Environment-Dependent Self-Organization of Positional Information Field in Chemotaxis of *Physarum* Plasmodium

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Physarum plasmodium shows highly coordinated tactic migration accompanied by a reorganization of its body shape, even in a complicated environment. We experimentally investigated the information integration mechanism to achieve such environment-dependent coordination from the viewpoint of self-organization of the positional information field. Since the organism is known to process information by mutual entrainment in distributed intracellular oscillators, we analysed the relationship between the global phase wave and migration velocity in the tactic response. It was thus clarified that the phase gradient in the wave linearly decreases according to the relative distance from the wave source corresponding to the best-conditioned site, and the migration velocity in each part was regulated by the phase gradient in position-dependent manner. These results indicate that the organized phase gradient pattern represents the environment-dependent positional information field which encodes the relative positional relationship between each part and the best-conditioned site in the whole system. Therefore, through the self-referential interaction process between self-organization of this information field and its interpretation, environmental information is integrated, and tactic migration towards the most suitable environment can be coordinately and relevantly controlled.

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1. Introduction

The *Physarum* plasmodium is a large amoeboid cell. Although it has no nervous system, highly coordinated tactic behavior accompanied by a global reorganization of its body shape is observed even in a complicated environment, as shown in Fig. 1. The important point is that the tactic pattern is not fixed, but changes flexibly according to the environmental conditions. How does the organism integrate infor-

mation to achieve such an environment-dependent global coordination?

In this study, we experimentally investigated the information integration mechanism from the viewpoint of self-organization of the positional information field. This concept, which provides a kind of space coordinate indicating positional relationships relative to the whole system, was first proposed by Wolpert (1969). It has since been mainly applied to insulated systems such as morphogenesis in the developmental process (Gierer & Meinhardt, 1972; Kauffman et al., 1978; Arcuri & Murray, 1986; Hunding et al., 1990), the environmental effects have not been sufficiently investigated. The tactic pattern formation is thought to necessitate both robustness and flexibility against the environment,

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because the changing environment requires the organism to generate a relevant and environment-dependent function. Thus the traditional concept could not be directly applied to the present problem. Meinhardt & Gierer (1974) showed that once a stationery, polarized pattern is established, alteration of the internal polarity upon a change in the external condition takes a long time, and they suggested the possibility that the periodic change of polarity permits the system to adapt more readily to the changing environment. These characteristics strongly suggest that the oscillation phenomena are important in tactic pattern formation.

In the taxis of *Physarum* plasmodium, it is widely suggested that mutual entrainment between distributed intracellular chemical oscillators plays an important role as a communication mechanism. Tension oscillations with a period of about 2 min are observed in the organism (Kamiya, 1970; Wohlfarth-Bottermann, 1975), and their local oscillations are spatially synchronized with each other (Yoshimoto & Kamiya, 1978; Grebecki & Cieslawska, 1978). Tension oscillation has a fixed phase relation with chemical oscillations such as Ca^{2+} (Yoshimoto *et al.*, 1981a; Kuroda *et al.*, 1988), ATP (Yoshimoto *et al.*, 1981b), and H^+ (Nakamura *et al.*, 1982). Attractive stimuli increase the frequency of tension oscillation, while repulsive stimuli decrease it (Durham & Ridgway, 1976; Miyake *et al.*, 1994b). Such a local frequency modulation generates a phase shift in the neighbouring region (Achenbach & Wohlfarth-Bottermann, 1980; Wohlfarth-Bottermann & Block,

1981), and the global phase wave propagating between the modulated site and the other regions is also organized (Hejnowicz & Wohlfarth-Bottermann, 1980; Matsumoto *et al.*, 1986; Tanaka *et al.*, 1987). In such a case, by artificially modulating the local frequency, the migratory direction was clarified to be controlled by the direction of phase waves (Matsumoto *et al.*, 1988).

We further investigated the relationship between mutual entrainment and information integration in the intracellular oscillator system. When spatial interaction between phase waves was inhibited, coordinated migration as a whole disappeared (Miyake *et al.*, 1991). In addition, it was clarified that such interaction is mediated by cytosolic Ca^{2+} oscillation (Miyake *et al.*, 1992; Natsume *et al.*, 1992). These two findings indicate that the global phase wave organized in the chemical oscillator system is essential to intracellular communication for coordinated migration. Thus, we proposed the diffusion-coupled nonlinear oscillator model, and suggested that information integration is based on the self-organization of the positional information field (Miyake *et al.*, 1993). However, how the integrated information is organized and encoded in the real organism is still obscure. Therefore, by analysing the spatio-temporal order of intracellular oscillation and migration velocity in the tactic response, and by artificially modulating the spatial relationship of phase and period, the information integration mechanism of such an environment-dependent coordination is clarified.
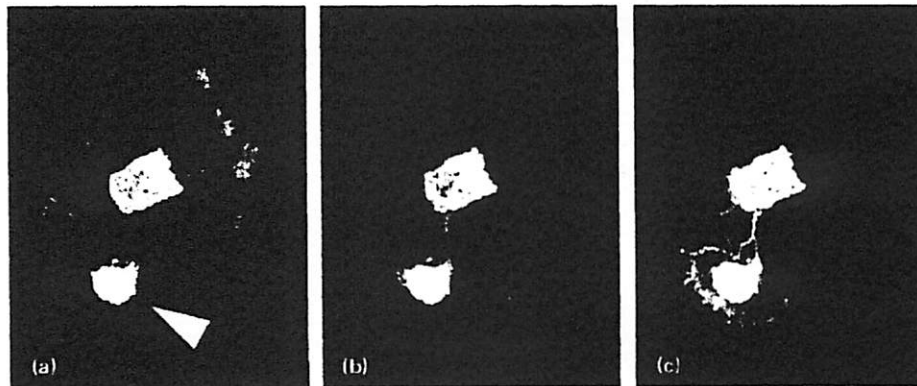

FIG. 1. Tactic pattern formation of *Physarum* plasmodium. The organism is a large amoeboid cell, and it shows highly coordinated tactic behavior accompanied by a reorganization of its body shape. (a) The organism migrates in the direction of the top of its fan-shaped body. An environmental stimulus (attractant) was applied to the rear part indicated by an arrowhead. (b) It started to change its body shape according to the stimulus. (c) Finally, reorganization of its body shape was completed and its polarity was reversed as a whole. The interval between photographs is 1 h. Bar: 5 cm.

