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ASYMMETRY OF FLUCTUATION WITH RESPECT TO TIME REVERSAL IN STEADY STATES OF BIOLOGICAL SYSTEMS

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The asymmetry of fluctuation with respect to time reversal which is expected in an energy-consuming steady state is discussed with special attention to biological systems. The necessary condition for asymmetry of fluctuation of an observed quantity is given. To show the usefulness of the experimental analysis of asymmetry of fluctuation, some calculations are carried out on two simple examples of three-state reactions. In one of them, the two-point time correlation function of the observed quantity has an oscillatory component, while in the other the function is nearly exponential, but in both cases, the fluctuation has a pronounced asymmetry. A method to estimate the degree of asymmetry of fluctuation is proposed, and the application of the present method to investigation of the molecular events in biological systems such as muscle is discussed.

1. Introduction

This paper is concerned with the asymmetry of fluctuation with respect to time reversal, which is expected in an energy-consuming steady state. Let us consider two examples in biological systems.

A large fluctuation of the electric potential difference across the cell membrane has been often observed in living cells. Such a fluctuation is not always merely noise, but is utilized by the living cells for regulation of their behavior [1,2]. The tactic response of swimming *Paramecium* cells, for example, is performed by modification of spontaneous fluctuation of the membrane potential depending on the environmental conditions [3,4]. The fluctuations are amplified by a positive feedback mechanism between the thermal movement of ions around the membrane and the fluctuation of the state of macromolecules in the membrane which controls the permeability of ions [5,6]. The free energy stored in the unequal distribution of ions across the membrane is consumed for amplification of fluctuation.

In a recording of the membrane potential measured in a *Paramecium* cell, we can find the asymmetry of fluctuation with respect to time reversal [2]. This is evidence that an energy-consuming process is involved in production of the fluctuation. It is an interesting problem to investigate the relationship between the degree of asymmetry of fluctuation and the amount of energy consumed, and to characterize the mechanism of amplification of fluctuation.

Another example is taken from the problem of biological motility. In muscle contraction, an active tension is generated by a cyclic interaction of myosin and actin molecules coupled with the splitting of ATP. All reactions in the cycle are reversible and proceed as stochastic processes [7,8]. Actually, the active tension has been found to have a large fluctuation [9]. Does this fluctuation contain any asymmetric component with respect to time reversal? Is there any signal from myosin and actin molecules which shows an asymmetric fluctuation? It is useful to find such a signal to identify the molecular events involved in the process of tension

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generation. The asymmetry of fluctuation of the signal may be found not only in a muscle fiber but also in a solution of myosin and actin molecules.

In a droplet of protoplasm of *Amoeba*, for example, we can often observe apparently random movements of particulates. How is an active movement distinguishable from a simple thermal movement? Besides the magnitude of fluctuation, the possible asymmetry of fluctuation should be considered. How does asymmetry of fluctuation appear during the transition from random thermal movement to organized active movement?

General theories on the fluctuation in nonequilibrium steady states have been treated previously, and remarks have been made on the asymmetry of fluctuation [10,11]. The purpose of this paper is not to treat the general theory but to show some calculations on simple realistic examples which can be expressed in terms of chemical reactions, and to emphasize the usefulness of experimental analysis of the asymmetry of fluctuation.

Measurements of fluctuation of various quantities in biological systems have been made in many laboratories. The data obtained are usually directly transferred to a computer to derive the ordinary time correlation function or the power density spectrum. To make the best use of the data, however, the asymmetry of fluctuation must be analyzed before putting the data into a conventional computer system. The experimentally useful definition of the asymmetry of fluctuation can be made by matrix presentation which is introduced later.

2. Asymmetry of fluctuation in a steady state

In an equilibrium state, the thermal fluctuation of any quantity should be symmetric with respect to time reversal. In a nonequilibrium steady state, however, the fluctuation does not necessarily satisfy this condition. Consider a molecule which can be in one of the three states, 1, 2 and 3. In these states, the molecule takes three different values of an observable quantity, A, a_1, a_2 and a_3. Assume that the molecule undergoes stochastic transitions among these states. If only cyclic transitions from 1 to 2, from 2 to 3 and from 3 to 1 are allowed, the observed quantity A changes with time as shown in fig. 1, where the change has an asymmetry with respect to time reversal.

