Press Releases

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JAMSTEC
Marine Works Japan Ltd.
Kitasato University

Finding of Phagocytic Activities of Hemocytes from Deep-sea *Bathymodiolus* Mussel Species

~Basic Observation for Elucidating Symbiotic Mechanism of Deep-sea Organisms from Perspective of Immune Defense System~

Overview

A research team from Department of Marine Biodiversity Research (BIO-DIVE), the Japan Agency for Marine-Earth Science and Technology (JAMSTEC: Asahiko Taira, President), Marine Works Japan Ltd. and Kitasato University found that the symbiotic *Bathymodiolus* mussel species (*Bathymodiolus japonicus, Bathymodiolus platifrons, and Bathymodiolus septemdierum*) have three types of hemocytes that play distinct roles in the host defense system[*1] with different phagocytic activity[*2].

Bathymodiolus species, which belong to the family Mytilidae, are important organisms living at seeps or hydrothermal vents in deep-sea. Deep-sea *Bathymodiolus* mussels harbor symbiotic bacteria in their gill epithelial cells, which is horizontally or environmentally transmitted to the next generation of hosts. The edible shallow-water mussels, such as *Mytilus edulis* and *Mytilus galloprovincialis*, are not symbiotic. On the other hand, animals have a host defense mechanism for protection against invading bacteria. In understanding the symbiosis of *Bathymoddiolus* mussels, it is one of important issues to elucidate that such defense system is compatible with intracellular symbiosis during the evolution. However, the classification and name of hemocytes have not been consistent among scientists, despite the importance in understanding the host defense mechanism.

The research team compared and classified types of hemocytes of mytilid mussels. In conclusion, the mussels belonging to the family Mytilidae have three distinct types of hemocytes, each of which has a different phagocytic activity. These findings will provide the basic observation of studying the relationship between symbiotic and defense system of mytilid mussels.

Further studies to elucidate the roles of these different hemocyte types in the host defense system against symbiotic bacteria and exogenously invading bacteria will lead to better understanding of the symbiotic mechanism of mytilid mussels.

These study results were posted on the online journal of *Fish and Shellfish Immunology* on April 9 (JST).

Title: Phagocytic activities of hemocytes from the deep-sea symbiotic mussels *Bathymodiolus japonicus, B. platifrons*, and *B. septemdierum*.

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*1 Host defense system
It is a system that cells defense themselves against invading foreign substances by trying to remove them after recognizing the difference between cells or substances that are “self” (part of you) versus “non-self” (not part of you and potentially harmful).

*2 Phagocytic activity
It is an activity of cell that engulfs and absorbs waste material, harmful microorganisms, or other foreign bodies.