2. Materials and Methods

2.1. ORGANISM

Physarum plasmodium was allowed to migrate on a 1.5% agar gel sheet overnight without feeding at room temperature (20–23°C) before use. A sheet of the plasmodium was carefully excised from the tip portion of the migrating plasmodium and used in the experiments.

2.2. IMAGE ACQUISITION AND ANALYSIS

By successively processing optical images of the plasmodium, oscillation and migration were measured without direct contact. The entire organism was illuminated homogeneously by diffuse white light of about 1000 lux from above, and its reflected light images were continuously taken by a video camera (*GR-60*, Victor). The red component of the image was digitized (*MT98-CVFM*01, Microtechnica) into 256 levels in each pixel and transmitted to a personal computer (*PC-98RX*, *NEC*). Data were acquired every 2 s. The digitized image of the organism was divided into square units, and the reflected light intensity from each unit area was calculated as a mean of all the pixels in it. To exclude pixels not corresponding to a part of the organism, pixels with intensity below a fixed threshold value were eliminated. Furthermore, cytoplasm thickness in each unit area was measured using a photointerrupter (*GP2L02*, Sharp), and the reflected light intensity was calibrated to be proportional to the thickness. Since change in thickness is thought to represent mechanochemical processes in the organism, the observed oscillation can be regarded as the intracellular chemical rhythm.

Temporal development of the reflected light intensity was separated into oscillation and offset components. The oscillation component was defined as the difference between the reflected light intensity and the offset component, as in eqn (1), and was regarded as the thickness oscillation in each unit area. The offset component was calculated by averaging the reflected light intensity, as shown in eqn (2), and was regarded as the cytoplasm distribution in the same unit area.

Thickness oscillation:

$$Th\ (x, t) = F(x, t) - \frac{1}{T}\int_{t-T/2}^{t+T/2} F(x, s)\,ds,\qquad (1)$$

Cytoplasm distribution:

$$Cy\ (x, t) = \frac{1}{T}\int_{t-T/2}^{t+T/2} F(x, s)\,ds,\qquad (2)$$

where $F(x, t)$ is the reflected light intensity at position x and time t, and T is 256 s.

The thickness oscillation was decomposed into the period and phase gradient. The period was defined as the time interval between two successive minimum peaks in the same oscillation. The phase gradient was defined as shown in eqn (3), and it was calculated approximately as the phase difference between two neighbouring unit areas by dividing the time interval between the corresponding maximum peaks from the neighbouring unit areas by the mean period between them. On the other hand, the cytoplasm distribution was analysed in terms of migration velocity. Since cytoplasm transport under the present experimental conditions can be regarded as the flow of a non-compressive fluid in a one-dimensional channel, the migration velocity was estimated as the velocity of cytoplasm transport using Laplace's equation, as shown in eqn (4). For simplicity, it was calculated as the relative velocity normalized by the total amount of cytoplasm distribution.

Phase gradient:

$$Pg\ (x, t) = -\frac{\partial Ph(r, t)}{\partial r}\bigg|_{r=x},\qquad (3)$$

Migration velocity:

$$Mv\ (x, t) = \int_0^x \left\{\frac{\partial Cy(r, s)}{\partial t}\bigg|_{s=t}\right\}dr,\qquad (4)$$

where $Ph(x, t)$ and $Cy(x, t)$ are the phase and the cytoplasm distribution, respectively.

2.3. ENVIRONMENTAL CHEMICAL STIMULATION

The plasmodium (80 × 5 mm or 50 × 5 mm) was transferred to a cellophane sheet placed on a 1.5% agar gel plate about 5 mm thick. About 2 hr after this procedure, the plasmodium was chemically stimulated. By carefully sliding the cellophane sheet on which the plasmodium was laid, one end of the organism was placed on another agar plate containing an attractant (20 mM galactose). The stimulated area was about 10 mm in length, as shown in Fig. 2(a). When two stimuli were applied, strong attractant (20 mM galactose) and weak attractant (0.5 mM galactose) were exposed to each end. A piece of thin film (Parafilm, American National Can) was inserted between these agar plates to prevent diffusion of stimulants. The specimen was always kept in humid air in a container at 20°C. Owing to the shape of the plasmodium, the size of the unit area for image analysis was fixed at about 5 mm in the longitudinal
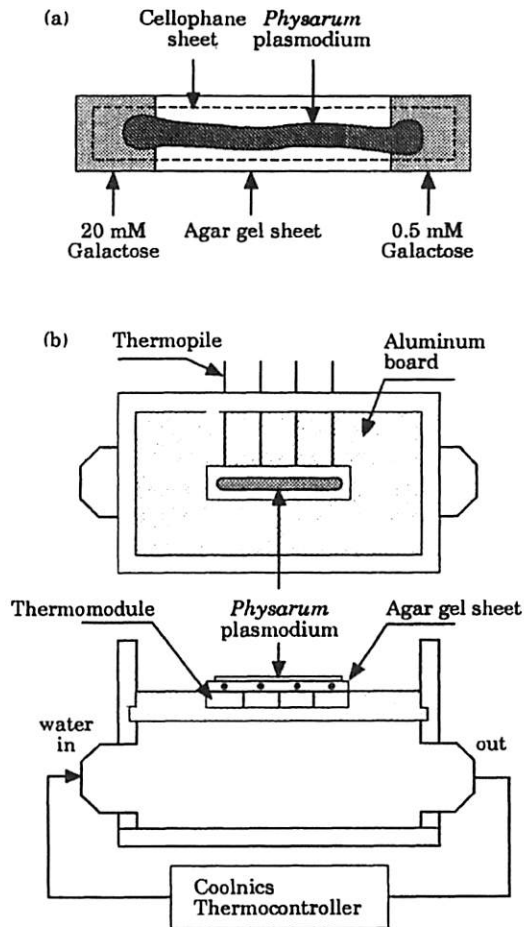
(a)   Cellophane *Physarum*
          sheet   plasmodium

20 mM   Agar gel sheet   0.5 mM
Galactose   Galactose

(b)   Thermopile   Aluminum
                              board

Thermomodule   *Physarum*   Agar gel sheet
                plasmodium

water
in   out

Coolnics
Thermocontroller

FIG. 2. Schematic illustrations of experimental setups. (a) Environmental chemical stimulation. The organism is exposed to chemical stimuli at both ends: one is 20 mM galactose and the other is 0.5 mM galactose. When the stimulus is applied at one end, stimulation with 0.5 mM galactose is not used. (b) Modification of intracellular oscillation. It is achieved by applying temperature oscillation to the organism, wherein the temperature is automatically controlled using a thermomodule and thermopile

direction of the organism and 8 mm width in the radial direction.

