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# 'STEADY/EQUILIBRIUM APPROXIMATION' IN RELAXATION AND FLUCTUATION

## II. MATHEMATICAL THEORY OF APPROXIMATIONS IN FIRST-ORDER REACTION

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A mathematical theory of the steady/equilibrium approximation for first-order reactions is presented. This gives the theoretical basis for the methods of simplifying the complex first-order reactions described in the preceding work. The steady/equilibrium relation holds on every fast component after a proper inducation period $T^0$. $T^0$ is either of O(1) or less, or nearly of O(1/ε) depending on the reaction scheme and on the initial condition but is always less than O(1/ε) (as in the preceding paper [1], we use the symbol O(1) to denote a positive number of the order of unity). In the open group, the determinant of the submatrix $M_p$, representing the interconversion between the fast components in the group and their dissipation, is of O(1). The concentration of the fast components in the open group can thus be expressed as a linear combination of those components neighboring the group after the establishment of a steady/equilibrium relation, and can be eliminated from the reaction scheme leaving the pathway through them. On the other hand, in the closed group the determinant of $M_p$ is of O(ε) or less and the components in the group are in quasi equilibrium with each other after $T^0$. They are eliminated from the reaction scheme leaving the sum of the components in the closed group as a slow component.

## 1. Introduction

In the preceding paper [1], we presented methods of simplifying complex first-order reactions by the two approximations: the steady-state approximation and the principle of fast equilibration (both were collectively denoted by the 'steady/equilibrium approximation'), without detailed mathematical proof. Here we present the mathematical theory of the steady/equilibrium approximation in a first-order reaction (or Markov process with discrete states). This clarifies under which conditions the two approximations hold and gives the foundation of the methods for the application of the two approximations.

First, we describe the formal solutions of the rate equation and roughly explain why every fast component can be put equal to zero to a good approximation, but detailed examination of the conditions under which the steady/equilibrium relation (cf. eq. 4 in ref. 1) holds is presented in appendix A and B.

Next, we deal with case where only open groups are included in the reaaction scheme and finally discuss the case where both open and closed groups are included.

## 2. Results

We treat in this work a general first-order reaction scheme consisting of $n$ components. The fundamental assumptions, definitions and notations concerning this reaction scheme are the same as those in the previous work [1].

We neglect throughout this work accidental cancellations or reduction of order of magnitude caused by assigning special values to the rate constants.

The rate equations of a first-order reaction are given in matrix form as

$$\frac{\mathrm{d}}{\mathrm{d}t}\boldsymbol{x}=\boldsymbol{Q}\boldsymbol{x} \tag{1}$$

Column vector $\boldsymbol{x}$ in eq. 1 is expressed as

$$\boldsymbol{x}=\begin{pmatrix} x_1\\ \vdots\\ x_n\end{pmatrix}=\begin{pmatrix} x_1\\ \vdots\\ x_r\\ y_1\\ \vdots\\ y_m\end{pmatrix}=\begin{pmatrix} \boldsymbol{x}_r\\ \boldsymbol{y}_m\end{pmatrix}, \tag{2}$$

and $n\times n$ matrix $\boldsymbol{Q}$ is given as

$$\boldsymbol{Q}=\left(\begin{array}{lll|ll} k_{11}, & k_{12},\dots, & k_{1r} & k_{1,r+1},\dots, & k_{1n}\\ \vdots & & & \vdots & \\ k_{r1}, & k_{r2},\dots, & k_{rr} & k_{r,r+1},\dots, & k_{rn}\\ \hline k_{r+1,1}, & k_{r+1,2},\dots, & k_{r+1,r} & k_{r+1,r+1},\dots, & k_{r+1,n}\\ \vdots & & & \vdots & \\ k_{n1}, & k_{n2},\dots, & k_{nr} & k_{n,r+1},\dots, & k_{nn}\end{array}\right)$$
$$=\left(\begin{array}{lll|ll} \epsilon k'_{11}, & \epsilon k'_{12},\dots, & \epsilon k'_{1r} & k_{1,r+1},\dots, & k_{1n}\\ \vdots & & & \vdots & \\ \epsilon k'_{r1}, & \epsilon k'_{r2},\dots, & \epsilon k'_{rr} & k_{r,r+1},\dots, & k_{rn}\\ \hline \epsilon k'_{r+1,1}, & \epsilon k'_{r+1,2},\dots, & \epsilon k'_{r+1,r} & k_{r+1,r+1},\dots, & k_{r+1,n}\\ \vdots & & & \vdots & \\ \epsilon k'_{n1}, & \epsilon k'_{n2},\dots, & \epsilon k'_{nr} & k_{n,r+1},\dots, & k_{nn}\end{array}\right) \tag{3}$$

In the second representation of $\boldsymbol{Q}$ in eq. 3 we put

$$k_{j,i}\equiv\epsilon k'_{j,i}\quad (j=1,\dots,n;\ i=1,\dots,r) \tag{4}$$

where $k_{j,r+p}$ and newly defined constant $k'_{j,i}$ ($i=1,\dots,r$; $j=1,\dots,n$; $p=1,\dots,m$) are all of the order of unity or less. The matrix $\boldsymbol{Q}$ is conveniently partitioned as shown by solid lines in eq. 4 and is represented by

$$\boldsymbol{Q}=\left(\begin{array}{c|c} \boldsymbol{Q}_{rr} & \boldsymbol{Q}_{rm}\\ \hline \boldsymbol{Q}_{mr} & \boldsymbol{Q}_{mm}\end{array}\right) \tag{5}$$

The exact solution of the rate equation, eq. 1, is given as [2,3],

$$\boldsymbol{x}(t)=\left(\sum_{h=1}^{n} e^{\lambda_h t}\boldsymbol{A}^{(h)}\right)\boldsymbol{x}(0)=\boldsymbol{x}(\infty)+\left(\sum_{h=2}^{n} e^{\lambda_h t}\boldsymbol{A}^{(h)}\right)\boldsymbol{x}(0) \tag{6}$$

where $\lambda_h$ ($h=1,\dots,n$) are the eigenvalues of $\boldsymbol{Q}$ obtained by solving the secular equation,

$$|\boldsymbol{Q}-\lambda\boldsymbol{E}|=0 \tag{7}$$

where $\boldsymbol{E}$ is a unit matrix, and $\lambda_1=0$ and $\lambda_h$ ($h=2,\dots,n$) are in general real and negative under the fundamental assumption that the principle of detailed balance holds on each reaction step. We assume here that the eigenvalues of $Q$ are not degenerate.

The projection matrix $A^{(h)}$ is given by the eigen-column vector $m^{(h)}$ and eigen-row-vector $n^{(h)}$ of $Q$ as

$$A^{(h)} = m^{(h)} n^{(h)} \tag{8}$$

where $m^{(h)}$ and $n^{(h)}$ satisfy

$$\left.\begin{aligned} n^{(h)} \cdot m^{(f)} &= \delta_{hf} \\ Q m^{(h)} &= \lambda_h m^{(h)} \\ n^{(h)} Q &= \lambda_h n^{(h)} \end{aligned}\right\} \tag{9}$$

From the above, the following relations hold on $A^{(h)}$ [2,3],

$$\left.\begin{aligned} A^{(h)} A^{(h)} &= A^{(h)} \\ A^{(h)} A^{(f)} &= 0 \;\; (h \neq f) \\ (Q - \lambda_h E) A^{(h)} &= A^{(h)} (Q - \lambda_h E) = 0 \end{aligned}\right\} \tag{10}$$

As shown later, among $n-1$ nonzero eigenvalues, $\lambda_h$ $(h = 2, \ldots, n)$, some are of O(1) (large eigenvalues) and others of O($\epsilon$) or less (small eigenvalues). We assume that there are $b$ small eigenvalues $(\lambda_2, \ldots, \lambda_b$, including $\lambda_1 = 0)$ and $n - b$ large eigenvalues $(\lambda_{b+1}, \ldots, \lambda_n)$. The number $b$ is determined later.

From eq. 6, we obtain,

$$\left.\begin{aligned} x(t) &= \sum_{h=1}^{b} e^{\lambda_h t} A^{(h)} x(0) + \sum_{f=b+1}^{n} e^{\lambda_f t} A^{(f)} x(0) \\ \frac{d}{dt} x(t) &= \sum_{h=2}^{b} \lambda_h e^{\lambda_h t} A^{(h)} x(0) + \sum_{f=b+1}^{n} \lambda_f e^{\lambda_f t} A^{(f)} x(0) \end{aligned}\right\} \tag{11}$$

The second terms in eq. 11 can be neglected for time $t$ $(t > T.\ T \approx O(1))$ on this time scale, because of rapid damping off of the term $e^{\lambda_f t}$ $(f = b+1, \ldots, n)$ (when $e^t = \epsilon$, $t = \ln(1/\epsilon) \approx O(1)$).

That is, for $t > T(T \approx O(1/\lambda_f) \approx O(1), f = b+1, \ldots, n)$

$$\left.\begin{aligned} x(t) &= \sum_{h=1}^{b} e^{\lambda_h t} A^{(h)} x(0) \\ \frac{d}{dt} x(t) &= \sum_{h=2}^{b} \lambda_h e^{\lambda_h t} A^{(h)} x(0) \end{aligned}\right\} \tag{12}$$

and

$$\begin{aligned} \left| \frac{d}{dt} x_i \right| &= \left| \sum_{h=2}^{b} \lambda_h e^{\lambda_h t} (A^{(h)} x(0))_i \right| \\ &\leq |\lambda_h|_{\max} \cdot \sum_{h=1}^{b} |e^{\lambda_h t} (A^{(h)} x(0))_i| \quad (i = 1, \ldots, n) \end{aligned} \tag{13}$$

If reduction of order of magnitude by cancelling out between the terms $e^{\lambda_h t}(A^{(h)} x(0))_i$ $(h = 1, \ldots, b)$ does not occur, $x_i(t)$ is of the order of the maximum term in $e^{\lambda_h t}(A^{(h)} x(0))_i$ $(h = 1, \ldots, b)$ (and thus, of the order of $\sum_{h=1}^{b} |e^{\lambda_h t}(A^{(h)} x(0))_i|$). Thus, for the fast component $X_j$ $(j = r+1, \ldots, n)$,

$$\left| \frac{d}{dt} x_j \right| \leq |\lambda_h|_{\max} \cdot |x_j(t)| \ll |k_{jj}| \cdot |x_j(t)| \tag{14}$$

because $|\lambda_h|_{max} \lesssim O(\epsilon)$ and $k_{jj} \approx O(1)$ for the fast components.

