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# Early Embryonic Development of *Reticulitermes speratus* (Kolbe, 1885) (Insecta: Isoptera, Rhinotermitidae)*

**Michiyo MATSUSHIMA[1, 3] and Ryuichiro MACHIDA[2, 3]**

[1] *Graduate School of Life and Environmental Sciences, University of Tsukuba, Tsukuba, Ibaraki 305–8572, Japan*
[2] *Faculty of Life and Environmental Sciences, University of Tsukuba, Tsukuba, Ibaraki 305–8572, Japan*
[3] *Current address: Sugadaira Montane Research Center, University of Tsukuba, Sugadaira Kogen, Ueda, Nagano 386–2204, Japan*
*E-mail: machida@sugadaira.tsukuba.ac.jp (RM)*

Polyneoptera are composed of 11 "orthopteroid" orders. In contrast to many open questions about polyneopteran interordinal relationships, the monophyly of Dictyoptera comprising Isoptera, "Blattaria" and Mantodea is widely accepted. Recent phylogenetic works based on morphological and molecular evidence suggest Isoptera to nest in "Blattaria". The monophyly of Isoptera and "Blattaria", of which assemblage is called "Blattodea", is well supported, but the phylogenetic position of Isoptera within Blattodea is variously argued as well as the affinities of each blattarian lineage. Thus, we have started an embryological study of Isoptera, using *Reticulitermes speratus* (Kolbe, 1885) as materials, to reconstruct the groundplan of Isoptera and discuss its affinity with other dictyopteran lineages from the comparative embryological standpoint.

Colonies of *Reticulitermes speratus* were collected in Sugadaira (Ueda, Nagano, Japan) and main campus of University of Tsukuba, (Tsukuba, Ibaraki, Japan), and those collected in Otani-ike (Iyo, Ehime, Japan) were kindly provided by Dr. Makiko Fukui of the Ehime University. Eggs were fixed with Bouin's fixative or Carl's fixative overnight and stored in 80% ethyl alcohol. Fixed eggs were stained with DAPI solution, and then observed with a fluorescence stereomicroscope under UV excitation. A part of eggs at earlier stages of embryonic development were processed into methacrylate resin sections of 2 $\mu$m thickness. Sections were stained with Delafield's hematoxylin, eosin G and fast green FCF, and then observed with a biological microscope.

A sac-shaped embryo forms at the posteroventral side of the egg: its inner and outer layers are the embryo proper and amnion, respectively. The embryo elongates posteriorly toward the anterior pole of the egg with its progressive segmentation. When the caudal end of the embryo reaches the anterior end of the egg, the abdomen bends and sinks into yolk. After appendages formation, the embryo appears on to the egg surface with its anteroposterior axis reversed. After the embryo rotates 180 degrees around the egg longitudinal axis, it acquires its definitive form to hatch out. In the present study, we focused on the early embryonic development from the cleavage to formation of the embryo.

The cleavage of *Reticulitermes speratus* occurs synchronously until at least 64-nucleus stage. Some cleavage nuclei migrate to the egg surface at 32-nucleus stage and proliferate to form a single-cell layer. About 15 nuclei are left in the yolk. At 200–240-cell stage, the inner cells proliferate, and take their position between the center and posteroventral side of the egg. Then, these inner cells concentrate to the posteroventral side of the egg, and the sac-shaped embryo forms, accompanied with the formation of amnioserosal fold. We will further examine the embryonic development of *R. speratus* covering the whole developmental stages, to discuss and reconstruct the groundplan of Isoptera.

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