2.4. MODIFICATION OF INTRACELLULAR OSCILLATION

As it has been reported that intracellular oscillation in the plasmodium is entrained into the temperature oscillation (Kawato, 1984; Matsumoto et al., 1988), we also used temperature to control the intracellular oscillator. The temperature was regulated using both a thermomodule (KSM-0131, Komatsu Electronics) and thermopile (T-316, Nimblox). The thermomodule (8.8 × 8.8 mm) is an electrical device used to control heat production and absorption using the Peltier effect. The plasmodium (4 × 3 mm or 30 × 3 mm) was laid on an agar gel sheet (35 × 8.8 mm) about 1.5 mm thick placed on four thermomodules, as

shown in Fig. 2(b). Four thermopiles 1 mm diameter were horizontally inserted into the agar gel sheet, corresponding to each thermomodule. To remove excess heat production from the module, 20°C water was circulated (CTE-42WS, Komatsu Electronics) under the aluminum board in which the modules were embedded. By using the PID control method in a personal computer (PC-98DX, NEC), a sinusoidal wave of $20 \pm 6°C$ was automatically generated and applied to the plasmodium. The amplitude was constant, and the spatial relationship of phase and that of period were modulated by independently controlling the four thermomodules. The modules were called regions 1, 2, 3 and 4 from the left. In this case, owing to the shape of the thermomodule, the size of the unit area for image analysis was fixed at about 2.2 mm in the longitudinal direction and 8 mm width in the radial direction.

3. Results

3.1. SELF-ORGANIZATION OF PHASE GRADIENT PATTERN

3.1.1. Temporal development in single cluster

First, information processing in the plasmodium after environmental stimulation at one site was investigated as the simplest case. Then, spatio-temporal development of thickness oscillation and cytoplasm distribution after attractive stimulation (20 mM galactose) at one end of the organism was measured. In the result shown in Fig. 3(a), the global phase wave was organized by mutual entrainment in thickness oscillation, and it propagated from the stimulated site (upper side) to the opposite end (lower side). In addition, as shown in Fig. 3(b), cytoplasm distribution increased at the stimulated site and decreased in other parts, indicating that the organism migrates toward the stimulated side as one whole body. In this process, the period was almost uniform within most parts of the system, as shown in Fig. 3(c). The phase gradient and the migration velocity were in the same direction, and the migration direction coincided with the phase advance, as shown in Figs 3(d) and 3(e). These results mean that one coherent cluster was self-organized.

To analyse this coherent spatio-temporal order, results obtained under the same experimental conditions were averaged. As shown in Fig. 4(a), the entrained period is almost the same in every part of the system. The phase gradient gradually decreases at each part depending on the distance from the wave source, which corresponds to the stimulated site, and the migration velocity changes similarly to this phase gradient pattern, as shown in Figs 4(b) and 4(c).

These results suggest that the phase gradient pattern can provide a kind of space coordinate representing the positional relationship between the stimulated site and the other regions in the whole system. Thus, the migration velocity at each part is thought to be determined depending on the phase gradient, that is that the region near the stimulated site migrates faster than that far from the stimulated site. This may be
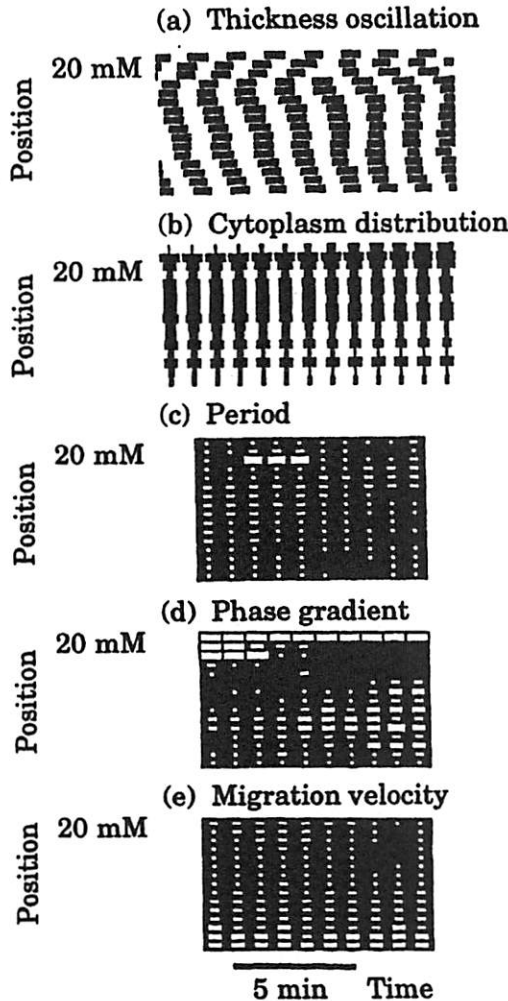
(a) Thickness oscillation