If the reduction of order of magnitude occurs for a fast component $X_j$ by summing the terms $e^{\lambda_h t}(A^{(h)}x(0))_j$, then

$$\sum_{h=1}^{b} |e^{\lambda_h t}(A^{(h)}x(0))_j| \gg \left| \sum_{h=1}^{b} e^{\lambda_h t}(A^{(h)}x(0))_j \right| = |x_j(t)| \tag{15}$$

and eq. 14 may not hold on the fast component $X_j$ at $t \approx O(1)$. Such cases may occur when the concentrations of a group of fast components, $x_j(t)$, at $t \approx O(1)$ are much smaller than their maximum values. Detailed discussions of these cases are given in appendix A and B.

As verified in appendix A, for every fast component $X_j$ (even in the cases when eq. 15 holds at $t \approx O(1)$), the following steady/equilibrium relation (eq. 16 or 17)

$$\left|\frac{d}{dt}x_j(t)\right| = \left|\sum_{i=1}^{n} k_{ji}x_i(t)\right| \ll |k_{jj}x_j(t)| \tag{16}$$

$$\left|\frac{d}{dt}x_j(t)\right| \leq O(\epsilon)x_j(t) \tag{17}$$

holds after a proper induction period, $T^{\circ}$ ($T^{\circ} < (1/\epsilon)$), which is small compared to the total reaction time span from the start to equilibrium. We denote hereafter the induction period after which the steady/equilibrium relation holds by $T^{\circ}$. When the above steady/equilibrium relation holds on $X_j$, $X_j$ is either in a quasi-steady state or in quasi equilibrium with some neighboring components or in both. The definitions of quasi-steady state and quasi equilibrium were given in ref. 1.

The steady/equilibrium relation (eq. 16) shows that to put

$$\frac{d}{dt}x_j = \sum_{i=1}^{n} k_{ji}x_i = 0 \quad (j = r+1, \ldots, n) \tag{18}$$

or

$$\frac{d}{dt}y = Q_{mr}x_r + Q_{mm}y = 0 \tag{19}$$

is a good approximation for $t > T^{\circ}$, in order to obtain $x_j$ ($j = r+1, \ldots, n$) as a linear combination of $x_i$ which are directly reacting with $x_j$. We denote this approximation by steady/equilibrium approximation. This is proved as follows. The steady/equilibrium relation is rewritten as

$$\frac{d}{dt}x_j = \sum_{i=1}^{n} k_{ji}x_i = c_j x_j \quad (j = r+1, \ldots, n), \tag{20}$$

where

$$|c_j| \leq |O(\epsilon)k_{jj}| \tag{21}$$

Solving eq. 20 with respect to $x_j$, we obtain

$$x_j = \frac{-1}{k_{jj} - c_j} \sum_{\substack{i=1 \\ (i \neq j)}}^{n} k_{ji}x_i \doteqdot \left(1 + \frac{c_j}{k_{jj}}\right)\bar{x}_j, \tag{22}$$

where

$$\bar{x}_j = \frac{-1}{k_{jj}} \sum_{\substack{i=1 \\ (i \neq j)}}^{n} k_{ji}x_i \tag{23}$$

is the true steady-state or equilibrium value of $x_j$ obtained by solving eq. 18.

From eq. 22, we obtain the relation,

$$|(x_j - \bar{x}_j)/x_j| \doteq |c_j/k_{jj}| \lesssim O(\epsilon) \tag{24}$$

Eq. 24, which is equivalent to the steady/equilibrium relation (eq. 16), shows the accuracy of the approximation (eq. 18).

Eq. 18 is equivalent to, neglecting small $\lambda_h$ $(h = 1,\ldots, b)$ in the $j$th row vector $(j = r+1,\ldots, n)$ in eq. 10,

$$((Q - \lambda_h E)A^{(h)})_j = (QA^{(h)})_j = 0 \quad (h = 1,\ldots,b; \quad j = r+1,\ldots,n) \tag{25}$$

By substituting $(QA^{(h)})_j = 0$ into the equation obtained by multiplying $Q$ from the left-hand side of the upper equation of eq. 12 and from eq. 1, we obtain eq. 18 for $t > T^{o}$.

In this way, the steady/equilibrium approximation holds on the fast components for the larger part of the reaction time course to equilibrium after a comparatively short induction period $T^{o}$.

In order to obtain the large (of O(1)) and small (of O($\epsilon$) or less) eigenvalues separately and introduce the approximation easily, $Q_{mm}$ is reduced to the following 'reduced' form with regard to the thick arrows by rearranging the components;

$$Q_{mm} = \begin{pmatrix} M_1 & & & [O(1)] \\ & M_2 & & \\ & & \ddots & \\ [O(\epsilon)] & & & M_k \end{pmatrix} \tag{26}$$

where $M_p$ $(p = 1,\ldots, k)$ are 'irreducible' submatrices corresponding to each group of fast components. When group A is the upper group of B, $M_A$ corresponding to A is placed to the upper left of $M_B$ corresponding to B. The lower left part in eq. 26 denoted by [O($\epsilon$)] contains the matrix elements of O($\epsilon$) or less (including zero), and the submatrix $M_p$ $(p = 1,\ldots, k)$ and the upper right part denoted by [O(1)] contain matrix elements of O(1) or less than O(1).

The solutions of the rate equation (eq. 1) are examined separately in the following two cases.

### 2.1. When all the groups of fast components are open ($\|Q_{mm}\| \approx O(1)$)

When the steady/equilibrium relation holds on the fast components in the open group, they are in a quasi-steady state or in quasi equilibrium with some neighboring components or in both [1].

We show that the molar fraction of each fast component in an open group can be expressed approximately by the linear combination of the components not contained in the group at $t > T^{o}$ and can be eliminated from the reaction scheme without disturbing the mass conservation law ($\Sigma_i x_i = 1$; $x_i$ are the remaining components). Here we deal with the case when all the groups are open.

We denote the fast components belonging to the open group G corresponding to submatrix $M_p$ in $Q_{mm}$ as $X_d,\ldots, X_{d+g}$.

The true equilibrium concentrations of the fast components belonging to the open group G are smaller by a factor $\epsilon$ (or less than $\epsilon$) than those of the components $X_v$ which do not belong to G and on which thick arrows from G terminate, because

$$\frac{x_{d+l}(\infty)}{x_v(\infty)} = \frac{k_{d+l,v}}{k_{v,d+l}} \lesssim O(\epsilon) \quad (l = 0,\ldots,g) \tag{27}$$

where $x_v(\infty)$ is the equilibrium molar fraction of $X_v$.

The fact that at least one thick arrow originates from fast component $X_{d+l}$ $(0 \leqq l \leqq g)$ belonging to G and is directed toward another slow or fast component $X_v$ $(1 \leqq v \leqq d-1)$ not belonging to G means that there is at least one rate constant $k_{v,d+l}$ $(1 \leqq v \leqq d-1)$ of O(1) and $-(\Sigma_{j=0}^{g} k_{d+j,d+l}) \approx O(1)$ for at least one $X_{d+l}$ $(0 \leqq l \leqq g)$ and so we can show that,

$$\|M_p\| \approx O(1), \tag{28}$$

where $\|M_p\|$ denotes the absolute value of the determinant of $M_p$. Because all the groups of fast components are open, all the $\|M_p\|$ $(p = 1, \ldots, k)$ are of O(1) and thus

$$\|Q_{mm}\| \approx O(1). \tag{29}$$

The approximate values of the large eigenvalues of $Q$ of O(1) are obtained by neglecting terms of $O(\epsilon)$ or less in the secular equation (eq. 7), i.e.,

$$|Q - \lambda E| = (-\lambda)^r \cdot |Q_{mm} - \lambda E| = 0. \tag{30}$$

We obtained $m$ $(= n - r)$ real and negative eigenvalues of O(1) from the above ($b = r$ in eqs. 11–13). Eq. 30 is approximately written as

$$(-\lambda)^r \cdot |M_1 - \lambda E| \cdot |M_2 - \lambda E| \cdot |M_k - \lambda E| = 0. \tag{31}$$

This shows that the large eigenvalues of O(1) can be obtained separately from each secular equation,

$$|M_p - \lambda E| = 0 \quad (p = 1, \ldots, k) \tag{32}$$

The $r - 1$ small eigenvalues of the secular equation (eq. 7) (one eigenvalue is zero $(= \lambda_1)$) are of $O(\epsilon)$ or less.

It can be shown that the approximate values of $r - 1$ small eigenvalues are obtained by neglecting all $\lambda$ in the submatrix $(Q_{mm} - \lambda E)$ of the secular equation (eq. 7). It is equal to solve,

$$|W(\lambda)| = 0 \tag{33}$$

where

$$W(\lambda) \equiv \left( \begin{array}{c|c} Q_{rr} - \lambda E & Q_{rm} \\ \hline Q_{mr} & Q_{mm} \end{array} \right) \tag{34}$$

Let us assign $r - 1$ small eigenvalues of $O(\epsilon)$ or less to $\lambda_2, \ldots, \lambda_r$ and $m$ large eigenvalues of O(1) to $\lambda_{r+1}, \ldots, \lambda_n$ ($b = r$ in eqs. 11–13).

After a proper induction period $T^{\circ}$, the steady/equilibrium relation holds on all the fast components. When $X_{d+l}$ belonging to an open group G is in quasi equilibrium with some components, $X_h$, at least one of them satisfies the following condition (a or b): (a) $X_h$ belongs to G and is also in a quasi-steady state; (b) $X_h$ does not belong to G and a thick arrow is directed to it from G.

Simplification of the reaction scheme by applying the steady/equilibrium approximation to the fast components is done as follows. At $t > T^{\circ}$, we apply the steady/equilibrium approximation to $(g + 1)$ rate equations for the components $X_{d+l}$ $(l = 0, \ldots, g)$ belonging to an open group G and we obtain,

$$\frac{d}{dt} x_{d+l} = - \sum_{\substack{i=1 \\ (i \neq d+l)}}^{n} k_{i,d+l} x_{d+l} + \sum_{\substack{i=1 \\ (i \neq d+l)}}^{n} k_{d+l,i} x_i = 0$$
$$(l = 0, \ldots, g) \tag{35}$$

Since $\|M_p\| \approx O(1)$, we can solve them with respect to $x_{d+l}$ $(l = 0, \ldots, g)$ and obtain them as a linear combination of $x_i$ (which are not contained in G but react directly with G).