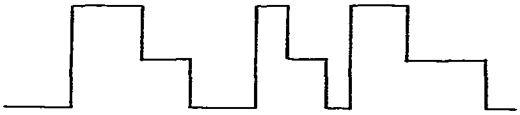

Fig. 1. Fluctuation of $A(t)$ of a single molecule.

Consider an assembly of N molecules, where each molecule undergoes the above cyclic transitions. The value of A of the assembly is given by

$$A(t) = n_1(t)a_1 + n_2(t)a_2 + n_3(t)a_3 \tag{1}$$

where n values are numbers of molecules in three states at time t. The value of $A(t)$ obtained by summation of the values taken by individual molecules should show an asymmetric fluctuation with respect to time reversal.

In the above example, the probability that a molecule is in a state i at t and is in other state j at $t+\tau$, $p(i,j;\tau)$, is not equal to the probability that a molecule is in the state j at t and is in the other state i at $t+\tau$, $p(j,i;\tau)$, for i and $j = 1$, 2 or 3 with $i \neq j$. In general, in a nonequilibrium steady state of an assembly of N molecules where each molecule can be in one of the states, $1, 2, \ldots, m$, the probability $p(i,j;\tau)$ is not necessarily equal to $p(j,i;\tau)$ for $i \neq j$. This inequality immediately results in the asymmetry of fluctuation of A if $a_i \neq a_j$. The asymmetry of fluctuation is due to the breaking of detailed balancing among different states.

It is obvious, however, that if each molecule in the assembly can be in only two states and make transitions independently, asymmetry of fluctuation cannot be produced. For example, if $a_2 = a_3$ in fig. 1, the asymmetry disappears. Even when each molecule can be in many different states, asymmetry of fluctuation cannot be produced, if the observed quantity can have only two different values. In other words, the observed quantity must distinguish three or more different states of the molecule to have asymmetry of fluctuation.

3. Simple examples

It is useful to give simple examples of a nonequilibrium steady state, where the probabilities, $p(i,j;\tau)$, can be easily calculated. In the cyclic reaction of fig. 1, let us denote the rate constants of transitions from 1 to 2, from 2 to 3 and from 3 to 1 as k_{+1}, k_{+2} and k_{+3}, respectively. In addition, we introduce reverse transitions from 2 to 1, from 3 to 2 and from 1 to 3 and denote their rate constants as k_{-1}, k_{-2} and k_{-3}, respectively.

3.1. Case 1

Let us assume that all rate constants of forward transitions and those of backward transitions are equal, respectively. That is, $k_{+1}=k_{+2}=k_{+3}(=k_+)$ and $k_{-1}=k_{-2}=k_{-3}(=k_-)$, as shown in figs. 2a. The average number of molecules in three states must be equal; i.e., $\langle n_1\rangle=\langle n_2\rangle=\langle n_3\rangle=(1/3)N$. Only when $k_+=k_-$ is an equilibrium state established. When $k_+\neq k_-$, a nonequilibrium steady state is established. The probability that a molecule is in a state i at t and is in a state j at $t+\tau$, $p(i,j;\tau)$, can be calculated, for $\tau\geqslant 0$, as:

$$p(1,1;\tau)=p(2,2;\tau)=p(3,3;\tau)$$
$$=(1/9)+(2/9)\exp(-(3/2)(1+\lambda)\tau)\cos((\sqrt{3}/2)(1-\lambda)\tau) \tag{2}$$

$$p(1,2;\tau)=p(2,3;\tau)=p(3,1;\tau)$$
$$=(1/9)-(1/9)\exp(-(3/2)(1+\lambda)\tau)\cos((\sqrt{3}/2)(1-\lambda)\tau)$$
$$+(1/3\sqrt{3})\exp(-(3/2)(1+\lambda)\tau)\sin((\sqrt{3}/2)(1-\lambda)\tau) \tag{3}$$

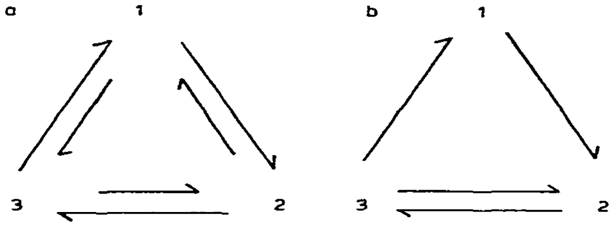

Fig. 2. (a) A three-state scheme where all reactions are reversible but rate constants of forward and backward reactions are different. (b) A three-state scheme where two reactions are unidirectional but the third one is bidirectional.

