Amplified expression of the gravity effect on the spatio-temporal formation of bioconvection pattern
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Abstract: We proposed a research that aims to reveal the possibility for gravity to develop new functions of the biological systems through collective interactions between the elements of the system. In this research bioconvection was focused as a research tool. Spatio-temporal analysis on the pattern formation revealed several interesting characteristics of the bioconvection, such as the pattern alteration response by *Chlamydomonas* and the responsibility to the altered gravity, which may lead to the development of the new concept in he space and gravitational biology.

*key words: collective interaction, bioconvection, parabolic flight, Tetrahymena, Chlamydomonas*

Gravity has been considered as a kind of restrictive environmental factor which provides the mechanical limits of growth and morphology of the organisms. As a part of “Ground-based Research Announcement for Space Utilization” promoted by Japan Space Forum, we proposed a research that aims to reveal a new concept concerning the effect of terrestrial gravity on the biological systems. In the proposed research we would like to demonstrate the possibility for gravity to develop new functions of the biological systems through collective interactions between the elements of the system.

Collective interactions, which are ubiquitous in nature, and the resultant dynamic instability of the system itself are known to have an ability to amplify the subtle effects of external forces, such as that of gravity on the biological event the cellular dimensions. In this paper we are stressing the importance of the bioconvective pattern formation as a research tool for our proposal which may lead to the development of the new research fields in the space and gravitational biology.

Bioconvection is a collective behavior of microorganisms. It refers to the spatial patterns that develop in suspensions of swimming microorganisms, including bacteria, ciliate and flagellate protozoa and the planktonic larvae of some invertebrates. When the top-heavy density gradient performed as the result of the gravitactic behavior of the microorganisms grows sufficiently large, an overturning convection occurs, leading to a formation of characteristic patterns, which involve highly concentrated aggregation of microorganisms extended in two-dimensional structures.

Collective interactions between individual organisms through their motile activities make a basis for the prolonged organization of the bioconvective pattern. In fact, microorganisms with different swimming activities (such as different species in the same genus and a behavioral mutant of the same species) showed different organization of bioconvective patterns. They also showed remarkable differences in the sensitivity to the altered gravitational force generated by centrifuge.

These facts suggest that the general theory of hydrodynamics is not simply applicable to the problem of the bioconvection although the theory is well suited to illustrate the many aspects of the thermal convection. It is, therefore, likely that researches should be necessary that focus much attention on the collective interactions which may make the system sensitive to gravity.

In this paper, we will show brief summary of the several interesting characteristics of the bioconvection.

1. Comparative analysis between the bioconvective pattern formation by *Tetrahymena* and *Chlamydomonas*

Since the bioconvection pattern emerges as the result of the collective interaction between microorganisms, it is informative to make comparative analyses on the bioconvective patterns generated by different organisms. *Tetrahymena* and *Chlamydomonas* have been major specimens used for the researches on the bioconvection. Each gave experimental evidences to the theories based on the completely different physical mechanisms; patterns by *Tetrahymena* gave evidences for the density-instability theory, and *Chlamydomonas* for the gyrotactic-instability theory.
Comparative analyses on the pattern formation by means of the space-time plot revealed the space-temporal aspects of the pattern formation in each microorganism.

When the suspension of *Tetrahymena* and *Chlamydomonas* was placed in a shallow chamber, bioconvective patterns emerged from the well-mixed homogeneous state. Following a short period of the initiation the convective movement became steady state. In the case of *Tetrahymena* patterns in the steady state convection were observed to move vigorously giving a crosshatch pattern in the space-time plot. *Chlamydomonas*, on the other hand, developed highly stable pattern with the regular array of dark dots surrounded by bright reticular lines. The dots in the steady state pattern appeared to keep their position, which makes parallel horizontal stripes in the space-time plot (cf. space-time plot between a and b in Fig. 1e). This difference in the stability was confirmed by the space-time plots from the side view recordings.

Analysis on the gravitactic behavior in the hyperdensity medium demonstrated that the mechanical torque is generated on the basis of the fore-aft asymmetry of the morphology rather than that of the density in *Chlamydomonas* as well as *Tetrahymena*\(^4\). This suggests that the differences found in the pattern formation is not dependent on the mechanical bias of the gravitactic orientation of the organisms.

2. **Pattern alteration response in the bioconvection of *Chlamydomonas***

We found a quite interesting behavior of bioconvective pattern created in the suspension of *Chlamydomonas*. The phenomenon, called the pattern alteration response\(^4\), was characterized by a rapid decrease in the pattern size (Fig 1). It occurred, much like a phase transition found in the physical events, spontaneously all over the suspension which already showed steady pattern formation. It was also observed that the response was initiated locally and propagated with the speed much faster than the swimming velocity of *Chlamydomonas*. Quantitative analysis of the response would stimulate further thought and research concerning gravity-related effects in biology.

3. **Responses of bioconvection pattern to the altered gravity***

In order to assess the stability of the collective motion under the influence of gravity, responses were investigated of the bioconvection pattern of *Tetrahymena* and *Chlamydomonas* to the altered gravity during...
parabolic flight of an airplane. Responses were analyzed by means of either space-time plot or discrete two-dimensional fast Fourier transformation (2D-FFT). In *Tetrahymena*, patterns were observed to increase their size under hypergravity and disappeared under microgravity (Fig 2). In *Chlamydomonas*, changes in the pattern size under hypergravity was less clear, although the patterns were observed to be enhanced under hypergravity and tended to increase in the wave number.

The patterns by *Chlamydomonas* remained less changed under microgravity. These differences in the response between organisms may be informative for the investigations for the development of the new research fields in space and gravitational biology.

The gravity dependent increase in the wave number observed in this research is almost in line with the response observed under the centrifugal, but not with those in the previous parabolic flight experiment.

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**Fig. 2** Responses of the bioconvection pattern by *Tetrahymena pyriformis* to the altered gravity during parabolic flight of the airplane (MU-300, Diamond Air Service). A, plan views of the convection pattern at the time indicated by arrows. Scale bar, 10mm. Dominant spatial frequency (wave number) calculated by 2D-FFT of convective patterns are shown in B in association with its amplitude (C). D, profiles of changes in gravity acceleration during the flight.

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**References**