By repeating the same procedure on every open group and summarizing the results, the fast components in open groups can be solved as a linear combination of the slow components: i.e., we obtain from eq. 19,

$$y = -Q_{mm}^{-1}Q_{mr}x_r, \tag{36}$$

where the existence of $Q_{mm}^{-1}$ is evident from $\|Q_{mm}\| \approx O(1)$.

By substituting eq. 36 into the upper half of the rate equation (eq. 1), we obtain

$$\left.\begin{aligned} \frac{d}{dt}x_r &= Q_{rr}x_r + Q_{rm}y = Q'_{rr}x_r \\ \text{where} \quad & \\ Q'_{rr} &= Q_{rr} - Q_{rm}Q_{mm}^{-1}Q_{mr} \end{aligned}\right\} \tag{37}$$

Eq. 37 is the rate equation of the reduced first-order reactions between the slow components, $X_1, \ldots, X_r$, after application of the steady/equilibrium approximation. The apparent rate constants between the slow components $X_i$ and $X_j$ in the reduced reaction scheme, $k_{ij}$, are given by the matrix elements $(Q'_{rr})_{ij}$ which are of $O(\epsilon)$ or less. The matrix $Q_{rr}$ represents direct interconversions between the slow components. $-Q_{rm}Q_{mm}^{-1}Q_{mr}$ gives the apparent rate constants of interconversion between the slow components through the eliminated fast components whose matrix elements are given as

$$(-Q_{rm}Q_{mm}^{-1}Q_{mr})_{ij} = -\sum_{k=1}^{m}\sum_{l=1}^{m}(Q_{rm})_{ik}\frac{\Delta_{lk}^{(mm)}}{|Q_{mm}|}(Q_{mr})_{lj}$$

$$(i, j = 1, \ldots, r) \tag{38}$$

where $(Q_A)_{ij}$ is the $(i, j)$ element of the matrix $Q_A$ and $\Delta_{lk}^{(mm)}$ the cofactor of matrix element $(Q_{mm})_{lk}$. The obtained conclusions about the reduced reaction scheme are as follows. If both slow components $X_i$ and $X_j$ are directly reacting with the fast components belonging to the same group G corresponding to $M_p$ $(p = 1, \ldots, k)$ or if they are reacting with the fast components belonging to a cluster of groups, the matrix element $(-Q_{rm}Q_{mm}^{-1}Q_{mr})_{ij}$ is not zero. In a cluster, open groups corresponding to $M_p, \ldots, M_{p+q}$ are connected with each other in some way by the thick or thin arrows. In all other cases, i.e., when there are other slow components in every route connecting $X_i$ and $X_j$, $(-Q_{rm}Q_{mm}^{-1}Q_{mr})_{ij} = 0$.

The concentrations of the fast components in open groups at quasi-steady state or at quasi equilibrium are of $O(\epsilon)$ or less, because in eq. 36 elements of $Q_{mm}^{-1}$, $Q_{mr}$ and $x_r$ are of O(1) or less, of $O(\epsilon)$ or less, and of O(1) or less, respectively. Thus, the mass conservation law of the reduced reaction scheme between the slow components holds approximately.

When the initial concentrations of some fast components in an open group G are of O(1), the induction period $T^\circ$ for the components in G is of O(1) (cf. appendix B), and almost all the mass in G is transferred in a definite manner to the components not belonging to G on which thick arrows from G terminate, within the induction period $T^\circ$, as shown in appendix C. This gives the initial condition of the reaction in the reduced scheme.

In this way, we can eliminate $m$ fast components in the reaction scheme, leaving the reaction pathway through them in general, and can obtain a reaction scheme consisting of $r$ slow components only. The simplified reaction scheme described by the rate equations (eq. 37) may sometimes be further simplified by applying the steady/equilibrium approximation again.

It can be shown by transforming eq. 33 to the next secular equation (eq. 39), that to solve the secular equation,

$$|Q'_{rr} - \lambda E| = 0 \tag{39}$$

for obtaining the relaxation rates, $-\lambda_h$ $(h = 1, \ldots, r)$, of the reaction described by the rate equation (eq.

37), is equivalent to solving the secular equation (eq. 33) for obtaining approximate values of small eigenvalues.

*2.2. When there are closed groups of fast components ($\|Q_{mm}\| \lesssim O(\epsilon)$): Appearance of the principle of fast equilibration*

We show here that the principle of fast equilibration holds on the fast components belonging to a closed group. Here we deal with the case in which there are both types of groups, closed and open. In this case, the steady/equilibrium relation (eq. 16) also holds for every fast component at $t > T^{\circ}$, but we cannot deal with them in just the same way as in section 2.1.

We denote $m - s$ fast components belonging to closed groups as $X_{r+1}, \ldots, X_{n-s}$, and s fast components belonging to open groups as $X_{n-s+1}, \ldots, X_n$.

The submatrix $Q_{mm}$ is also reduced to the form of eq. 26 in this section. The fast components belonging to a closed group G corresponding to the submatrix $M_f$ are denoted by $X_d, \ldots, X_{d+g}$ in the same way as in section 2.1. Because the matrix elements $k_{i,d+l}$ $(i = 1, \ldots, d-1, d+g+1, \ldots, n;\ l = 0, \ldots, g)$ are all of $O(\epsilon)$ or less and $\sum_{j=1}^{n} k_{j,d+l} = 0$ $(l = 0, \ldots, g)$, $|\sum_{i=0}^{g} k_{d+i,d+l}| \lesssim O(\epsilon)$ $(l = 0, \ldots, g)$ and so for closed groups,

$$\|M_f\| \lesssim O(\epsilon) \quad (f = 1, \ldots, h, h \gtrless 1), \tag{40}$$

and thus, $\|Q_{mm}\| = \|M_1| \cdots |M_k\| \lesssim O(\epsilon)$. The submatrices corresponding to closed groups are placed at the head of $Q_{mm}$ and those corresponding to open groups and satisfying $\|M_s\| = O(1)$ $(s = h+1, \ldots, k)$ are at the tail of $Q_{mm}$.

Since $\|M_f\| \lesssim O(\epsilon)$, $x_{d+l}$ $(l = 0, \ldots, g)$ cannot be obtained as the linear combination of $x_i$ only ($X_i$ are the components neighboring G). This is explained as follows. We eliminate $x_{d+g}$ in the rate equations of $X_d, \ldots, X_{d+g-1}$ by applying the steady/equilibrium approximation to $X_{d+g}$ and obtain $g$ rate equations not including $x_{d+g}$. If $g > 1$, the coefficient of $x_{d+l}$ $(l = 0, \ldots, g-1)$ in the resultant rate equation for $X_{d+l}$ is of $O(1)$ and the steady/equilibrium approximation holds again. Thus, we further apply the above procedure and eliminate $x_{d+g-1}$ in the rate equations for $X_d, \ldots, X_{d+g-2}$. Repeating the above procedure we finally obtain the rate equation for $X_d$ not including $x_{d+1}, \ldots, x_{d+g}$. This time, the coefficient of $x_d$ is of $O(\epsilon)$ or less because $\|M_f\| \lesssim O(\epsilon)$ and $x_d$ cannot be obtained by putting the rate equation for $x_d$ equal to zero.

By applying the steady/equilibrium approximation to the $g$ rate equations for $X_{d+1}, \ldots, X_{d+g}$, we can obtain $x_{d+1}, \ldots, x_{d+g}$ as the linear combination of $x_i$ ($X_i$ are the components neighboring G) and the sum of the components in G, $\sum_{l=0}^{n} x_{d+l}$. This is done as follows. Rate equations for $X_{d+1}, \ldots, X_{d+g}$ are transformed to the following form by replacing $x_d$ by the sum of the group, $\sum_{l=0}^{g} x_{d+l}$.

$$\frac{\mathrm{d}}{\mathrm{d}t} x_i = \sum_{j=1}^{n} k_{ij} x_j = \sum_{j=1}^{d-1} k_{ij} x_j + k_{id} \left( \sum_{l=0}^{g} x_{d+l} \right) + \sum_{j=d+1}^{d+g} (k_{ij} - k_{id}) x_j + \sum_{j=d+g+1}^{n} k_{ij} x_j \quad (i \neq d) \tag{41}$$

We denote the matrix whose $(l, l')$ elements are, $k_{d+l,d+l'} - k_{d+l,d}$ $(l, l' = 1, \ldots, g)$, by $M_f^*$, to which the fast components, $X_{d+1}, \ldots, X_{d+g}$, correspond.

$$M_f^* = \begin{pmatrix} k_{d+1,d+1} - k_{d+1,d}, & k_{d+1,d+2} - k_{d+1,d} & \cdots\cdots & k_{d+1,d+n} - k_{d+1,d} \\ k_{d+2,d+1} - k_{d+2,d}, & & & \vdots \\ \vdots & & & \vdots \\ k_{d+g,d+1} - k_{d+g,d} & & \cdots\cdots & k_{d+g,d+g} - k_{d+g,d} \end{pmatrix} \tag{42}$$

The diagonal element, $k_{d+l,d+l} - k_{d+l,d}$ $(l = 1,\ldots,g)$, which is the coefficient of $x_{d+l}$ $(l = 1,\ldots,g)$ in the rate equation of $X_{d+l}$ in eq. 41, is of O(1) and $X_{d+1},\ldots,X_{d+g}$ are treated as fast components. The dimension of $M_f^*$ is $g$ and $\|M_f^*\| = O(1)$ (in contrast to $\|M_f\| \lesssim O(\epsilon)$) because at least one of the sum of the column, $\sum_{l=1}^{g} (k_{d+l,d+l'} - k_{d+l,d})$ $(l' = 1,\ldots,g)$, is of O(1). (because $\sum_{l=0}^{g} k_{d+l,d+l'} \lesssim O(\epsilon)$, and at least one of $d_{d+l,d}$ is of O(1)). Thus, at $t > T^{o}$, we can apply the steady/equilibrium approximation to $X_{d+1},\ldots,X_{d+g}$ and obtain $x_{d+l}$ $(l = 1,\ldots,g)$ as the linear combination of $x_i$ and $\sum_{l=0}^{g} x_{d+l}$.