(b) Cytoplasm distribution

(c) Period

(d) Phase gradient

(e) Migration velocity

5 min   Time

FIG. 3. Spatio-temporal development of intracellular oscillation and migration after stimulation at one end of the plasmodium. Ordinate shows position and each unit area corresponds to 5 mm. Abscissa indicates time from 4 min after stimulation. (a) Thickness oscillation. Black and white parts represent positive and negative values of oscillation, respectively. Phase wave can be seen as the spatially continuous regions of the black and white parts. (b) Cytoplasm distribution. It is shown for 1 min intervals. The extent of the black area in each unit indicates the degree of distribution. (c) Period. Greater extent of the black area in each unit corresponds to shorter period. (d) Phase gradient. Greater extent of the black area in each unit represents larger gradient on the 20-mM-side, and greater extent of the white area, on the 0.5-mM-side. (e) Migration velocity. Representation is the same as in (d).
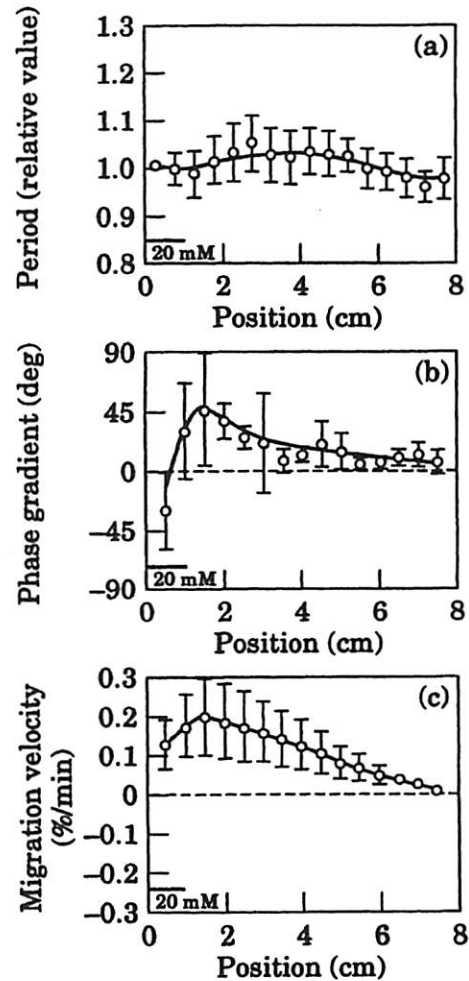
FIG. 4. Spatial pattern of period, phase gradient and migration velocity under stimulation at one end. Data were averaged over 10 min and further averaged among many samples (n = 5). In each figure, abscissa indicates the position where left end corresponds to the stimulated site. (a) Period. It is normalized by the value at the left end. (b) Phase gradient. It is calculated as the phase difference between neighbouring unit areas. Positive value means the direction to the stimulated side, and vice versa. (c) Migration velocity. It is calculated as the velocity of cytoplasmic transport normalized by the total amount of cytoplasm. Expressions are the same as for the phase gradient. Bar: S.D.

very convenient for reorganizing the fan-shaped body in the tactic response of the plasmodium.

If this phase gradient pattern encodes the positional information field, it could represent the relative positional relationship between each part and the whole system. Thus, the relationship between the system size and the organized phase gradient pattern was further investigated for another size of the plasmodium (50 × 5 mm) under the same experimental conditions. As shown in Fig. 5, the phase gradient

linearly decreases depending on the distance from the stimulated site, while the phase gradients at the two ends are constant and independent of the system size. It is, therefore, suggested that the phase gradient in each part is scaled relative to the system size. Because pf this characteristic, it can represent the relative positional relationship between the stimulated site and other regions in the whole system. This means that the organized phase gradient pattern can encode the positional information field, at least in a single cluster.

3.1.2. Temporal development between two clusters

Next, as the simplest case of information integration in a complicated environment, the integration process after environmental stimulation at both ends of the organism was investigated. One stimulant was 20 mM galactose and the other was 0.5 mM galactose. As shown in Figs 6(a) and 6(b), two phase waves propagating from the two ends were observed, and the boundary layer between the two wavefronts gradually moved toward the 0.5-mM-side. Finally, only the wave from the 20 mM side entrained the whole system, and the cytoplasm transport changed in a similar manner to this phase wave. As shown in Figs 6(c–e), two clusters having different periods were observed in the first stage (a), in which the polarities of the phase gradient and migration velocity were opposite between the two clusters. In the next stage (b), the cluster on the 20 mM side gradually enlarged until it finally became dominant.

Figures 7(a–c) show the statistical results obtained from stage (a). It is clear that the period on the 20 mM side cluster is shorter than that on the 0.5 mM side cluster, indicating that a more suitable environ-

ment induces a shorter period. Corresponding to this period difference, the phase gradient and the migration velocity become positive on the 20 mM side and negative on the 0.5 mM side. In stage (b), as shown in Figs 7(d–f), the period distribution is uniform in most of the system. The phase gradient and the migration velocity become positive in most part, and they gradually decrease with increasing distance from the 20 mM side. These characteristics are similar to those in the case of Figs 4(a–c). Since 20 mM galactose is a stronger attractant than 0.5 mM galactose, these results suggest that the cluster with shorter period, representative of the more suitable environmental condition, entrains the other clusters. Thus, the phase gradient pattern is thought to represent the positional relationship between the best-conditioned site and the other regions in the whole system. This means, therefore, that the phase gradient pattern can encode the environment-dependent positional information field around the best conditioned site even in a complicated environment.

3.2. MODIFICATION OF PHASE GRADIENT PATTERN

3.2.1. Causal relation between phase gradient and migration

The causal relation between phase gradient pattern and migratory response was investigated. In this experiment, the spatial gradient in a small organism (4 × 3 mm) was regulated by applying temperature oscillations at two different regions, and thickness oscillation and cytoplasm distribution were measured under this condition. A result for the case of 90° phase difference in temperature oscillation between neighbouring regions is shown in Figs 8(a–c). In this case, thickness oscillation was stably entrained into the temperature oscillation, and the cytoplasm distribution increased in region 1 and decreased in region 2. This means that the organism migrated toward the phase-advanced side as a whole body.