$$p(2,1;\tau)=p(3,2;\tau)=p(1,3;\tau)$$
$$=(1/9)-(1/9)\exp(-(3/2)(1+\lambda)\tau)\cos((\sqrt{3}/2)(1-\lambda)\tau)$$
$$-(1/3\sqrt{3})\exp(-(3/2)(1+\lambda)\tau)\sin((\sqrt{3}/2)(1-\lambda)\tau) \tag{4}$$

where λ is the ratio of two rate constants, k_-/k_+, and the time τ is the real time multiplied by k_+. If λ is not equal to unity, $p(i,j;\tau)$ is not equal to $p(j,i;\tau)$ for $i\neq j$. Therefore, asymmetry appears in the transition probabilities and in the fluctuation of $A(t)$ if $a_1\neq a_2\neq a_3$. The difference between $p(i,j;\tau)$ and $p(j,i;\tau)$ for $i\neq j$ has a maximum value around $\tau=0.4$ as a function of τ, where the asymmetry is most pronounced.

3.2. Case 2

Let us assume another condition, where $k_{+1}=k_{+2}=k_{+3}(=k_+)$ as before, but $k_{-1}=k_{-3}=0$ and $k_{-2}=k_{+2}(=k_+)$. The transitions from 1 to 2 and from 3 to 1 are unidirectional but the transition between 2 and 3 is reversible, as shown in fig. 2b. In the steady state, the average numbers of molecules in three states satisfy the condition $\langle n_1\rangle=\langle n_3\rangle=(1/2)\langle n_2\rangle=(1/4)N$. The probabilities are given, for $\tau>0$, by:

$$p(1,1;\tau)=(1/16)(1+(3+2\tau)\exp(-2\tau)) \tag{5}$$
$$p(2,2;\tau)=(1/4)(1+\exp(-2\tau)) \tag{6}$$
$$p(3,3;\tau)=(1/16)(1+(3-2\tau)\exp(-2\tau)) \tag{7}$$
$$p(1,2;\tau)=(1/8)(1-\exp(-2\tau)) \tag{8}$$
$$p(2,3;\tau)=(1/8)(1-(1-2\tau)\exp(-2\tau)) \tag{9}$$
$$p(3,1;\tau)=(1/16)(1-(1-2\tau)\exp(-2\tau)) \tag{10}$$
$$p(2,1;\tau)=(1/8)(1-(1+2\tau)\exp(-2\tau)) \tag{11}$$
$$p(3,2;\tau)=(1/8)(1-\exp(-2\tau)) \tag{12}$$
$$p(1,3;\tau)=(1/16)(1-(1+2\tau)\exp(-2\tau)) \tag{13}$$

The time τ is the real time multiplied by k_+. It is found that $p(i,j;\tau)\neq p(j,i;\tau)$ for $i\neq j$ and, therefore, asymmetry is expected in the fluctuation of $A(t)$. The difference between two probabilities has a maximum around $\tau=0.5$.

4. Time correlation functions

The ordinary time correlation function of $A(t)$,

$\phi_A(\tau)$, is defined by

$$\phi_A(\tau) = (1/T)\int_0^T A(t)A(t+\tau)\,dt$$
$$= \langle A(t)A(t+\tau)\rangle \qquad (14)$$

where the averaging is performed over a sufficiently long period, $0 < t < T$. This time correlation function does not retain any information about the asymmetry with respect to time reversal. Actually, $\phi_A(\tau)$ must be equal to $\phi_A(-\tau)$. In order to analyze the asymmetry, we have to have more information. For example, higher-order time correlation functions may be useful. Before introducing them, we calculate the ordinary time correlation function $\phi_A(\tau)$ for the above two cases. Eq. 14 is rewritten as:

$$\phi_A(\tau) = \sum_{i,j} N a_i a_j p(i,j;\tau) \qquad (15)$$

Therefore, in case 1, for $\tau > 0$,

$$\phi_A(\tau) - \phi_A(\infty) = (\phi_A(0) - \phi_A(\infty))\exp(-(3/2)(1+\lambda)\tau)$$
$$\times\cos((\surd 3/2)(1-\lambda)\tau) \qquad (16)$$

for all values of a. The time correlation function has an oscillatory component if λ is not equal to unity. In this example, the condition necessary for asymmetry of fluctuation with respect to time reversal is the same as that necessary for the contribution of an oscillatory component in the time correlation function. The presence of a cyclic process is apparent in the time correlation function.