Moreover, at $t > T^{o}$, we can neglect terms smaller than the largest terms by the factor $\epsilon$ or less using eq. B1, i.e.,

$$k_{d+l,i}x_i \ll x_{d+l} \quad (l = 0,\ldots,g;\ i = 1,\ldots,d-1, d+g+1,\ldots,n) \tag{43}$$

for closed groups (cf. appendix B) and thus,

$$\frac{\mathrm{d}}{\mathrm{d}t}\begin{pmatrix} x_{d+1} \\ \vdots \\ x_{d+g} \end{pmatrix} = \begin{pmatrix} k_{d+1,d} \\ \vdots \\ k_{d+g,d} \end{pmatrix} \sum_{l=0}^{g} x_{d+l} + M_f^* \begin{pmatrix} x_{d+1} \\ \vdots \\ x_{d+g} \end{pmatrix} = 0 \tag{44}$$

The accuracy of the principle of fast equilibration is not changed by the neglect introduced into eq. 44. Of course, eq. 44 holds also at true equilibrium, so it can be solved for $x_{d+l}$ $(l = 1,\ldots,g)$ in the following form:

$$\begin{pmatrix} x_{d+1} \\ \vdots \\ x_{d+g} \end{pmatrix} = -M_f^{*-1} \begin{pmatrix} k_{d+1,d} \\ \vdots \\ k_{d+g,d} \end{pmatrix} \sum_{l=0}^{g} x_{d+l} = \begin{pmatrix} K_{d+1} \\ \vdots \\ K_{d+g} \end{pmatrix} \sum_{l=0}^{g} x_{d+l} \tag{45}$$

where $K_{d+l}$ $(l = 1,\ldots,g)$ is the proportion of $x_{d+l}$ to $\sum_{l=0}^{g} x_{d+l}$ at true equilibrium. Eq. 45 shows that $(g+1)$ components, $X_{d+l}$ $(l = 0,\ldots,g)$, which belong to a closed group and corresponding to the submatrix $M_f$ in $Q_{mm}$ ($\|M_f\| \lesssim O(\epsilon)$), attain quasi equilibrium at $t > T^{o}$.

On the other hand, by summing up the rate equations for $X_d,\ldots,X_{d+g}$, the rate equation for the sum of $x_d,\ldots,x_{d+g}$, $\sum_{l=0}^{g} x_{d+l}$, is obtained as

$$\begin{aligned} \frac{\mathrm{d}}{\mathrm{d}t}\left(\sum_{l=0}^{g} x_{d+l}\right) &= \sum_{l=0}^{g} \frac{\mathrm{d}}{\mathrm{d}t} x_{d+l} = \sum_{j=1}^{d-1}\left(\sum_{i=d}^{d+g} k_{ij}\right) x_j \\ &+ \left(\sum_{i=d}^{d+g} k_{id}\right) \sum_{l=0}^{g} x_{d+l} \\ &+ \sum_{j=d+1}^{d+g}\left(\sum_{i=d}^{d+g} (k_{ij} - k_{id})\right) x_j \\ &+ \sum_{j=d+g+1}^{n}\left(\sum_{i=d}^{d+g} k_{ij}\right) x_j \end{aligned} \tag{46}$$

By introducing eq. 45 into eq. 46 and into the rate equations for $X_1,\ldots,X_{d-1}$, $X_{d+g+1},\ldots,X_n$, transformed into the form of eq. 41, we obtain the simplified new set of rate equations corresponding to the reduced reaction scheme containing the components $x_1,\ldots,x_{d-1}$, $x_{d+g+1},\ldots,x_n$ and $\sum_{l=0}^{g} x_{d+l}$. The mass conservation law holds in this scheme.

In eq. 46 the coefficient of the sum of the elements in G, $\sum_{l=0}^{g} x_{d+l}$, is of $O(\epsilon)$ or less because rate constants of O(1) are cancelled and it can be treated as a slow component.

When all the closed groups are in quasi equilibrium at $t > T^{o}$ $(T^{o} < O(1/\epsilon))$, we apply the above procedure to all the closed groups. This is done as follows. One component $X_{d(f)}$ in every closed group corresponding to the submatrix $M_f$ ($f = 1, \ldots, h$, $\|M_f\| \leq O(\epsilon)$) is replaced by the sum of the components in the group, $\sum_{l=0}^{g(f)} x_{d(f)+l}$, and every rate equation except for $X_{d(f)}$ ($f = 1, \ldots, h$) is changed to the form of eq. 47.

$$\begin{aligned}\frac{\mathrm{d}}{\mathrm{d}t}x_i &= \sum_{j=1}^{r} k_{ij}x_j + \sum_{f=1}^{h}(k_{i,d(f)})\sum_{l=0}^{g(f)} x_{d(f)+l} \\ &+ \sum_{f=1}^{h}\sum_{l=1}^{g(f)}(k_{i,d(f)+l} - k_{i,d(f)})x_{d(f)+l} \\ &+ \sum_{j=n-s+1}^{n} k_{i,j}x_j \quad (i = 1,\ldots,r \text{ and } r+h+1,\ldots,n)\end{aligned} \tag{47}$$

The rate equations for $h$ sums of the components in $h$ closed groups are given in eq. 48

$$\begin{aligned}\frac{\mathrm{d}}{\mathrm{d}t}\left(\sum_{l=0}^{g(f)} x_{d(f)+l}\right) &= \sum_{j=1}^{r}\left(\sum_{l=0}^{g(f)} k_{d(f)+l,j}\right)x_j \\ &+ \sum_{f'=1}^{h}\left(\sum_{l=0}^{g(f)} k_{d(f)+l,d(f')}\right)\sum_{l'=0}^{g(f')} x_{d(f')+l'} \\ &+ \sum_{f'=1}^{h}\sum_{l'=1}^{g(f')}\sum_{l=0}^{g(f)}(k_{d(f)+l,d(f')+l'} - k_{d(f)+l,d(f')})x_{d(f')+l'} \\ &+ \sum_{j=n-s+1}^{n}\left(\sum_{l=0}^{g(f)} k_{d(f)+l,j}\right)x_j \quad (f = 1,\ldots,h)\end{aligned} \tag{48}$$

where $x_{d(f)}, \ldots, x_{d(f)+g(f)}$ are the fast components belonging to the closed group corresponding to each $M_f$ ($f = 1, \ldots, h$). Because $h$ coefficients of $\sum_{l=0}^{g(f)} x_{d(f)+l}$ in their rate equations are of $O(\epsilon)$ or less, the sums are treated as slow components. And thus, the $h$ rate equations (eq. 48) for sums of $h$ closed groups are placed in the row from $r+1$ to $r+h$ in the following matrix form of the rate equations (eq. 49) in which the rate equations in the form of eqs. 47 and 48 are contained:

$$\frac{\mathrm{d}}{\mathrm{d}t}\begin{pmatrix} x_r^* \\ y^* \end{pmatrix} = Q^*\begin{pmatrix} x_r^* \\ y^* \end{pmatrix} \tag{49}$$

where

$$x_r^* = \begin{pmatrix} x_1 \\ \vdots \\ x_r \\ \sum_{l=0}^{g(1)} x_{d(1)+l} \\ \vdots \\ \sum_{l=0}^{g(h)} x_{d(h)+l} \end{pmatrix} \qquad y^* = \begin{pmatrix} y_{h+1}^* \\ \vdots \\ y_m^* \end{pmatrix} \tag{50}$$

where $m-h$ elements $y^*_{h+1},\ldots,y^*_m$ are the molar fractions of fast components obtained by omitting one component, $x_{d(f)}$, from each closed group corresponding, respectively, to $M_f$ ($f=1,\ldots,h$).

Thus, $Q^*$ is expressed as

$$Q^* = \begin{array}{c} r \\ r+h \\ n-s \\ \\ \end{array} \left(\begin{array}{cc|cc} Q^*_{rr} & [O(\epsilon)] & [O(\epsilon)] & [O(1)] \\ [O(\epsilon)] & [O(\epsilon)] & [O(\epsilon)] & [O(1)] \\ \hline [O(\epsilon)] & [O(1)] & M^* & [O(1)] \\ [O(\epsilon)] & [O(\epsilon)] & [O(\epsilon)] & M \end{array}\right)$$

$$\equiv \begin{array}{c} r+h \\ \\ \end{array} \left(\begin{array}{c|c} Q^*_{rr} & Q^*_{rm} \\ \hline Q^*_{mr} & Q^*_{mm} \end{array}\right) \tag{51}$$

where $Q^*_{rr}$ is the $(r+h)\times(r+h)$ submatrix, whose elements are all of $O(\epsilon)$ or less. In eq. 51, the numbers of rows and columns are shown on the outside of the matrix $Q^*$. $Q^*_{mm}$ is the $(m-h)\times(m-h)$ submatrix, given as

$$Q^*_{mm} = \left(\begin{array}{c|c} M^* & [O(1)] \\ \hline [O(\epsilon)] & M \end{array}\right) = \left(\begin{array}{ccc|ccc} M^*_1 & & [O(\epsilon)] & & [O(1)] & \\ & \ddots & & & & \\ [O(\epsilon)] & & M^*_h & & & \\ \hline & & & M_{h+1} & & [O(1)] \\ & [O(\epsilon)] & & & \ddots & \\ & & & [O(\epsilon)] & & M_k \end{array}\right) \tag{52}$$

where $M^*$ and $M$ are the submatrices of $Q_{mm}$ in which $M_f$ ($f=1,\ldots,h$) and $M_s$ ($s=h+1,\ldots,k$) are contained, respectively. $\|M^*_f\| \approx O(1)$ and thus,

$$\|Q^*_{mm}\| \approx O(1) \tag{53}$$

$Q^*_{rm}$ and $Q^*_{mr}$ are $(r+h)\times(m-h)$ and $(m-h)\times(r+h)$ matrices, respectively, and the orders of magnitude of their elements are shown in eq. 51, which are a little different from those of the elements of $Q_{rm}$ and $Q_{mr}$ given in eqs. 3 and 5.

In spite of some difference between $Q_{rm}$ and $Q_{mr}$ and those of the matrices denoted by an asterisk, $Q^*_{rm}$ and $Q^*_{mr}$, we can proceed in almost the same way as in section 2.1. The eigenvalues obtained from the secular equation

$$|Q^*-\lambda E|=0 \tag{54}$$

are the same as those obtained from the original secular equation (eq. 7), because the relation $|Q^*-\lambda E| = |Q-\lambda E|$ is obtained by adding and subtracting properly rows and columns of the determinant $|Q-\lambda E|$.