Furthermore, results obtained under various phase differences of temperature oscillation were analysed, and the relationship between the migration velocity and the phase gradient of thickness oscillation is plotted in Fig. 9. The migration velocity has a biphasic relationship with the phase gradient, and the velocity is maximum at about 90° phase gradient and minimum at both 0 and 180°. The direction of migration always, therefore, coincides with that of phase advance of the thickness oscillation. These results indicate that the migration velocity is directly controlled by the phase gradient in the intracellular oscillator. Therefore, it was clarified that the coherent phase gradient pattern encodes the positional
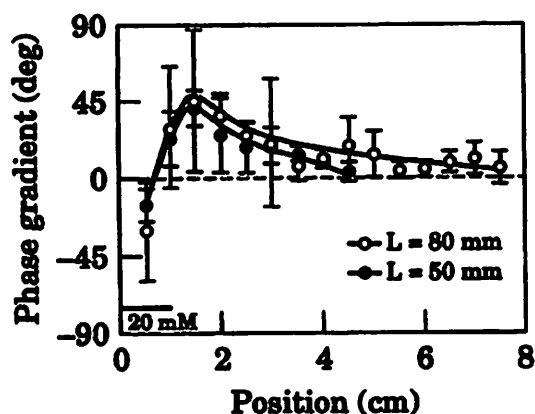

FIG. 5. Spatial pattern of phase gradient for different sizes of the organism. Data were averaged over 10 min and further averaged among many samples (n = 5). Phase gradient pattern shown in Fig. 4b for 80 mm organism is superimposed. Bar: S.D.
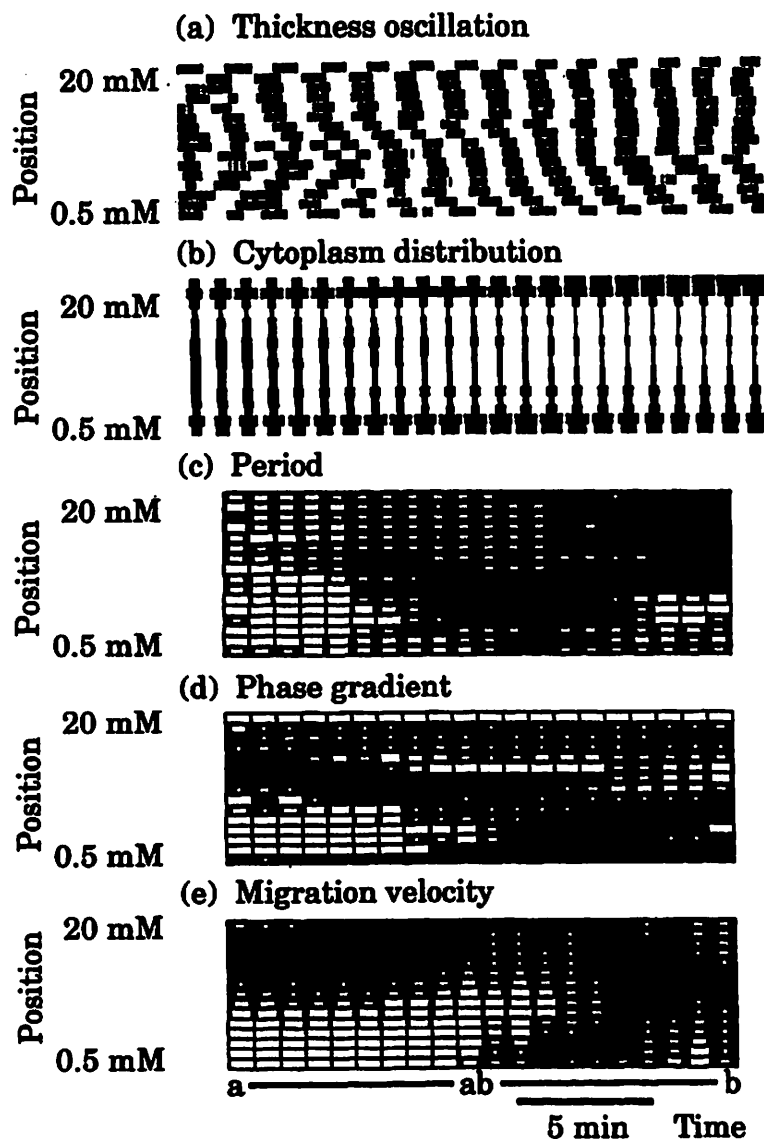
Fig. 6. Spatiotemporal development of intracellular oscillation and migration after stimulation at both ends of the plasmodium. Horizontal lines at bottom indicate stage a and stage b. Other details are the same as for Fig. 3. (a) Thickness oscillation. (b) Cytoplasm distribution. (c) Period. (d) Phase gradient. (e) Migration velocity.

information field which regulates the tactic migration and pattern formation.

3.2.2. Causal relation between original period and phase gradient

The causal relation between the original period and the phase gradient pattern was clarified. In this experiment, the spatial period difference in the organism (30 × 3 mm) was regulated by applying temperature oscillations at four different regions. Regions 1 and 2 had one period, and regions 3 and 4 another. The phase of region 1 was advanced 90°

from that of region 2, and the phase of region 4 was similarly advanced from that of region 3. Two clusters having opposite phase gradients were, therefore, artificially organized. Under this condition, the period difference between the two clusters was modified.