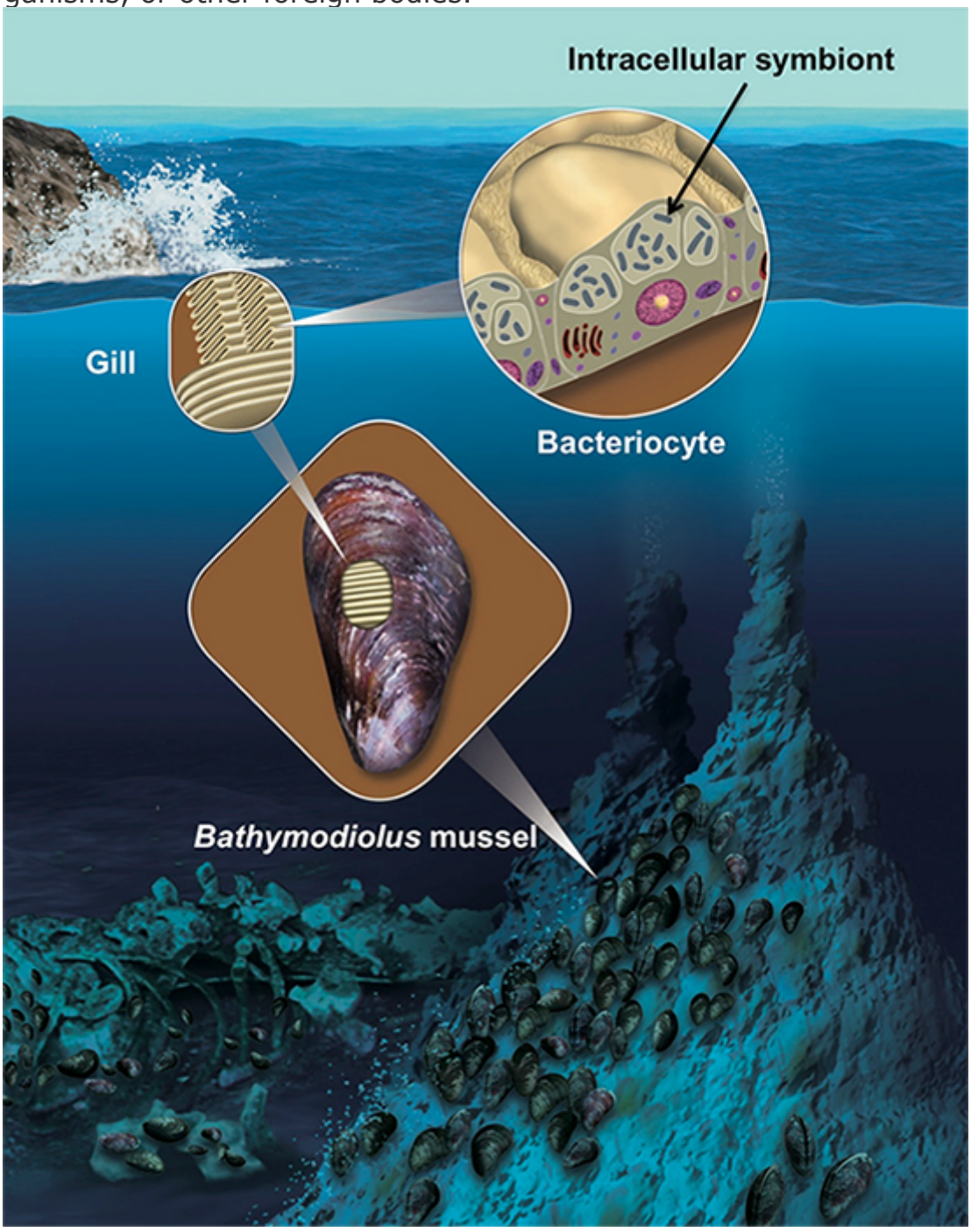

Figure 1: Symbiotic *Bathymodiolus* mussels
Symbiotic *Bathymodiolus* mussels harbor chemosynthetic bacteria in their gill epithelial cells. It is considered that chemosynthetic bacteria utilize the chemical energy to synthesize organic materials and establish their symbiotic relationship (Illustration by Nariyuki Yoshiwara).

Bathymodiolus japonicus

Bathymodiolus platifrons

Bathymodiolus septemdierum

Figure 2: Three symbiotic *Bathymodiolus* mussel species used in this study *Bathymodiolus japonicus, Bathymodiolus platifrons*, and *Bathymodiolus Septemdierum* *Bathymodiolus japonicus* and *Bathymodiolus platifrons* were collected at a seep site in Off Hatsushima Island in Sagami Bay at a depth of 850m, using the Remotely Operated Vehicle (ROV) *Hyper-Dolphin*.
Bathymodiolus septemdierum was collected at a hydrothermal vent in Myojin Knoll, Izu-Ogasawara arc at a depth of 1,300 m.