The approximate values of $m-h$ large eigenvalues of $O(1)$ and of $r+h$ small eigenvalues of $O(\epsilon)$ or less are obtained from

$$|Q^*_{mm}-\lambda E|=0 \tag{55}$$

and

$$|W^*(\lambda)|=0 \tag{56}$$

respectively, where

$$W^*(\lambda) = \left( \begin{array}{c|c} Q^*_{rr} - \lambda E & Q^*_{rm} \\ \hline Q^*_{mr} & Q^*_{mm} \end{array} \right) \tag{57}$$

The orders of magnitudes of the molar fractions of the elements in closed groups are of O(1) or less, in contrast to the magnitudes of elements in open groups after the proper induction period $T^\circ$.

At $t > T^\circ$,

$$y^* = Q^*_{mr} x^*_r + Q^*_{mm} y^* = 0 \tag{58}$$

is a good approximation for the fast components, $y_{h+1}, \ldots, y_m$.

Eq. 58 can be solved for $y^*$ because $|Q^*_{mm}| \approx O(1)$ as

$$y^* = Q^{*-1}_{mm} Q^*_{mr} x^*_r \tag{59}$$

By substituting the above into the rate equations for $x^*_r$, we obtain,

$$\frac{\mathrm{d}}{\mathrm{d}t} x^*_r = Q^{*\prime}_{rr} x^*_r \tag{60}$$

where

$$Q^{*\prime}_{rr} = Q^*_{rr} - Q^*_{rm} Q^{*-1}_{mm} Q^*_{mr} \tag{61}$$

Eq. 60 is the rate equation of the reduced scheme after application of the steady/equilibrium approximation. In $x^*_r$, the sum of the components in every closed group corresponding to $M_f$ ($|M_f| < O(\epsilon)$, $f = 1, \ldots, h$), for which the principle of fast equilibration holds, is involved as a new slow component (cf. ref. 3). So the mass conservation law holds.

The above procedure gives a unified treatment of both the steady-state approximation and the principle of fast equilibration. However, a more simple and practically more useful expression of the matrix is obtained instead of $Q^*$ when the coefficients of eq. 44 are used for the rows of $Q^*$ corresponding to $M^*$ (cf. eq. 44 and also appendix B). This is to put the coefficients of $x_i$ ($i = 1, \ldots, d-1$ and $d+g+1, \ldots, n$) in the rate equations for $x_{d+l}$ ($l = 1, \ldots, g$) equal to zero.

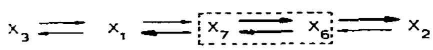
The matrix elements of $Q^{*\prime}_{rr}$ are of O($\epsilon$) or less in the same way as in section 2.1. Thus, further application of two simplifying principles to the reduced scheme is possible by making use of the difference in magnitude of the reduced kinetic constants of O($\epsilon$) or less, if a difference exists.

## 3. Examples

We show here examples of the matrix treatment of steady/equilibrium approximation developed in section 2.

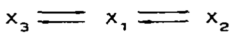
The reaction schemes examined here are those shown as examples in the preceding work [1].

*3.1. A reaction scheme (I) containing an open group, which is equal to scheme XII in ref. 1.*




(I)

This scheme reduces to scheme II after $T^\circ$ by applying the steady/equilibrium approximation to the fast




(II)

components, $X_6$ and $X_7$, in the open group. The rate equations for scheme I are given as

$$\frac{d}{dt}\begin{pmatrix} x_1 \\ x_2 \\ x_3 \\ x_6 \\ x_7 \end{pmatrix} = Q \begin{pmatrix} x_1 \\ x_2 \\ x_3 \\ x_6 \\ x_7 \end{pmatrix} \tag{62}$$

where

$$Q = \left(\begin{array}{c|c} Q_{rr} & Q_{rm} \\ \hline Q_{mr} & Q_{mm} \end{array}\right)$$
$$= \left(\begin{array}{ccc|cc} -(k_{31}+k_{71}), & 0, & k_{13} & 0, & k_{17} \\ 0, & -k_{62}, & 0 & k_{26}, & 0 \\ k_{31}, & 0, & -k_{13} & 0, & 0 \\ \hline 0, & k_{62}, & 0 & -(k_{26}+k_{76}), & k_{67} \\ k_{71}, & 0, & 0 & k_{76}, & -(k_{17}+k_{67}) \end{array}\right), \tag{63}$$

$$Q_m = M_1 = \begin{pmatrix} -(k_{26}+k_{76}), & k_{67} \\ k_{76}, & -(k_{17}+k_{67}) \end{pmatrix}. \tag{64}$$

Fast relaxation rates of O(1), $-\lambda_4$ and $-\lambda_5$, are obtained by solving the secular equation:

$$|Q_{mm} - \lambda E| = 0. \tag{65}$$

The rate equation for the reduced reaction scheme (scheme II) is given as,

$$\frac{d}{dt}\begin{pmatrix} x_1 \\ x_2 \\ x_3 \end{pmatrix} = Q'_{rr} \begin{pmatrix} x_1 \\ x_2 \\ x_3 \end{pmatrix} \tag{66}$$

where,

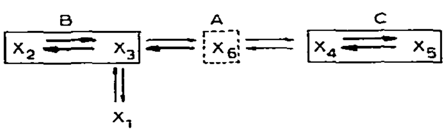
$$Q'_{rr} = Q_{rr} - Q_{rm} Q_{mm}^{-1} Q_{mr}$$
$$= \begin{pmatrix} -k_{31} - (k_{26}k_{67}k_{71}/d), & k_{17}k_{76}k_{62}/d, & k_{13} \\ k_{26}k_{67}k_{71}/d, & -k_{17}k_{76}k_{62}/d, & 0 \\ k_{31}, & 0, & -k_{13} \end{pmatrix} \tag{67}$$

where

$$d = k_{26}k_{17} + k_{26}k_{67} + k_{17}k_{76}.$$

The apparent rate constants of the reduced scheme (scheme II) are given by the matrix elements of $Q'_{rr}$ shown above.

### 3.2. *A scheme (III) containing both open and closed groups, which is equal to scheme XX in ref. 1*




(III)

$$x_1 \rightleftharpoons (x_2 \cdot x_3) \rightleftharpoons (x_4 \cdot x_5) \qquad \text{(IV)}$$

This scheme is reduced to scheme IV (scheme XXI in ref. 1) by applying the steady-equilibrium approximation to an open group A and to two closed groups B and C. The matrix $Q$ in the rate equation (eq. 1) for scheme III is given as,

$$Q=\left(\begin{array}{c|c} Q_{rr} & Q_{rm} \\ \hline Q_{mr} & Q_{mm} \end{array}\right)$$

$$=\left(\begin{array}{c|ccccc} -k_{31} & 0, & k_{13} & 0, & 0, & 0 \\ \hline 0 & -k_{32}, & k_{23} & 0, & 0, & 0 \\ k_{31} & k_{32}, & -(k_{13}+k_{23}+k_{63}) & 0, & 0, & k_{36} \\ 0 & 0, & 0 & -(k_{54}+k_{64}), & k_{45} & k_{46} \\ 0 & 0, & 0 & k_{54}, & -k_{45} & 0 \\ 0 & 0, & k_{63}, & k_{64}, & 0 & -(k_{36}+k_{46}) \end{array}\right), \tag{68}$$

$$Q_{mm}=\left(\begin{array}{cc|c|c} \multicolumn{2}{c|}{M_1} & 0, & 0 \\ & & 0, & k_{36} \\ \hline 0, & 0 & M_2 & k_{46} \\ 0, & 0 & & 0 \\ \hline 0, & k_{63}, & k_{64} & M_3 \end{array}\right). \tag{69}$$

$$\left.\begin{aligned} M_1 &= \begin{pmatrix} -k_{32}, & k_{23} \\ k_{32}, & -(k_{13}+k_{23}+k_{63}) \end{pmatrix}, \\ M_2 &= \begin{pmatrix} -(k_{54}+k_{64}), & k_{45} \\ k_{54}, & -k_{45} \end{pmatrix}, \\ M_3 &= -(k_{36}+k_{46}). \end{aligned}\right\} \tag{70}$$

We make a new set of rate equations replacing $x_2$ and $x_4$ by $(x_2+x_3)$ and $(x_4+x_5)$, respectively, and rearranging,

$$\frac{d}{dt}\begin{pmatrix} x_1 \\ x_2+x_3 \\ x_4+x_5 \\ x_3 \\ x_5 \\ x_6 \end{pmatrix} = Q^*\begin{pmatrix} x_1 \\ x_2+x_3 \\ x_4+x_5 \\ x_3 \\ x_5 \\ x_6 \end{pmatrix}, \tag{71}$$

where,

$$Q^*=\left(\begin{array}{c|c} Q^*_{rr} & Q^*_{rm} \\ \hline Q^*_{mr} & Q^*_{mm} \end{array}\right)$$

$$=\left(\begin{array}{ccc|ccc} -k_{31}, & 0, & 0 & k_{13}, & 0, & 0 \\ k_{31}, & 0, & 0 & -(k_{13}+k_{63}), & 0, & k_{36} \\ 0, & 0, & -k_{64} & 0, & k_{64}, & k_{46} \\ \hline k_{31}, & k_{32}, & 0 & -(k_{13}+k_{23}+k_{63}+k_{32}) & 0, & k_{36} \\ 0, & 0, & k_{54} & 0 & -(k_{54}+k_{45}) & 0 \\ 0, & 0, & k_{64} & k_{63}, & -k_{64} & -(k_{36}+k_{46}) \end{array}\right) \tag{72}$$

$$Q^{*}_{mm}=\left(\begin{array}{cc|c} \multicolumn{2}{c|}{M^{*}} & \begin{matrix} k_{36} \\ 0 \end{matrix} \\ \hline k_{63}, & -k_{64} & M \end{array}\right) \tag{73}$$

$$\left.\begin{aligned} M^{*} &= \left(\begin{array}{c|c} M^{*}_{1} & O \\ \hline 0 & M^{*}_{2} \end{array}\right) \\ M^{*}_{1} &= -(k_{13}+k_{23}+k_{63}+k_{32}) \\ M^{*}_{2} &= -(k_{54}+k_{45}) \\ M &= -(k_{36}+k_{46}) \end{aligned}\right\} \tag{74}$$

The matrix $Q^{*}$ can further be simplified to $Q^{\dagger}$ shown below without losing the accuracy of the approximation, as described in section 2.2 (cf. eq. 44).