When the periods of temperature oscillations in regions 1, 2 and regions 3, 4 were fixed at 120 s and 140 s, respectively, the phase wave with the shorter period propagated a longer distance than that with the longer period, as shown in Figs 10(a–c). Thus, the thickness oscillation in region 3 was entrained into the
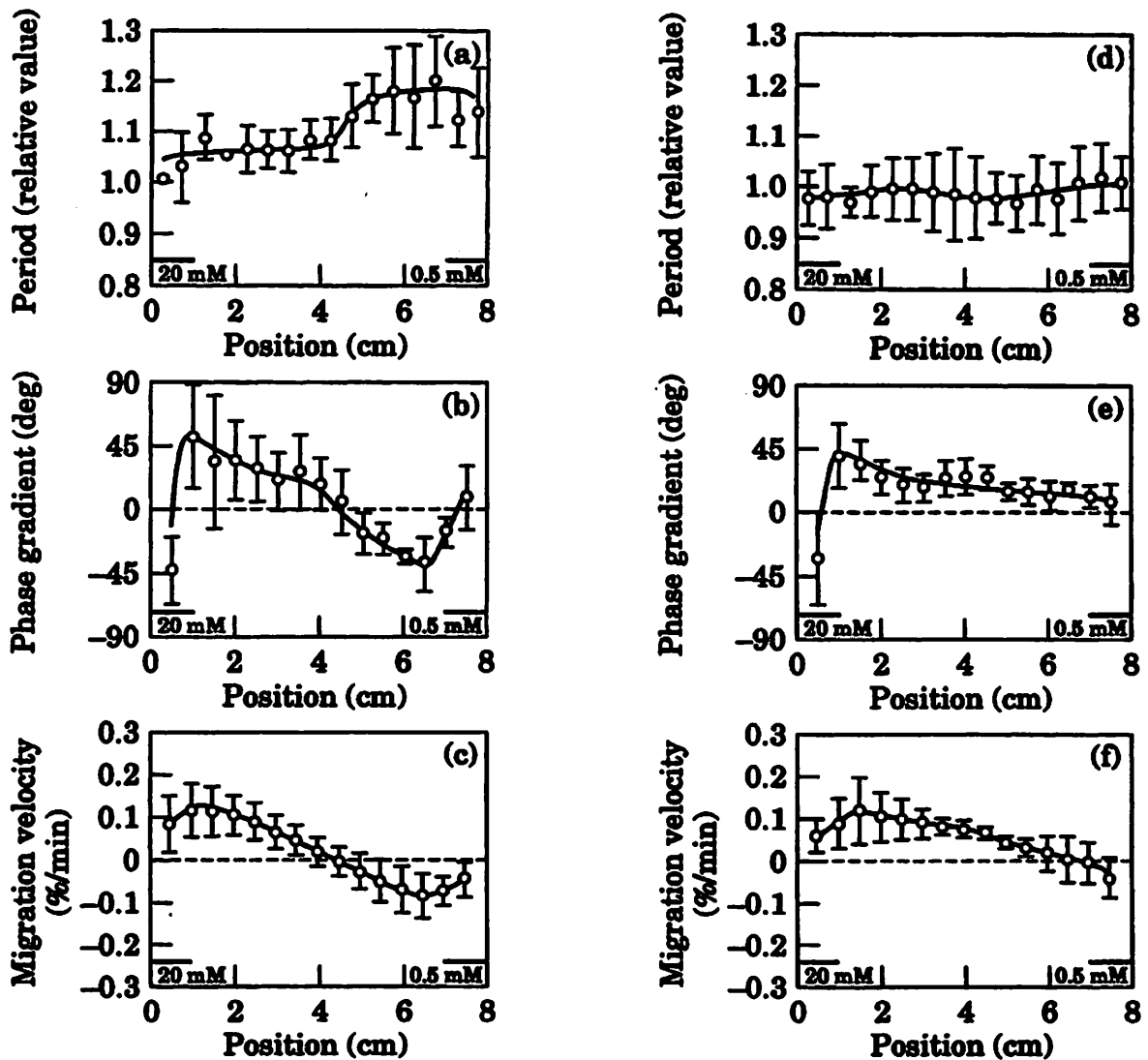
Fig. 7. Spatial pattern of period, phase gradient and migration velocity with stimulation at both ends. Data were averaged over 10 min and further averaged among many samples (n = 5). Other details are the same as for Fig. 4. (a) Period in stage a. (b) Phase gradient in stage a. (c) Migration velocity in stage a. (d) Period in stage b. (e) Phase gradient in stage b. (f) Migration velocity in stage b. Bar: S.D.

shorter period, which is different from the temperature oscillation in the same region. Under this condition, the boundary layers between the period, phase gradient and migration velocity shifted toward the cluster with longer period, as shown in Figs 10(d–f). The cluster size with the shorter period became larger than that with the longer period, indicating that the two clusters compete with each other depending on the original period.

The results obtained under the same experimental conditions were averaged, as shown in Figs 11(a–c). The point of period change in the thickness oscillation moves to the region with the longer period of

temperature oscillation. Accordingly, the boundary layers indicated by the zero intersect the phase gradient and the migration velocity also shifts to the same side. In the case of a same period modification, as superimposed on the same figures, the period of the thickness oscillation coincides with that of the temperature oscillation in every part, and the boundary layers are located in the central part of the system. These characteristics depended only on the relative difference of period, not on the absolute value of the period modulation (data not shown). Thus, it is indicated that the phase wave with the shorter period entrains the wave with the longer
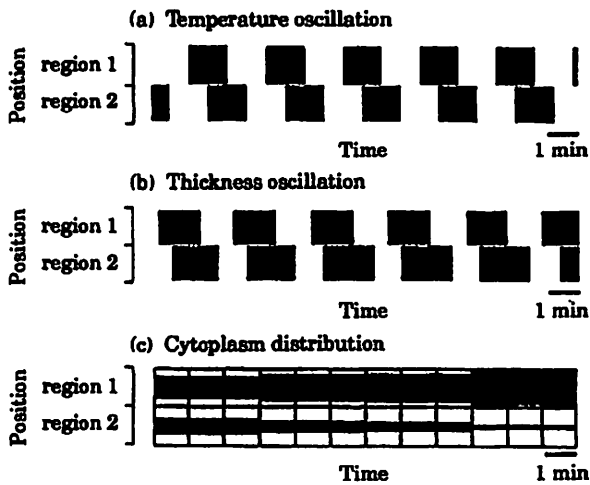
Fig. 8. Spatio-temporal development of applied temperature oscillation, intracellular oscillation and migration under the modification of spatial phase pattern. Ordinate shows position and each unit area corresponds to 2.2 mm. Other details are the same as in Fig. 3. (a) Temperature oscillation. Black and white areas stand for positive and negative changes around the mean value, respectively. (b) Thickness oscillation. (c) Cytoplasm distribution.
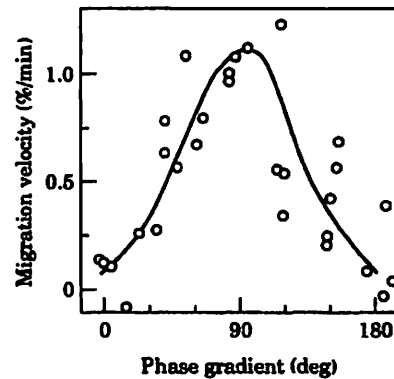

Fig. 9. Relationship between migration velocity and modified phase gradient of thickness oscillation. Migration velocity and phase gradient were averaged over 10 min for each data point; every value obtained is plotted (n = 28).

period in the coupled intracellular oscillator system. It was shown, therefore, that much information from a complicated environment is integrated by the competitive dynamics in mutual entrainment, and that the organized phase gradient pattern can encode the environment-dependent positional information field, even in a complicated environment.