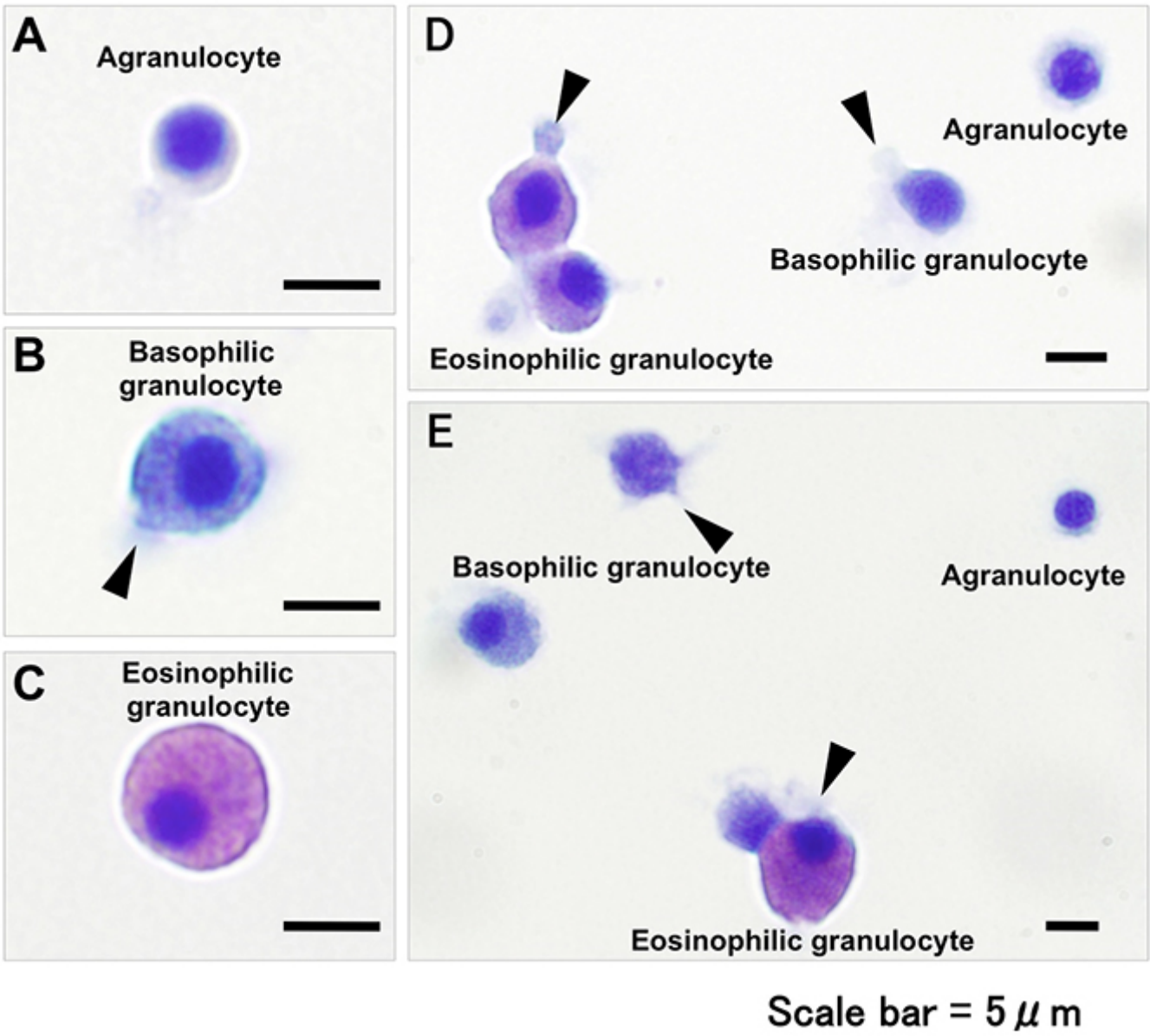

Figure 3: Light micrographs of the hemocytes of the three *Bathymodiolus* mussel species stained with May-GrünwaldeGiemsa (MGG) stain.
A, B, C:Three types of hemocytes of *Bathymodiolus japonicus* D:Three types of hemocytes of *Bathymodiolus platifrons* E: Three types of hemocytes of *Bathymodiolus septemdierum*
The arrowheads indicate pseudopodia. The nucleus and the granules of basophilic granulocyes appear in blue, while eocinophoilic granulocytes in red.

