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無腸目コンボルータ科ワミノア属とその近縁群および共生藻(Symbiodiniaceae)における生態学的・分子系統学的研究

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Ecological and phylogenetic studies on genus *Waminoa* and related acoel flatworms (family Convolutidae) and their symbiotic Symbiodiniaceae

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Abstract

It is known that the genus *Waminoa* (order Acoela, Family Convolutidae) associates with anthozoans (Scleractinia, Alcyonacea, and Zoantharia) and feeds on the mucus of their hosts, and occasionally it has been found that hosts may have their whole surface covered by these flatworms. Thus, genus *Waminoa* is epizoic and is thought to have negative effects on their hosts.

In Chapter 2, we conducted phylogenetic and morphological analyses on specimens considered as *Waminoa* collected from Japan, Palau, and Indonesia. There were 18 morphotypes in the collected specimens from on at least 20 family or genera of anthozoan hosts. Furthermore, a *Linckia* starfish and another, unidentified starfish species were observed as hosts in Indonesia. The results suggest that host diversity is higher than previously reported from previous studies. Two main body shapes (obcordate and molar-like) were observed, and each formed a subclade in 18S rDNA and mitochondrial cytochrome c oxidase subunit 1 (COI) phylogenetic trees. We estimated the number of species using Automatic Barcode Gap Discovery (ABGD) analyses on COI sequences, and there were five operational taxonomic units (OTUs) in the collected specimens. Two large subclades in five OTUs contain numerous morphotypes and were associated with a variety of hosts. Finally, based on genetic distances, the molar-like shaped specimens may be considered as a genus-group separate from *Waminoa*; future studies including histology and additional molecular data are needed to clarify this.

In Chapter 3, we investigated the endosymbionts of obcordate *Waminoa*. *Waminoa* are known to contain two endosymbiotic photosynthetic dinoflagellate taxa, Symbiodiniaceae and *Amphidinium*. Moreover, these dinoflagellates have been observed in the oocytes of *Waminoa*, suggesting these algal symbionts are inherited via vertical transmission. It is known from

phylogenetic analyses of sequences of the internal transcribed spacer 2 of ribosomal DNA (ITS-2) that photosymbiotic *Cladocopium* (Symbiodiniaceae) isolated from *Waminoa* are different from Symbiodiniaceae of their anthozoan hosts. In order to re-examine the diversity of *Cladocopium* of *Waminoa*, a high resolution DNA marker, the hyper-variable non-coding region of the plastid minicircle ($psbA^{ncr}$), was used for phylogenetic analyses. These results confirmed *Cladocopium* from obcordate *Waminoa* collected from various depths formed a unique group within *Cladocopium*. Therefore, it is suggested that *Cladocopium* of obcordate *Waminoa* may be a generalist that is able to survive under different environmental conditions.

In Chapter 4, we conducted field surveys focused on distribution and occurrence of obcordate *Waminoa* on coral reefs around Okinawa Main Island. Obcordate *Waminoa* individuals were significantly more abundant close to the sandy bottom at the lower end of reef slopes and were always absent in the shallowest 5 m of every site. In Okinawa, the distribution of obcordate *Waminoa* on coral reefs is not random, but has patterns specific to certain anthozoan host species, with the highest occurrences within 10 m in depth from the sandy bottom areas of reefs.

From these results of this thesis, we suggest that at least obcordate *Waminoa* may be able to thrive as a generalist on coral reefs. Future work examining *Waminoa* diversity would be well served by examining a wide variety of potential hosts including not only cnidarians but also echinoderms and other mucus-producing benthos across a wide variety of locations and depths.