In case 2, with $a_1 = 0$, $a_2 = 1$ and $a_3 = a$,

$$\phi_A(\tau) - \phi_A(\infty) = (\phi_A(0) - \phi_A(\infty))\big(1 + ((a - (a^2/2))/$$
$$(1 - a + (3/4)a^2))\tau\big)\exp(-2\tau) \qquad (17)$$

The time correlation function has no oscillatory component. This is a good example of where conventional computation of the time correlation function loses information about whether or not the observed quantity comes from a cyclic process in a steady state. The asymmetry of fluctuation with respect to time reversal is not always equivalent to the presence of an oscillatory component in the time correlation function. The condition necessary for the asymmetry is broader than that necessary for the oscillation, as long as the molecules give three or more different values of the observed quantity in different states. Nonequilibrium steady states in a network of chemical reactions contain cyclic processes in general. The cycling is not necessarily evident in the ordinary time correlation function.

One of the higher-order time correlation functions is defined by

$$\psi_A(\tau_1,\tau_2) = \langle A(t)A(t+\tau_1)A(t+\tau_1+\tau_2)\rangle \qquad (18)$$

In a nonequilibrium steady state,

$$\psi_A(\tau_1,\tau_2) \neq \psi_A(\tau_2,\tau_1) \quad \text{for } \tau_1 \neq \tau_2 \qquad (19)$$

In a special case where $\tau_1 = 0$,

$$\psi_A(0,\tau) - \psi_A(\tau,0) = \sum_{i,j} N\big(a_i^2 a_j - a_i a_j^2\big)p(i,j;\tau) \qquad (20)$$

In the scheme of fig 2a and b, with $a_1 = 0$, $a_2 = 1$ and $a_3 = a$, we find, for case 1,

$$\psi_A(0,\tau) - \psi_A(\tau,0) = Na(1-a)(2/3\surd 3)$$
$$\times\exp(-(3/2)(1+\lambda)\tau)\sin((\surd 3/2)(1-\lambda)\tau) \qquad (21)$$

and for case 2,

$$\psi_A(0,\tau) - \psi_A(\tau,0) = Na(1-a)(1/4)\tau\exp(-2\tau) \qquad (22)$$

The comparison of $\psi_A(0,\tau)$ and $\psi_A(\tau,0)$ is a simple way to find asymmetry of fluctuation with respect to time reversal.

It should be noted here that if in a nonequilibrium steady state, two different quantities $A(=\Sigma n_i a_i)$ and $B(=\Sigma n_i b_i)$ were measured in an assembly of N molecules, the time correlation function between them would be asymmetric with respect to time reversal; i.e. [10]:

$$\phi_{AB}(\tau) = \langle A(t)B(t+\tau)\rangle \neq \langle A(t+\tau)B(t)\rangle = \phi_{AB}(-\tau) \qquad (23)$$

The inequality comes from the asymmetry of the correlation function between the numbers of molecules in different states;

$$\langle n_i(t)n_j(t+\tau)\rangle \neq \langle n_i(t+\tau)n_j(t)\rangle \qquad (24)$$

In an equilibrium, these two correlation functions must be equal. The previous analysis simply means that information about the inequality of eq. 24 is preserved in the fluctuation of a single quantity $A(t)$, if a_i values are not equal.

5. Matrix presentation

In principle, all information about the fluctuation is contained in a series of time correlation functions of higher orders. The analysis of these functions, however, is not always practical. In some cases, it is rather easy to find asymmetry of fluctuation directly in the original data. For example, the spontaneous fluctuation of membrane potential in a *Paramecium* cell often shows a sharp rise and slow decay. Is there any simple manner to express such specific features of the observed fluctuation?

The following matrix presentation may be helpful. In a record of a fluctuating quantity, A, let us count the number of times of the event that at time t, A has a value A_i and at $t+\tau$, A has another value A_j, by scanning the record from $t=0$ to T with a fixed value of the time difference τ. Denoting this number as $M_{ij}(\tau) = M(A_i, A_j; \tau)$, we can compose a matrix of components $M_{ij}(\tau)$. In an equilibrium state, this matrix must be symmetric. In a nonequilibrium steady state, it may be asymmetric. The asymmetry of fluctuation with respect to time reversal can be defined by the asymmetry of this matrix.