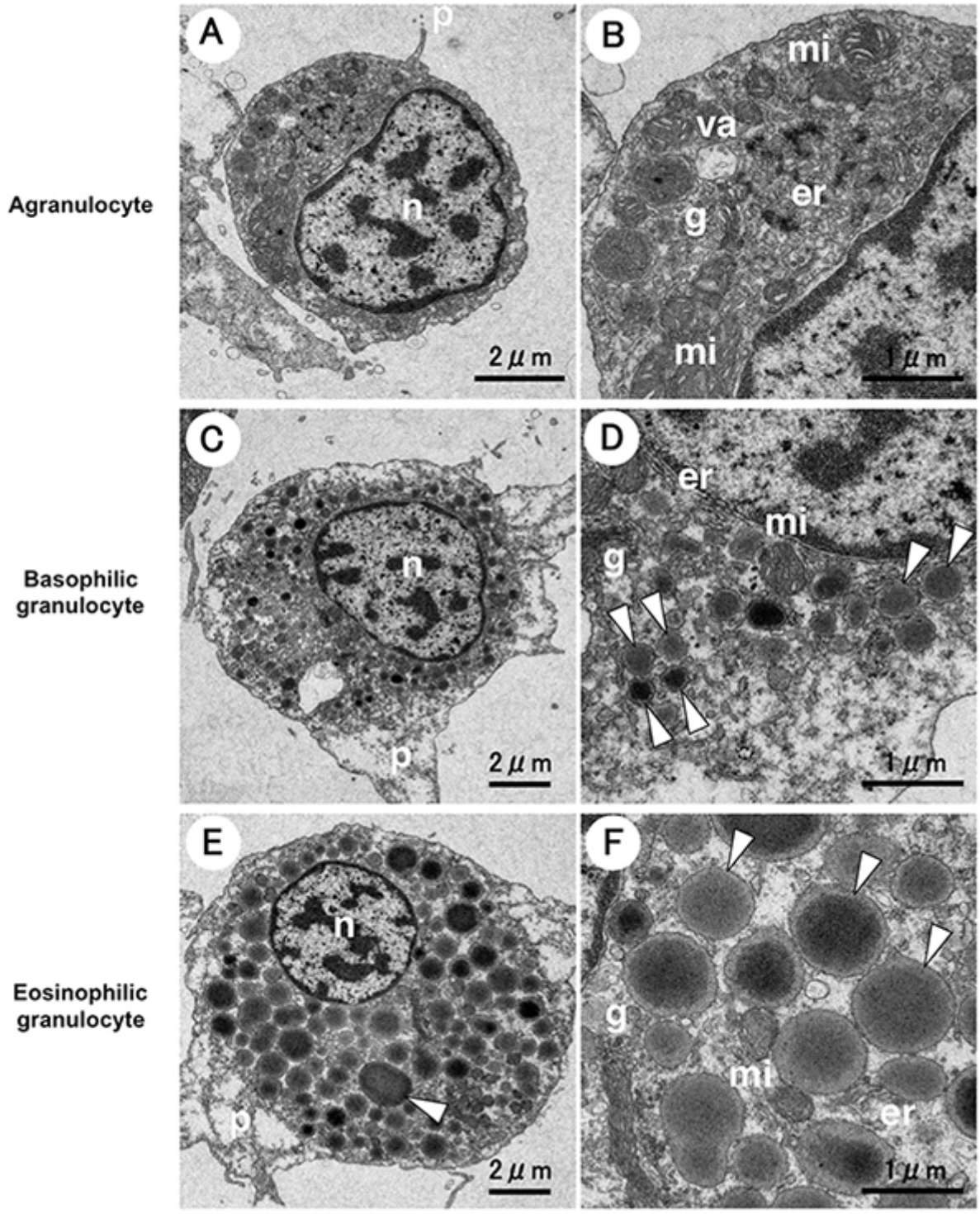

Figure 4: Transmission electron micrographs of the hemocytes of *Bathymodiolus japonicus*.
A and B: Agranulocytes; C and D: Basophilic granulocytes, which contain small electronlucent to electron-dense granules (arrowheads) with electron dense cores (arrows); E and F: Eosinophilic granulocytes containing large electron-dense granules (arrowhead) with electron-dense cores (white arrow) and more electron-lucent peripheries (black arrow). bp, broad pseudopodia; p, pseudopodia; er, endoplasmic reticulum; g, Golgi apparatus; mi, mitochondria; n, nucleus; va, vacuole.

