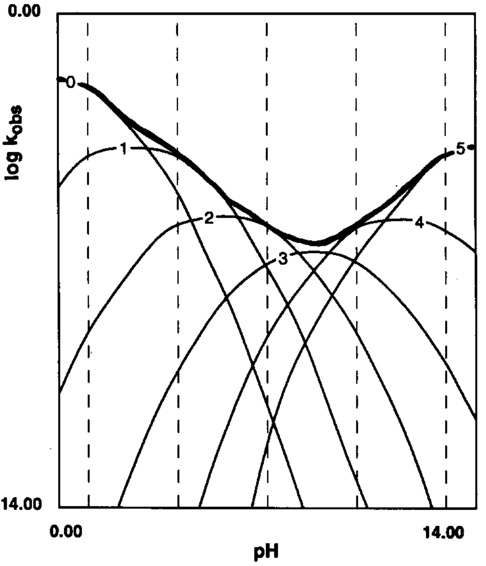

Fig. 4. Logarithm of the contribution of the macro reaction constants (M_0–M_5) to k_{obs} as a function of pH together with the overall degradation profile of AH$_2$.

4. Application of the model equation for the interpretation of degradation profiles

When the degradation of solutes in buffered media is studied, a combination of proton-, hydroxyl-, solvent- and buffer-catalyzed reactions should be taken into account. The contributions of the buffer-catalyzed reactions are proportional to the buffer concentration (Eq. 7). Those of the proton-, hydroxyl- and solvent-catalyzed reactions are independent of the buffer concentration. The contribution of the buffer at a given pH can therefore be calculated from a series of measurements at constant pH and varying buffer concentrations (Eq. 23):

$$k_{obs} = k_{obs}^0 + k_{buf} \cdot [buf] \qquad (23)$$

According to this equation the observed rate constant, k_{obs}, plotted against the buffer concentration [buf] yields a straight line with a slope equal to the contributions of the buffer-catalyzed reactions (k_{buf}). The intercept corresponds with the contributions of the proton-, hydroxyl- and solvent-catalyzed reactions (k_{obs}^0).

The calculation of the contributions of buffer-catalyzed reactions is possible if these contribute significantly to the overall rate constant. This is more likely to occur for intermediate pH values where the proton and hydroxyl concentrations are low compared to those of the species of the buffering agent.

Apart from 'specific' buffer ion catalytic influences on k_{obs}, a general influence of ionic strength on degradation reaction rates can be expected in cases where two charged species are involved in the reaction (Martin et al., 1983). To eliminate this influence, measurements of the degradation rate constants must be carried out at constant ionic strength. A decrease in buffer concentration should be compensated for by a corresponding increase in the concentration of inert salt.

If such measurements are repeated at a series of pH values, a series of measured values of k_{buf} are obtained. The value of the constants M_i can then be calculated from the values of k_{buf} using Eq. 7 as a model for computer fitting.

If the protolytic dissociation constants of the solute and the buffer are known, the value of constant K_i^c in the numerator of Eq. 7 can be calculated. This results in less unknown parameters and therefore more reliable results from the computer fitting.

5. Conclusions

The pH profile of the degradation of a substance AH_n catalyzed by a buffering agent BH_m can be described with a general equation containing $m + n + 1$ macro reaction constants and $m + n$ combinations of the protolytic dissociation constants.

References

Martin, A., Swarbrick, J. and Cammarata, A., *Physical Pharmacy*, 3rd Edn, Lea & Febiger, Philadelphia, 1983, p. 367.

Van der Houwen, O.A.G.J., Beijnen, J.H., Bult, A. and Underberg, W.J.M., A general approach to the interpretation of pH degradation profiles. *Int. J. Pharm*, 45 (1988) 181–188.