Fig. 3 gives a graphic presentation of the matrix obtained by computer simulation for case 1. A small degree of asymmetry is noticeable around the peak along the diagonal. The asymmetry grows and decays with increasing time difference.

To make a matrix of suitable size, the observed value of $A(t)$ must be digitized with a limited resolution, and to obtain reliable values of components $M_{ij}(\tau)$, $A(t)$ must be measured for a sufficiently long period.

The ordinary time correlation function is calculated from the matrix as

$$\Phi_A(\tau) = \sum_{A_i > A_j} A_i A_j \left(M_{ij}(\tau) + M_{ji}(\tau) \right) / M_0 \tag{25}$$

where M_0 is a normalization factor arising from the total number of events. In contrast with this time correlation function, the degree of asymmetry of fluctuation may be estimated by the following correlation function obtained from the matrix;

$$\Phi_A^*(\tau) = \sum_{A_i > A_j} A_i A_j \left| M_{ij}(\tau) - M_{ji}(\tau) \right| / M_0 \tag{26}$$

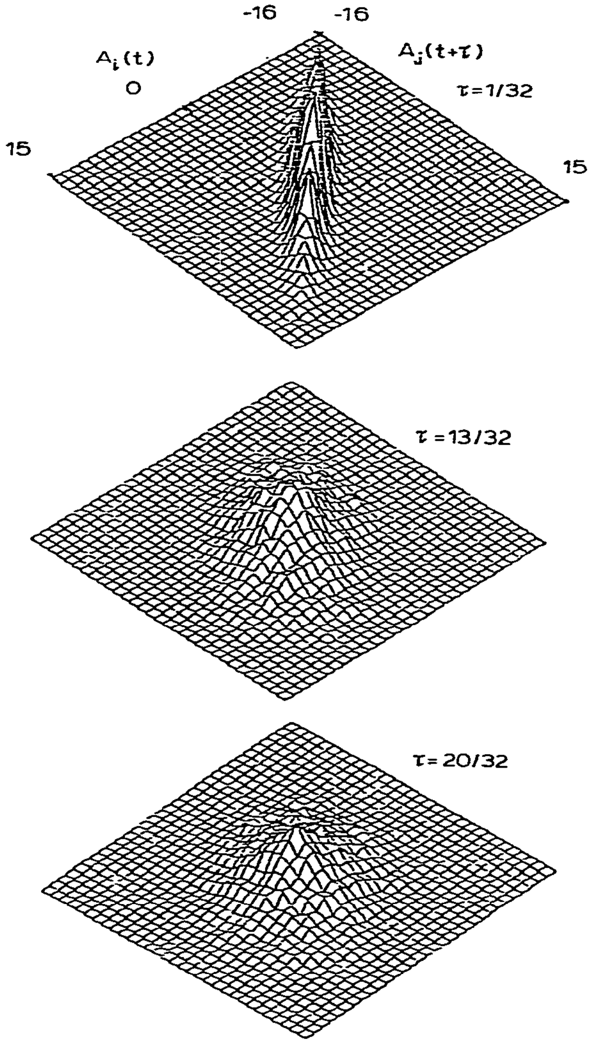

Fig. 3. Graphic presentation of the matrix M_{ij} obtained from $A(t)$ produced by computer simulation of the three-state scheme of fig. 2a. $M(A_i, A_j; \tau)$ is given by the height in a two-dimensional plane of A_i and A_j at three different values of the time difference $\tau = 1/32$, 13/32 and 20/32. $N = 900$, $a_1 = 0$, $a_2 = 1$ and $a_3 = 3$. Probabilistic transitions were made by computer with $k_+ = 1/32$ at every time interval $\Delta t = 1$, to produce $A(t)$ in a total period $T = 10^4$. The average of A must be 1200. The step of A to digitize the values of A_i and A_j, ΔA, was taken to be 1200/128.