$$Q^{\dagger}=\left(\begin{array}{c|c} Q^{*}_{rr} & Q^{*}_{rm} \\ \hline Q^{\dagger}_{mr} & Q^{\dagger}_{mm} \end{array}\right)$$

$$=\left(\begin{array}{ccc|ccc} -k_{31}, & 0, & 0, & k_{13}, & 0, & 0 \\ k_{31}, & 0, & 0, & -(k_{13}+k_{63}), & 0, & k_{36} \\ 0, & 0, & -k_{64} & 0 & k_{64}, & k_{46} \\ \hline 0, & k_{32}, & 0 & -(k_{13}+k_{23}+k_{63}+k_{32}) & 0, & 0 \\ 0, & 0, & k_{54} & 0 & -(k_{45}+k_{54}) & 0 \\ 0, & 0, & k_{64}, & k_{63}, & -k_{64} & -(k_{36}+k_{46}) \end{array}\right) \tag{75}$$

The reduced rate equation after simplification is given as.

$$\frac{\mathrm{d}}{\mathrm{d}t}\begin{pmatrix} x_1 \\ x_2+x_3 \\ x_4+x_5 \end{pmatrix}=Q^{\dagger\prime}_{rr}\begin{pmatrix} x_1 \\ x_2+x_3 \\ x_4+x_5 \end{pmatrix} \tag{76}$$

where,

$$Q^{\dagger\prime}_{rr}=Q^{*}_{rr}-Q^{*}_{rm}Q^{\dagger-1}_{mm}Q^{\dagger}_{mr}$$

$$=\begin{pmatrix} -k_{31}, & \dfrac{k_{13}k_{32}}{k_{23}+k_{32}}, & 0 \\ k_{31}, & -\left(\dfrac{k_{13}k_{32}}{k_{23}+k_{32}}+\dfrac{k_{46}k_{63}k_{32}}{(k_{23}+k_{32})k_{36}}\right), & \dfrac{k_{64}k_{45}}{k_{45}+k_{54}} \\ 0, & \dfrac{k_{46}k_{63}k_{32}}{k_{36}(k_{23}+k_{32})}, & -\dfrac{k_{64}k_{45}}{k_{45}+k_{54}} \end{pmatrix} \tag{77}$$

The approximate values of fast relaxation rates of O(1), $-\lambda_4$, $-\lambda_5$ and $-\lambda_6$ are obtained by solving the secular equation,

$$|Q^{*}_{mm}-\lambda E|=0 \tag{78}$$

as $k_{23}+k_{32}$, $k_{45}+k_{54}$ and $k_{36}$, respectively.

The apparent rate constants of the reduced scheme (scheme IV) are given in $Q^{\dagger\prime}_{rr}$.

## 4. Discussions and conclusions

In the present work we presented the mathematical basis for the methods of simplifying the first-order reaction by the steady/equilibrium approximation, described in the preceding paper [1]. We could determine the conditions under which the approximation holds and evaluate the accuracy of the approximation. When the steady/equilibrium relation (eq. 16) holds on a fast component, the relative deviation of its concentration from the true steady-state or true equilibrium value is small: i.e., the steady/equilibrium approximation is applicable to the component. The steady/equilibrium relation was shown to hold on every fast component after a proper induction period $T^{o}$, which is always smaller than the time of $O(1/\epsilon)$.

As the elementary system including fast components, let us imagine $U_o$ and $U_c$, where $U_o$ consists of fast components belonging to an open group G and the components directly reacting with G (but without direct interactions between the components not belonging to G), and $U_c$ consists of fast components belonging to a closed group.

When the reaction scheme consists of only one $U_o$ or $U_c$ and does not contain other components, the steady/equilibrium relation is established when the terms with large relaxation rates of O(1) damp off and $T^{o} \leq O(1)$ for any initial condition (cf. appendix B). When the reaction scheme is more complicated, $T^{o}$ is dependent on the initial condition. In the case where mass is rapidly flowing into one of $U_o$ (or $U_c$) from the outside of $U_o$ (or $U_c$) at a time of O(1), the steady/equilibrium relation may sometimes not hold on the fast components in it even after damping off of the terms with large relaxation rates. In these cases $T^{o}$ for the fast components in that elementary system is larger than O(1) (nearly of $O(1/\epsilon)$, cf. appendix B). However, when the time is near $O(1/\epsilon)$, the concentration of fast component $X_j$ belonging to $U_o$ (or $U_c$) becomes much larger (by a factor $1/\epsilon$) than at the time of O(1) and the steady/equilibrium relation comes to hold, although the absolute value of $dx_j/dt$ may be as large as that at the time of O(1). This is shown in appendix A and B.

The concept of 'open' and 'closed' groups is important. It was shown that the open and closed groups of the fast components correspond, respectively, to the case when the determinant of the submatrix $M_p$ in the irreducible form of the matrix $Q_{mm}$ is of O(1) and when it is of $O(\epsilon)$ or less. When $|M_p| \approx O(1)$, the steady/equilibrium value of the fast components in the open group G corresponding to $M_p$ is given by the concentration of the components not belonging to G. While, when $|M_p| \leq O(\epsilon)$, the rate equations for the components in the closed group C corresponding to $M_p$ are not independent of each other and the components are in quasi equilibrium with each other after $T^{o}$ only within the group.

## Appendix A

We prove here the following two theorems.

### A1. Theorem I

There is an appropriate period $T_{hi}$ which satisfies $T_{hi} < O(\epsilon^{-h})$ ($h = 0, 1, 2, \ldots$) on each component $X_i$ ($i = 1, \ldots, n$). At $t > T_{hi}$ the following relation holds

$$\left|\frac{d}{dt}x_i\right| \leq O(\epsilon^{h})x_i \tag{A1}$$

Thus, the steady/equilibrium relation (eq. 17) holds on every fast component at $t \gtrsim O(1/\epsilon)$.

### A2. Theorem II

At $t > T_{0i}$ ($T_{0i} < O(1)$), $O(k_{ij}x_j) \leq O(x_i)$. Thus, at $t > T_0$ ($T_0 < O(1)$), fast components in a group are of the same order of magnitude.

### A3. Proof

(I) The rate equation for component $X_i$ is given as

$$\frac{d}{dt}x_i = k_{ii}x_i + \sum_{j(\neq i)} k_{ij}x_j \tag{A2}$$

where $X_j$ are components reacting with $X_i$, $k_{ii}$ is negative and $-k_{ii} \lesssim O(1)$ and $k_{ij}$ $(i \neq j)$ are positive. We put

$$\frac{d}{dt}x_i = a_i(t)x_i \tag{A3}$$

First, we prove that if the following equation (eq. A4) holds at $t = t_1$ $(> 0)$, there must be at least an influx, $k_{ij}x_j$, which is increasing (in the case where $x_i$ is increasing) or decreasing (in the case where $x_i$ is decreasing) more rapidly than $x_i$ following eq. A6 for some period around $t_2$ $(> t_1)$, in order that eq. A4 becomes invalid and eq. A5 comes to hold at $t_2$ $(> t_1)$ for the reason as follows.

$$|a_i| \lesssim O(\epsilon^h) \quad (h = 0, 1, 2, \ldots) \tag{A4}$$

$$|a_i| > O(\epsilon^h) \tag{A5}$$

$$\frac{d}{dt}x_j = a_j(t)x_j, \quad |a_j| > O(\epsilon^h)$$
$$(\text{for some period around } t_2(> t_1)) \tag{A6}$$

We assume that: (1) eq. A4 holds at $t_1$; (2) every $X_j$ which is reacting with $X_i$ is always increasing or decreasing following eq. A7 at $t > t_1$.

$$\frac{d}{dt}x_j = a_j(t)x_j \quad |a_j| \lesssim O(\epsilon^h) \quad (t > t_1) \tag{A7}$$

The function $a_i(t)$ has maximum or minimum values or approaches a constant value at $t_m(> t_1)$ because $x_i$ does not increase or decrease infinitely. This is because,

$$O(\epsilon^p) \lesssim x_i \lesssim 1 \quad (p \approx O(1)) \quad \text{at } t \gtrsim O(1), \tag{A8}$$

from the fundamental assumptions (i, iii and iv) given in the preceding paper [1]. In order to know their values, we differentiate eqs. A2 and A3,

$$\begin{aligned}\frac{d^2}{dt^2}x_i &= \frac{d}{dt}a_i \cdot x_i + a_i\frac{d}{dt}x_i = \frac{d}{dt}a_i \cdot x_i + a_i\left(k_{ii}x_i + \sum_{j(\neq i)} k_{ij}x_j\right)\\ &= k_{ii}\frac{d}{dt}x_i + \sum_{j(\neq i)} k_{ij}\frac{d}{dt}x_j\\ &= k_{ii}a_ix_i + \sum_{j(\neq i)} k_{ij}a_jx_j.\end{aligned} \tag{A9}$$

The time derivative of $a_i$ is zero or nearly zero at $t_m$. So by setting $(d/dt)a_i = 0$ or $\doteqdot 0$ in eq. A9, we obtain the maximum, minimum or their asymptotic values as,

$$|a_i(t_m)| = \left|\sum_{j(\neq i)} k_{ij}a_jx_j\right| \Big/ \left(\sum_{j(\neq i)} k_{ij}x_j\right) \lesssim O(\epsilon^h). \tag{A10}$$

This shows that at any time $t(> t_1)$,

$$|a_i| \lesssim O(\epsilon^h) \tag{A11}$$

Thus, eq. A1 never becomes invalid at the time when eq. A7 holds.

Next we prove that eq. A12 holds only for periods shorter than $T(<O(\epsilon^{-h}))$.

$$\frac{d}{dt}x_i = a_i(t)\cdot x_i, \quad a_i(t) > O(\epsilon^h) \quad (h = \ldots, -2, -1, 0, 1, 2, \ldots) \tag{A12}$$

Putting the minimum value of $a_i(t)$ as $a$,

$$\frac{d}{dt}x_i \geq ax_i, \quad a > O(\epsilon^h) \tag{A13}$$

By integrating eq. A13, we obtain

$$x_i(t_1 + T) \geq x_i(t_1)e^{aT} \tag{A14}$$

When $t_1 = 0$ and $x_i(0) = 0$, we can replace them by $t_2 \approx O(1)$ and $x_i(t_2) \approx O(\epsilon^p)$ $(p \lesssim O(1))$ (cf. eq. A8). Eq. A14 shows that if $T \gtrsim O\epsilon^{-h})$, $x_i(t_1 + T)$ becomes infinite. This contradicts the fact that $x_i \leq 1$.