4. Discussion

From the viewpoint of environment-dependent self-organization of the positional information field, we elucidated the information integration mechanism which allows coordinated tactic responses in *Physarum* plasmodium.

By analysing the relationship between the global phase wave and migration velocity in the tactic response, and by artificially modulating the spatial relationship of its phase and period, the following characteristics were clarified. The phase gradient in the wave decreases linearly according to the relative distance from the wave source, and the migration velocity in each part is regulated by the phase gradient. Thus, the spatial pattern of the phase gradient was shown to represent a kind of space coordinate which regulates the position dependent coordinated migration. Furthermore, the wave with the shorter original period corresponding to the better environmental condition, entrains a wider region than that with the longer original period. This means that

the wave source represents the best conditioned site in the whole system, even in a complicate environment. From these experimental results, it was clarified that the organized phase gradient pattern represents the environment dependent positional information field which encodes the relative positional relationship between each part and the best conditioned site in the whole system.

As shown in Fig. 12, the information integration model in the plasmodium is proposed and its dynamics is thought to be achieved by the following process. (i) Information from the environment is first incorporated as a period modulation of intracellular oscillation in each subsystem. At this stage, the local role of the subsystem in organizing the global function cannot yet be determined. (ii) These oscillations are mutually entrained and the oscillation with the shortest original period corresponding to the best conditioned site dominates the whole system. Thus, the global phase gradient pattern representing the positional information field is self-organized depending on the environmental conditions. This information field indicates the relative positional relationship between each part and the best conditioned site in the whole system. (iii) Upon interpreting this information, each subsystem is informed of its relative position within the whole system, enabling it to determine its local role in the global function for realizing coordinated migration toward the best conditioned site. Based on this self-referential information circulation between each subsystem and the whole system, the subsystems can be self-regulated in the position- and environment-dependent manner.

The theoretical model of the coupled nonlinear oscillator system has been widely investigated as a model of the spinal cord in swimming fish (Cohen

(a) Temperature
    oscillation

(b) Thickness
    oscillation

(c) Cytoplasm
    distribution

(d) Period

(e) Phase
    gradient

(f) Migration
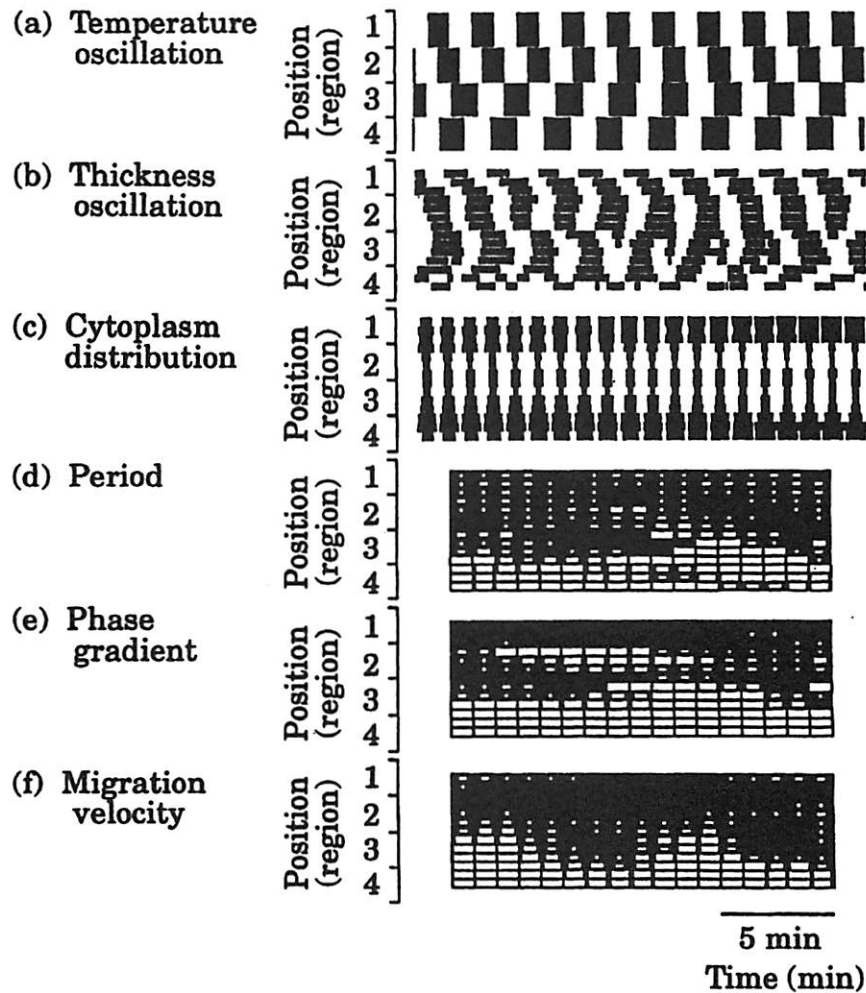   velocity

5 min

Time (min)

FIG. 10. Spatio-temporal development of applied temperature oscillation, intracellular oscillation and migration under the modification of spatial period pattern. Ordinate shows position and each unit area corresponds to 2.2 mm. Other details are the same as in Fig. 3. (a) Temperature oscillation. (b) Thickness oscillation. (c) Cytoplasm distribution. (d) Period. (e) Phase gradient. (f) Migration velocity.

et al., 1982; Ermentrout & Koppel, 1982; Koppel & Ermentrout, 1986; Koppel & Ermentrout, 1988). However their nonlinear dynamics were not analysed from the viewpoint of the positional information field. Goodwin & Cohen (1969) proposed the phase shift model for generating the positional information in biological pattern formation. They assumed two waves having different propagation velocities, and positions in the system were measured from the increase in the phase difference between the two waves. As the plasmodium is a mutual entrainment system which is open to the environment, however their model could not be directly applied to the present problem. We therefore proposed the mutual entrainment based coupled nonlinear oscillator model for the self-organization of the environment depen-

dent positional information field in the plasmodium (Miyake *et al.*, 1993). The characteristics found in the present experiments, such as the global phase gradient pattern and its environment dependence, support our model. Recently, we have been modelling the self-referential interaction process between the self-organization of the positional information field and its interpretation in a coordinated multirobot system as an engineering application (Miyake *et al.*, 1994a).