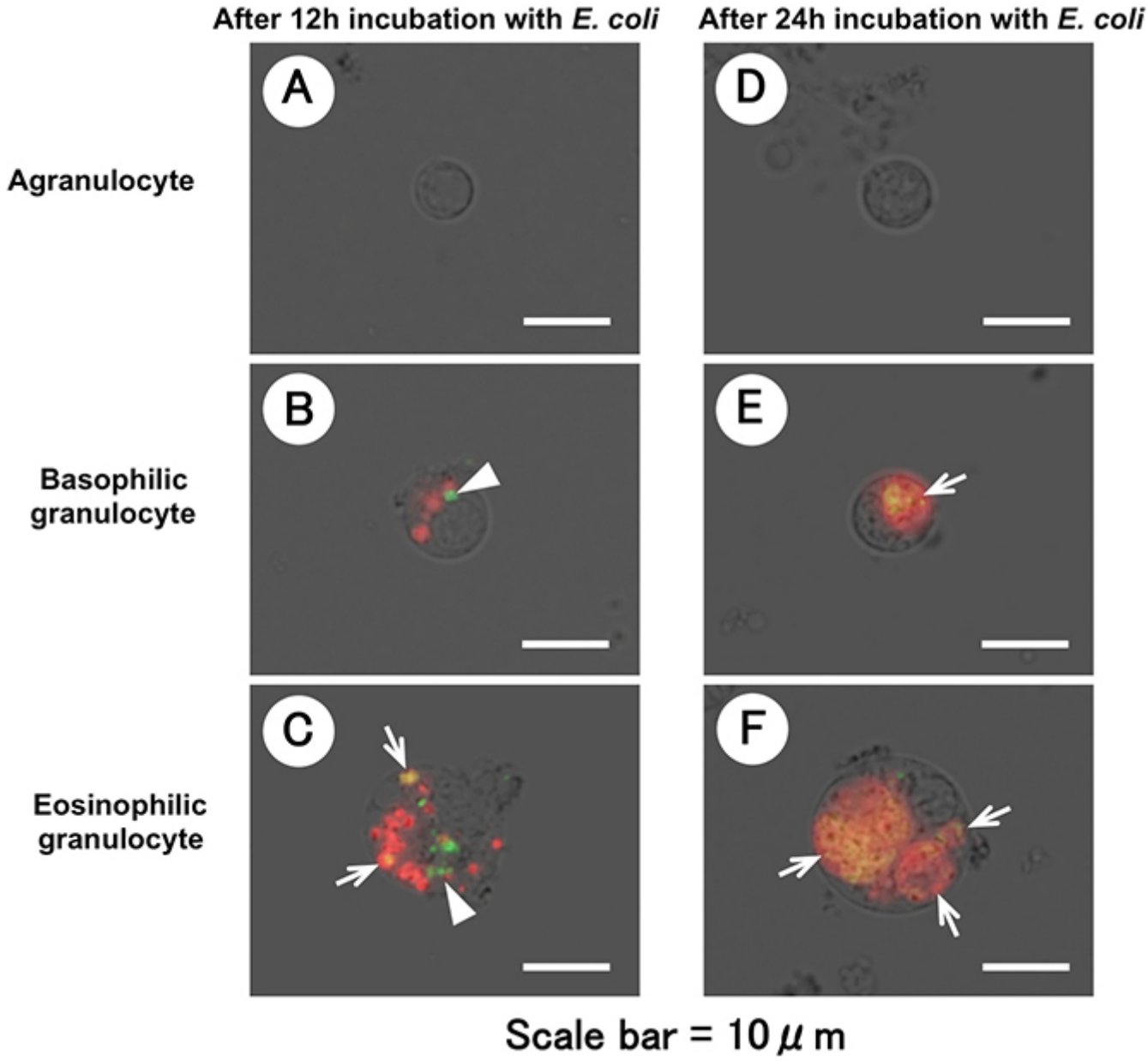

Figure 5: Analysis of phagosome-lysosome fusion in the hemocytes of *Bathymodiolus japonicus*.
Bathymodiolus japonicus hemocytes were incubated with *Escherichia coli* bio-particles (ECBP) for 2 h (A, B, and C) or 24 h (D, E, and F), and then with LysoTracker® Red solution for an additional 1 h, and observed under a fluorescence microscope. A and D: Agranulocyte (AG); B and E: Basophilic granulocyte (BG); and C and F: Eosinophilic granulocyte (EG). Arrowheads indicate green fluorescent ECBPs present in the phagosome prior to lysosome fusion. Arrows indicate orange fluorescent ECBPs present in phagolysosomes.

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