The difference $\psi_A(0,\tau)-\psi_A(\tau,0)$ previously introduced is equivalent to the function;

$$\Phi_A(\tau)=\sum_{A_i>A_j} A_iA_j(A_i-A_j)(M_{ij}(\tau)-M_{ji}(\tau))/M_0 \quad (27)$$

6. Applications

Let us consider the three-state model which has been often employed to understand the molecular mechanism of muscle contraction [8,12,13]. Actin molecules in the thin filament and myosin molecules in the thick filament undergo a cyclic process consisting of three states. In state 1, two molecules are dissociated; in states 2 and 3, they are bound in different manners. Assuming an isometric condition where the distance between the centers of actin and myosin molecules along the filaments is fixed, the tension developed by the interaction of actin and myosin molecules is different in the two bound states 2 and 3. In these two states, the binding free energies between two molecules as a function of the distance along the filaments have minima at different positions [12,13]. The rate constants of transitions among three states depend on the environmental conditions, particularly on the concentrations of ATP and Ca^{2+}. Roughly speaking, however, the cycle has similar features to those depicted in fig. 2b. Namely, the transitions between 1 and 2 and between 3 and 1 are almost unidirectional and the transition between 2 and 3 is bidirectional. Therefore, as in the case of fig. 2b, the tension developed by a pair of actin and myosin molecules at a fixed distance is expected to show fluctuation asymmetric with respect to time reversal. The fluctuation of the tension developed by a large number of pairs of actin and myosin molecules in a muscle fiber must preserve the asymmetry.

Actually, the fluctuation of the tension of a muscle fiber was measured under an isometric condition [9]. Upon activation of the fiber, the fluctuation became very large, where its time correlation function was apparently exponential. It is desirable to examine whether or not fluctuation of the tension contains a component asymmetric with respect to time reversal. The simplest way is to calculate the functions $\psi_A(\tau_1,\tau_2)$ and $\psi_A(\tau_2,\tau_1)$. The inequality of these functions, if it were found, would give support for the observed fluctuation coming from an active process to which the three-state model may be applied. The matrix of $M_{ij}(\tau)$, if obtained, would be very useful.

A large fluctuation in light scattering was found in an activated muscle fiber. Its time correlation function had some deviation from a simple exponential type [14], and a possible structural origin was discussed [15]. The fluctuation of the fluorescence intensity from labelled myosin molecules in a muscle fiber was also measured [16]. It was large during development of active tension and had a correlation time of the same order as that of the tension. If the fluorescence intensity is different between two bound states of myosin and actin molecules, it is very probable that its fluctuation has a component asymmetric with respect to time reversal. Then, the fluorescence intensity gives valuable information about the cyclic interaction of myosin and actin molecules coupled with the splitting of ATP.

Similar measurements can be made in a solution of myosin and actin molecules. In the presence of ATP, the solution was found to give a large fluctuation of light scattering having a long correlation time [17]. If the scattering intensity changes simply with binding and detachment of myosin and actin molecules, its fluctuation cannot have asymmetry. If, however, the scattering intensity contains some contribution from active movements of myosin and actin molecules, the fluctuation may have asymmetry. Recently, active movements of the filaments of actin and myosin have been realized in a solution with ATP [18,19]. It is hoped to investigate the fluctuation of fluorescence and light scattering under the same condition.

The bacterial flagellar motor is rotated by a flow of H^+ due to the electrochemical potential difference across the membrane. The threshold of the potential difference of H^+ for driving the motor has been found to be very small, in the range from 30 to 8 mV [20,21]. Then, the free energy released by an H^+ flowing across the membrane is of the same order as the energy of thermal fluctuation. Accordingly, it is interesting to measure the fluctuation in the speed and torque of the motor near the threshold of the potential difference [20]. The asymmetry of fluctuation, if it were found, would give a clue to understanding the mechanism of rotation of the motor [22].

The other example is the fluctuation of the membrane potential. Channel molecules in the membrane usually have three states, a closed state, an opened state and an inactive or desentisized state [23]. All transitions among these states are reversible and the rate constants depend on the membrane potential. Under a fixed membrane molecules is usually symmetric and, thus, fluctuation of the membrane current is symmetric [24]. However, if the membrane potential is free to fluctuate, fluctuation of the state of channel molecules takes place closely correlated with fluctuation of the membrane potential. In the case of *Paramecium* cells, such a correlation gives rise to cooperative fluctuation of a number of channel molecules and, consequently, fluctuation of the membrane potential is very much amplified [2]. The amplification may be associated with a flow of free energy carried by the ion current across the membrane. Actually, fluctuation of the membrane potential of *Paramecium* cells consists of two components, a basic fluctuation and a spike-like fluctuation triggered by the basic one [2]. The asymmetry of fluctuation is more pronounced in the latter. In such a case, the matrix presentation of M is useful to see where the asymmetry is produced.