Thus, it is concluded that $T$ is less than $O(\epsilon^{-h})$ (i.e., for $t_1 \leq t \leq t_1 + T$, eq. A12 is valid).

Next, we prove that the following equation (eq. A15) also holds only for a duration less than $O(\epsilon^{-h})$.

$$\frac{d}{dt}x_i = -a_i(t)\cdot x_i, \quad a_i > O(\epsilon^h) \quad (h = 0, 1, 2, \ldots) \tag{A15}$$

We assume that at any time $t$ $(t_1 \leq t \leq t_1 + T)$, eq. A15 holds. Putting the minimum value of $a_i(t)$ as $a$ we get

$$\frac{d}{dt}x_i \leq -ax_i, \quad a > O(\epsilon^h). \tag{A16}$$

Integrating the above we obtain,

$$x_i(t_1 + T) \leq x_i(t_1)e^{-aT}, \quad x_i(t_1) \leq 1 \tag{A17}$$

On the other hand, eq. A8 holds at $t \gtrsim O(1)$. Thus, $T$ is less than $O(\epsilon^{-h})$, i.e., eq. A15 holds for a period less than $O(\epsilon^{-h})$.

Finally we prove that there is an appropriate time $T_{hi}$ $(<O(\epsilon^{-h}))$ for each $X_i$, at which eq. A1 holds and after which $(t > T_{hi})$ it never becomes invalid.

First we deal with the case where $x_i$ is increasing. We assumed that eq. A18 holds at $t_0$.

$$\frac{d}{dt}x_i = a_i x_i \quad 0 \leq a_i \leq O(\epsilon^h) \quad (h = 0, 1, 2, \ldots). \tag{A18}$$

In order that eq. A18 becomes invalid at $t_1$ $(> t_0)$, there must be at least one component $X_j$ reacting with $X_i$ and which increases for some period around $t_1$, satisfying eq. A19.

$$\frac{d}{dt}x_j = a_j x_j, \quad a_j > O(\epsilon^h) \tag{A19}$$

As shown before, eq. A19 continues to hold only for the duration $T$ $(<O(\epsilon^{-h}))$. So this came to hold at $t_2$ $(>(t_1 - T))$. If eq. A19 is the equation continuing from $t = 0$, $t_1 \leq T < O\epsilon^{-h})$. If not, then in order that eq. A19 comes to hold at $t_2$ there must be a component $X_p$ reacting with $X_j$ and which satisfies eq. A20 at $t_3$ $(\geq t_2 - T \geq t_1 - 2T)$.

$$\frac{d}{dt}x_p = a_p x_p, \quad a_p > O(\epsilon^h) \tag{A20}$$

In this way we obtain a series of components $X_i, X_j, X_p, \ldots$ which came to satisfy equations like eq. A19 or A20 at $t_q$ $(\geq t_1 - (q-1)T)$ $(q = 2, 3, \ldots)$. But this series cannot continue infinitely or form cycles, because of the fundamental assumption (v) in ref. 1 (energy is not supplied) and we certainly arrive at an equation like eq. A19 or A20 which held at $t = 0$. This shows that $t_1 \lesssim O(1)T < O(\epsilon^{-h})$. Thus, $T_{hi}$ exists and must be less than $O(\epsilon^{-h})$. At $t > T_{hi}$, eq. A18 never becomes invalid.

For the case where $x_i$ is decreasing, we can prove the above in a similar way.

(II) From theorem I we obtain,

$$\left|\frac{d}{dt}x_q\right| \leq O(1)x_q \quad (q = i \text{ or } j) \quad \text{at } t > T_{0q}\ (T_{0q} < O(1)) \tag{A21}$$

This means,

$$O(k_{ij}x_j) \leq O(x_i) \quad \text{and} \quad O(k_{ji}x_i) \leq O(x_j),$$
$$\text{at} \quad t > T_{0q}\ (T_{0q} < O(1)) \tag{A22}$$

because if $O(k_{ij}x_j) > O(x_i)$ then $(d/dt)x_i > O(1)\ x_i$. When components $X_i$ and $X_j$ are connected by thick arrows in both directions (i.e., $O(k_{ij}) = O(k_{ji}) = 1$), we get,

$$O(x_i) = O(x_j) \quad \text{at} \quad t > T_0\ (T_0 < O(1)) \tag{A23}$$

Thus, from the definition of the group, all the fast components in a group are of the same order of magnitude at $t \gtrsim O(1)$, and the total mass of the components in the group is also of the same order with them because one group has not so many components.

## Appendix B

We show here the necessary and sufficient conditions in order that $T^o$ is of O(1) or less for the fast components (A) in the closed group and (B) in the open group. $T^o$ is the induction period required for establishment of a quasi-steady state or quasi-equilibrium.

### *B1. Closed group*

The necessary and sufficient condition under which the induction period $T^o$, before the establishment of a quasi equilibrium on the fast components in the closed group G, $x_d, \ldots, x_{d+g}$, is of O(1) or less is that the following relation (eq. B1) continues to hold at $t > T'$ ($T' \lesssim O(1)$) on every fast component in G, $X_{d+l}$, and its neighboring component, $X_i$,

$$x_{d+l} \gg k_{d+l,i}x_i \quad (l = 0, \ldots, g;\ i = 1, \ldots, d-1, d+g+1, \ldots, n). \tag{B1}$$

This is proved as follows. When the principle of fast equilibration holds on each component in G, the steady/equilibrium relation holds on them, i.e.,

$$\left|\frac{d}{dt}x_{d+l}\right| \ll |k_{d+l,d+l}x_{d+l}| = x_{d+l} \quad (l = 0, \ldots, g) \tag{B2}$$

By summing the above we obtain,

$$\left|\frac{d}{dt}\left(\sum_{l=0}^{g} x_{d+l}\right)\right| \ll \sum_{l=0}^{g} x_{d+l} \tag{B3}$$

The rate equation for the sum of the components in group G is given as,

$$\frac{d}{dt}\left(\sum_{l=0}^{g} x_{d+l}\right) = \sum_{i=1}^{d-1}\left(\sum_{l=0}^{g} k_{d+l,i}\right)x_i$$

$$+\sum_{j=0}^{g}\left(\sum_{l=0}^{g}k_{d+l,d+j}\right)x_{d+j}$$
$$+\sum_{i=d+g+1}^{n}\left(\sum_{l=0}^{g}k_{d+l,i}\right)x_i \tag{B4}$$

The coefficients of $x_{d+j}$ $(0 \leqq j \leqq g)$ in the right-hand side of the above rate equation are all negative or zero and their absolute values are of O($\epsilon$) or less because they contain the diagonal elements of $Q$, $k_{d+l,d+l} = -\sum_{i=1(\neq d+l)}^{n} k_{i,d+l}$, and the rate constants of O(1) connecting between $x_{d+j}$ $(0 \leqq j \leqq g)$ are cancelled out by summation. The coefficients of $x_i$ $(i = 1,\ldots, d-1, d+g+1,\ldots, n)$ are all positive. If any one of $k_{d+l,i}x_i$ $(l = 0,\ldots, g;\ i = 1,\ldots, d-1, d+g+1,\ldots, n)$ is of the same order or larger than $x_{d+l}$, then

$$\left|\frac{d}{dt}\sum_{l=0}^{g}x_{d+l}\right| \geq x_{d+l} \simeq \sum_{l=0}^{g}x_{d+l} \tag{B5}$$

The second relation in eq. B5 is proved in appendix A. The relation eq. B5, contradicts eq. B3. Thus, the eq. B1 is necessary for the establishment of a quasi-equilibrium.

Differentiating eq. B4, the second derivative of the sum of group G is,

$$\frac{d^2}{dt^2}\left(\sum_{l=0}^{g}x_{d+l}\right) = \sum_{i=1}^{d-1}\left(\sum_{l=0}^{g}k_{d+l,i}\right)\frac{d}{dt}x_i$$
$$+\sum_{j=0}^{g}\left(\sum_{l=0}^{g}k_{d+l,d+j}\right)\frac{d}{dt}x_{d+j}$$
$$+\sum_{i=d+g+1}^{n}\left(\sum_{l=0}^{g}k_{d+l,i}\right)\frac{d}{dt}x_i \tag{B6}$$

The necessary condition to maintain eq. B3 stable is the following relation (eq. B7),

$$\left|\frac{d^2}{dt^2}\left(\sum_{l=0}^{g}x_{d+l}\right)\right| \ll \sum_{l=0}^{g}x_{d+l}. \tag{B7}$$

From eq. B6 and the relation $|(d/dt)x_k| \lesssim O(1)x_k$ $(k = 1,\ldots, n)$ at $t \gtrsim O(1)$ as shown in appendix A, eq. B1 holds when eq. B1 holds.

To hold, the relation eq. B7, at $t > T'$ $(T' \lesssim O(1))$ is sufficient for the establishment of quasi equilibrium on the fast component in the closed group within the time of O(1) as shown below. When eq. B7 holds, $k_{d+l,i}x_i$ is sufficiently smaller than $|k_{d+l,d+l'}x_{d+l'}|$ $(l' = 0,\ldots, g)$ in the rate equation of $x_{d+l}$ and can be neglected. Thus, we obtain the rate equations,

$$\frac{d}{dt}x_{d+l} = \sum_{l'=0}^{g}k_{d+l,d+l'}x_{d+l'} \quad (l' = 0,\ldots,g) \tag{B8}$$

The solutions of the above are described by the sum of the exponential terms with large $\lambda_i$ of O(1) and the constant term, $x_{d+l}(\infty)$ and do no contain the terms with small $\lambda_i$ of O($\epsilon$) or less. Thus, eq. B2 holds within the time of O(1), because of the rapid damping off of the terms with large $\lambda_i$.

Using this necessary and sufficient condition, we obtained method iv in section 2.4 of ref. 1 and determined the order of $T^{\circ}$ in the examples described in previous work [1].