Furthermore, the information integration model proposed in the present paper will extend the framework of positional information applicable to an open system. The conventional concept of positional information is based on self-organization under a fixed boundary condition. However, the present model regards the boundary as changeable, and it
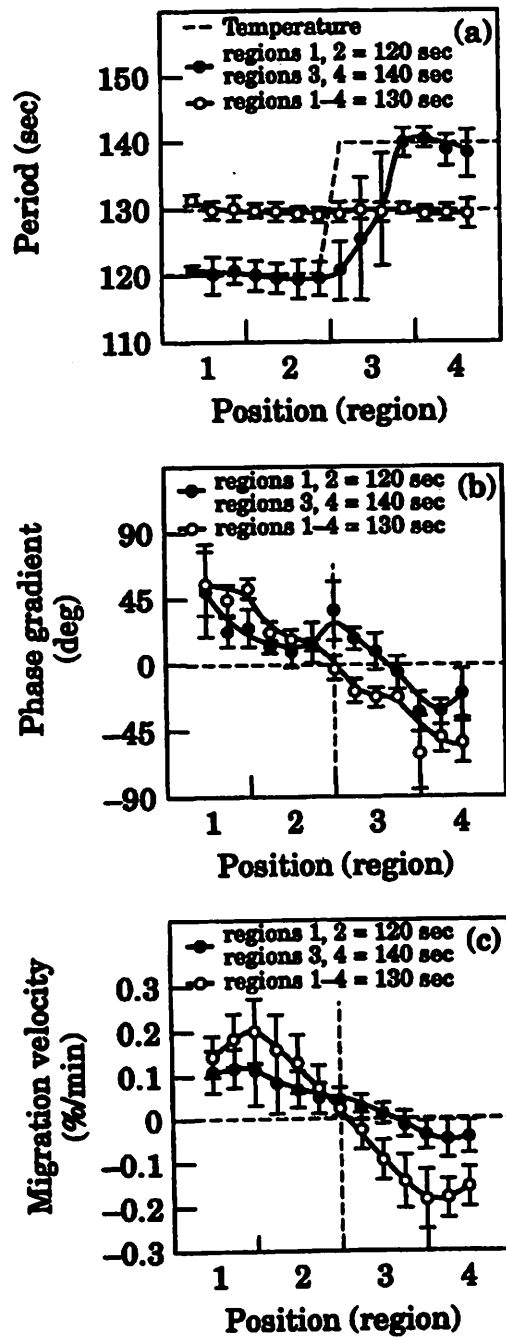
Fig. 11. Spatial pattern of period, phase gradient and migration velocity under the modification of spatial period pattern. Data were averaged over 15 min and further averaged among many samples (n = 5) under each experimental condition. The abscissa indicates the position and the left end corresponds to region 1. (a) Period of temperature oscillation and thickness oscillation. (b) Phase gradient. (c) Migration velocity. Bar: S.D.

may be embedded in a complicated and unpredictable environment. Through dynamical interaction between the system and its environment, therefore, the global attractor that determines the boundary for self-organizing the positional information is thought to be generated. Such a state of global coupling can be regarded as a consistent and functional state, taking into consideration both internal and external
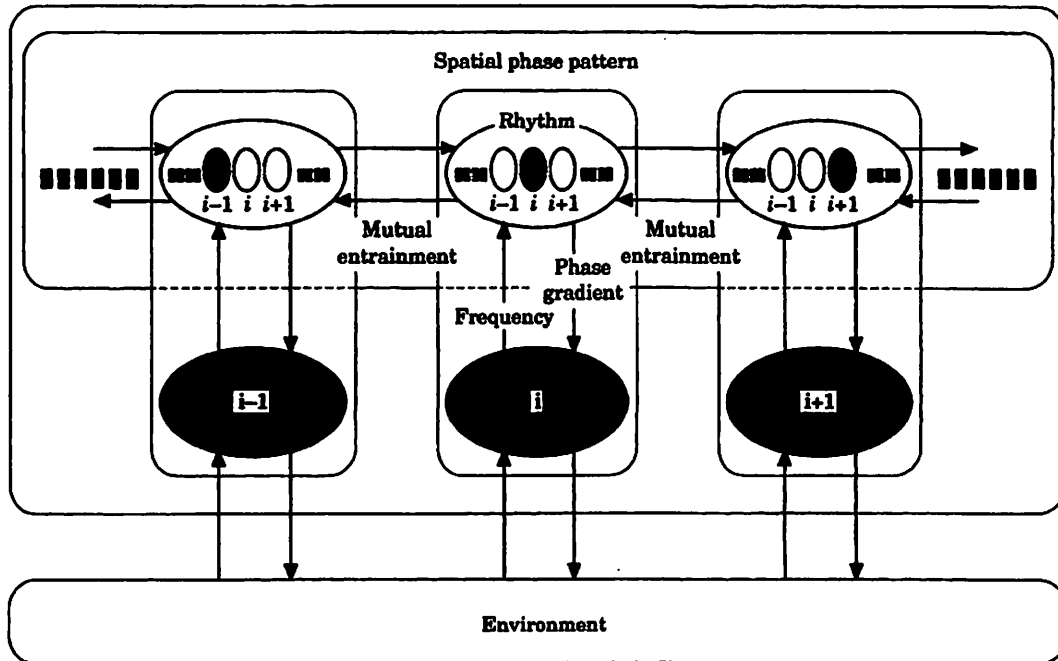
FIG. 12. Schematic illustration of information integration model in *Physarum* plasmodium. Based on this self-referential information circulation between self-organization of the positional information field and its interpretation, organism can be self-regulated to organize a coordinate function in an environment dependent manner.

dynamics. This kind of extended framework should be further investigated experimentally and theoretically.

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