7. Remarks

A few remarks must be added. It was pointed out that asymmetry of fluctuation of an observed quantity $A(t)$ in a nonequilibrium steady state of an assembly of N molecules (molecule X) is expected if each molecule can assume three or more different states where it has different values of A. In such an assembly let us put the other kind of molecules (molecule Y) which have some interaction with the assembly of X. Assume that these molecules Y make transitions among different states where an observable quantity B takes different values. Because of the interaction, the rate constants of transitions of Y among different states may depend on the state of the assembly of X, or the value of $A(t)$. Then, the fluctuation of $B(t)$ may have an asymmetric component, if the fluctuation of $A(t)$ is asymmetric. If the fluctuation of $B(t)$ is really asymmetric, it means that there is a flow of free energy between two assemblies of X and Y. In this case, asymmetry of fluctuation of $B(t)$ may appear even if molecules Y can take only two different values of B. This kind of problem may be formulated by treating a reaction scheme in which the rate constants are not constant but have fluctuations due to fluctuation in the environment [5].

Finally, it must be noted that in several papers, the higher-order time correlation functions or the multispectral densities were already studied, although the asymmetry with respect to time reversal was not always noticed [25]. The third-order correlation function of eq. 20 was discussed in relation to the problem of the $1/f$ noise [26]. It may be interesting to investigate the possible asymmetry in the $1/f$ noise. Similar problems have been often treated also in the field of mathematics of stochastic processes. The high-order time correlation functions give important information in nonlinear systems.

Experiments along the line discussed in this paper are now in progress. The results will be reported in another paper.

References

1 W. Moolenaar, J. DeGoede and A. Verveen, Nature 260 (1976) 344.
2 T. Majima, Biophys. Chem. 11 (1980) 101.
3 Y. Naitoh and R. Eckert, Experiments in physiology and biochemistry vol. 5 (Academic Press, New York, 1975).
4 F. Oosawa and Y. Nakaoka, J. Theor. Biol. 66 (1977) 747.
5 F. Oosawa, J. Theor. Biol. 52 (1975) 175.
6 F. Oosawa, Ann. N. Y. Acad. Sci. 303 (1977) 38.
7 A.F. Huxley, Prog. Biophys. 7 (1957) 255.
8 A.F. Huxley and R. Simmons, Nature 233 (1971) 533.
9 J. Borejdo & M. Morales, Biophys. J. 20 (1977) 315.
10 Y. Chen, Adv. Chem. Phys. 37 (1978) 67.
11 K. Tomita and H. Tomita, Prog. Theor. Phys. 51 (1974) 1731.
12 T.L. Hill, Prog. Biophys. Mol. Biol. 29 (1975) 107.
13 H. Shimizu, Adv. Biophys. 13 (1979) 195.

14 F. Carlson, R. Bonner & A. Fraser, Cold Spring Harbor Symp. Quant. Biol. 37 (1973) 389.
15 S. Fujime and S. Yoshino, Biophys. Chem. 8 (1978) 305.
16 J. Borejdo, S. Putnam and M. Morales, Proc. Natl. Acad. Sci. U.S.A. 76 (1979) 6446.
17 F. Oosawa, S. Fujime, S. Ishiwata and K. Mihashi, Cold Spring Harbor Symp. Quant. Biol. 37 (1973) 277.
18 S. Higashi-Fujime, J. Cell Biol. 87 (1980) 569.
19 S. Higashi-Fujime, Cold Spring Harbor Symp. Quant. Biol. 46 (1982) in the press.
20 M. Manson, P. Tedesco and H. Berg, J. Mol. Biol. 138 (1980) 541.
21 J. Shioi, S. Matsuura and Y. Imae, J. Bacteriol. 144 (1980) 891.
22 F. Oosawa and J. Masai, J. Phys. Soc. Jap. 51 (1982) 631.
23 T. Heidmann and J.-P. Changeux, Eur. J. Biochem. 94 (1979) 255.
24 E. Neher and C. Stevens, Annu. Rev. Biophys. Bioeng. 6 (1977) 345.
25 G. Phillies, J. Chem. Phys. 72 (1980) 6123.
26 M. Nelkin and A. Tremblag, J. Stat. Phys. 25 (1981) 253.