*B2. Open group*

The necessary and sufficient condition in order that the induction period $T^{\circ}$ required for the establishment of the steady/equilibrium relation on each component $X_{d+l}$ $(l = 0,\ldots, g)$ in an open group

G is of O(1) or less is that the following relation (eq. B9) continues to hold at $t > T'$ ($T' \lesssim$ O(1)) on every component $X_j$ which does not belong to G but directly reacting with $X_{d+l}$,

$$k_{d+l,j}\left|\frac{d}{dt}x_j\right| \leq O(\epsilon)x_{d+l} \quad (j=1,\ldots,d-1, d+g+1,\ldots,n). \tag{B9}$$

When the steady/equilibrium relation holds stably on $X_d, \ldots, X_{d+g}$, at $t > T^o$,

$$\left|\frac{d}{dt}x_{d+l}\right| \leq O(\epsilon)x_{d+l} \quad (l=0,\ldots,g) \tag{B10}$$

and

$$\left|\frac{d^2}{dt^2}x_{d+l}\right| \leq O(\epsilon)x_{d+l}. \tag{B11}$$

Introducing eqs. B10 and B11 into eq. B6 and using $|\sum_{l=0}^{g} k_{d+l,d+j}| \lesssim$ O(1), we obtain

$$\left|\sum_{\substack{i=1,\ldots,d-1;\\ d+g+1,\ldots,n}} \left(\sum_{l=0}^{g} k_{d+l,i}\right)\frac{d}{dt}x_i\right| \leq O(\epsilon)\sum_{l=0}^{g} x_{d+l} \tag{B12}$$

Each $x_{d+j}$ is of the same order as $\sum_{j=0}^{g} x_{d+j}$ at $t > T^o$ from theorem II in appendix A. The left-hand side of eq. B12 may contain both the terms

$$k_{d+l,i}\frac{d}{dt}x_i \approx O(1)\sum_{l=0}^{g} x_{d+l} \tag{B13}$$

and

$$k_{d+l,j}\frac{d}{dt}x_j \approx -O(1)\sum_{l=0}^{g} x_{d+l} \tag{B14}$$

and eq. B12 may hold due to cancelling out between both terms. In this case, from the relation $(d/dt)x_j \gtrsim -O(1)\ x_j$ and theorem II in appendix A, the component $X_j$, on which eq. B14 holds satisfies

$$\frac{d}{dt}x_j \approx -O(1)x_j, \tag{B15}$$

and the order of magnitude of $x_j$ decreases within the time of O(1). Thus, the relation

$$\left|k_{d+l,j}\frac{d}{dt}x\right| \leq k_{d+l,j}x_j \leq O(\epsilon)\sum_{l=0}^{g} x_{d+l} \tag{B16}$$

holds within the time of O(1). At that time, the component $X_i$ does not satisfy eq. B13 because eq. B12 holds. Therefore, eq. B9 holds on every component neighboring the open group; i.e., eq. B9 is necessary for eqs. B10 and B11.

To hold the relation, eq. B9, continuously for $t > T'$ ($T' \lesssim$ O(1)) is sufficient in order that $T^o$ is of O(1) or less. This is shown as follows. If the following relation, eq. B17 holds at $t_1$ ($\lesssim$ O(1)), eq. B18 also holds within the time of O(1) because if eq. B18 does not hold, theorem II in appendix A does not hold.

$$\frac{d}{dt}\left(\sum_{l=0}^{g} x_{d+l}\right) \approx \pm O(1)\sum_{l=0}^{g} x_{d+l} \tag{B17}$$

$$\frac{d}{dt}x_{d+l} = \pm O(1)\sum_{l=0}^{g} x_{d+l} \quad (l=0,\ldots,g) \tag{B18}$$

Thus we obtain eq. B19 by introducing eqs. B9 and B18 into eq. B6,

$$\frac{d^2}{dt^2}\left(\sum_{l=0}^{g} x_{d+l}\right) = \mp O(1)\sum_{l=0}^{g} x_{d+l} = -O(1)\frac{d}{dt}\left(\sum_{l=0}^{g} x_{d+l}\right) \tag{B19}$$

because at least one of the coefficients ($\sum_{l=0}^{g} k_{d+l,d+j}$) is negative and its absolute value is of O(1) as G is an open group. Thus,

$$\left|\frac{d}{dt}\left(\sum_{l=0}^{g} x_{d+l}\right)\right| \le O(\epsilon)\sum_{l=0}^{g} x_{d+l} \tag{B20}$$

holds after a short period ($\lesssim O(1)$). It is stable because from eqs. B6, B9 and the above we obtain the following relation (eq. B21),

$$\left|\frac{d^2}{dt^2}\left(\sum_{l=0}^{g} x_{d+l}\right)\right| \le O(\epsilon)\sum_{l=0}^{g} x_{d+l}. \tag{B21}$$

Making use of the condition, eq. B9, we obtained method iii of section 2.2 in ref. 1 and determined $T^o$ in the examples given in the preceding paper [1].

## Appendix C

### *C1. Apparent rate constants of the reduced scheme when an open group was eliminated*

In this appendix, $g+1$ fast components belonging to an open group G are denoted by $X_d, \ldots, X_{d+g}$. The rate equations on the components in G and those on the components directly reacting with the components in G are given as,

$$\frac{d}{dt}\boldsymbol{x}_c = \boldsymbol{M}_c\boldsymbol{x}_c + \boldsymbol{D}_{cu}\boldsymbol{x}_u \tag{C1}$$

$$\frac{d}{dt}\boldsymbol{x}_u = \boldsymbol{D}_{uu}\boldsymbol{x}_u + \boldsymbol{D}_{uc}\boldsymbol{x}_c + \boldsymbol{D}_{uw}\boldsymbol{x}_w \tag{C2}$$

where $\boldsymbol{x}_c$, $\boldsymbol{x}_u$ and $\boldsymbol{x}_w$ are the vectors whose elements are molar fractions of $g+1$ fast components in G, those of $u$ components not belonging to G but directly reacting with the components in G and those of the rest of the components in the system respectively; i.e.,

$$\boldsymbol{x}_c = \begin{pmatrix} x_d \\ \vdots \\ x_{d+g} \end{pmatrix}, \quad \boldsymbol{x}_u = \begin{pmatrix} x_{v+1} \\ \vdots \\ x_{v+u} \end{pmatrix}, \tag{C3}$$

and $\boldsymbol{M}_c$, $\boldsymbol{D}_{cu}$, $\boldsymbol{D}_{uc}$ and $\boldsymbol{D}_{uw}$ are the matrices consisting of appropriate rate constants, $k_{ij}$. After an induction period $T^o$ the rate equation (eq. C1) can be set to be zero to a good approximation. Since $|\boldsymbol{M}_c| \approx O(1)$, $\boldsymbol{x}_c$ is given as,

$$\boldsymbol{x}_c = -\boldsymbol{M}_c^{-1}\boldsymbol{D}_{cu}\boldsymbol{x}_u \tag{C4}$$

By introducing the above into eq. C2, we obtain,

$$\boldsymbol{x}_u = (\boldsymbol{D}_{uu} - \boldsymbol{D}_{uc}\boldsymbol{M}_c^{-1}\boldsymbol{D}_{cu})\boldsymbol{x}_u + \boldsymbol{D}_{uw}\boldsymbol{x}_w \tag{C5}$$

This is the rate equation of the partially simplified scheme obtained by eliminating the fast components in the open group G. The matrix $-\boldsymbol{D}_{uc}\boldsymbol{M}_c^{-1}\boldsymbol{D}_c$ gives the apparent rate constants of the reaction through eliminated fast components in G in the reduced scheme.

*C2. The mass distribution at $T^o$ ($T^o \approx O(1)$) in the case when mass of O(1) exists in an open group G at $t = 0$*

Next we discuss the mass distribution at the end of the induction period $T^o$, when initial concentrations of some fast components in an open group G are of O(1) and those in the upper groups of G are of O($\epsilon$) or less. This gives the initial condition of the reaction in the reduced scheme after elimination of the fast components in G. The rapid change of the fast components in G is expressed by neglecting terms of O($\epsilon$) or less at $0 < t < T^o$ in the rate equation, eq. C1, as,

$$\frac{\mathrm{d}}{\mathrm{d}t}\boldsymbol{x}_c = \boldsymbol{M}_c\boldsymbol{x}_c$$

or,

$$\boldsymbol{x}_c = \boldsymbol{M}_c^{-1}\frac{\mathrm{d}}{\mathrm{d}t}\boldsymbol{x}_c. \tag{C4}$$

Integrating above from $t = 0$ to $t = T^o$ and taking into account that every $x_{d+l}(T^o)$ is of O($\epsilon$) or less, we have,

$$\int_0^{T^o}\boldsymbol{x}_c\mathrm{d}t = -\boldsymbol{M}_c^{-1}\boldsymbol{x}_c(0). \tag{C5}$$

The mass flow to the component $X_{v+i}$ ($i = 1,\ldots,u$) neighboring G, through $X_{d+l}$ during the period from $t = 0$ to $T^o$, is given as,

$$k_{v+i,d+l}\int_0^{T^o}x_{d+l}\mathrm{d}t \tag{C6}$$

where, $k_{v+i,d+l}$ is of O(1).

When $X_{v+i}$ is a slow component, the mass transferred to $X_{v+i}$ is all approximately reserved on $X_{v+i}$ at $t = T^o$ ($T^o \approx O(1)$) and $x_{v+i}(T^o)$ is obtained as,

$$\begin{aligned} x_{v+i}(T^o) &= \sum_{l=0}^{g} k_{v+i,d+l}\int_0^{T^o}x_{d+l}\mathrm{d}t \\ &= -\sum_{l=0}^{g} k_{v+i,d+l}\sum_{l'=0}^{g} c_{l,l'}x_{d+l'}(0) \end{aligned} \tag{C7}$$

where $c_{l,l'}$ is the $(l+1, v+1)$ element of $\boldsymbol{M}_c^{-1}$.

When $X_{v+i}$ is a fast component belonging to a closed group C, the mass transferred to $X_{v+i}$ distributes in C and attains quasi equilibrium within the time of O(1).

When $X_{v+i}$ is a fast component belonging to an open group F, the mass transferred to it is transferred to the components to which thick arrows is directed from the components belonging to F, within the time of O(1) and the above procedure is repeated again.

**Note**: Appendices A, B and C are cited in ref. 1 as appendices I, II and III, respectively.

## References

1 H. Kijima and S. Kijima, Biophys. Chem. 16 (1982) 181.
2 D.R. Cox and H.D. Miller, The theory of stochastic processes, ch. 4 (Chapman and Hall, London, 1965).
3 D. Colquhoun and A.G. Hawks, Proc. R. Soc. B199 (1977) 231.