論文の内容の要旨

Shell matrix proteins of brachiopods: Implications for the mechanisms of shell formation (腕足動物の殻体タンパク質: 殻形成メカニズムの解明に向けて)

磯和幸延

Brachiopods are sessile marine invertebrates with calcitic or phosphatic shells. The brachiopod shells contain proteins that play pivotal roles in shell formation and are important in understanding the evolution of biomineralization. In this study, I performed a large-scale exploration of shell matrix proteins in the rhynchonelliform brachiopod Laqueus rubellus using proteomic analysis and transcriptome analysis. A transcriptome analysis in mantle tissue of the closely related species, Coptothyris grayi was also performed. Then, comparative analysis of shell matrix proteins in brachiopods was performed using my results with proteome data of shell matrix proteins in the rhynchonelliform brachiopod Magellania venosa and linguliform brachiopod Lingula anatina published recently (Jackson et al., 2015; Luo et al., 2015). These results provided suggestions for possible molecular mechanisms and evolution of brachiopod shell.

In chapter 1, I reviewed previous studies related to this work. Firstly, I summarized the anatomical structure of brachiopod soft body and shells as well as phylogenetic position of Brachiopoda and phylogenetic relationship of the three brachiopod subphyla. Then some previous studies of shell matrix proteins in molluscs and brachiopods were shown to emphasize the importance of sequencing shell matrix proteins to understand the mechanisms and evolution of biomineralization. Finally, I discussed significance of this study to understanding of the evolution of biominerals and to paleontological applications.

In chapter 2, I performed proteome analysis in the shell matrix proteins in Laqueus

rubellus. As a result, I identified a total of 77 shell matrix proteins. Many shell matrix proteins identified in this study have signal peptide and transmembrane region. I identified ICP-1, which is a chromoprotein detected in brachiopod shells in previous studies. ICP-1 has been sequenced completely for the first time, and showed the highest abundance in the shell. MSP130, which is a skeletal protein identified from sea urchins and oysters, was also identified. In addition, many extracellular proteins, digestive enzyme, and trypsin inhibitor were identified. Among these proteins, possible ion transporter domains, EGF-CA and EF-hand were also identified. On the other hand, 48 out of the 77 had no homologues in public databases. Among these unknown proteins, one shell matrix protein was identified with a domain architecture that includes a NAD(P) binding domain, a transmembrane region, and an aspartic acid rich region. The repertoire of brachiopod shell matrix proteins also contains a basic amino acid-rich protein and proteins. These pieces of sequence information of shell matrix proteins in Laqueus rubellus suggest that some unique mechanisms exist in brachiopod biomineralization.

In chapter 3, I performed comparative analyses of shell matrix proteins in brachiopods. The comparison of the shell matrix proteins in rhynchonelliform brachiopods showed that about half of the shell matrix proteins in Laqueus rubellus and Magellania venosa share a high sequence similarity with each other. This conserved proteins contained some extracellular, trypsin and trypsin inhibitor as well as many rhynchonelliform brachiopod specific shell matrix proteins. These shared proteins are thought to have important functions in the calcitic brachiopod shell formation. I also carried out comparative analyses of shell matrix proteins between rhynchonelliform and linguliform brachiopods. The shell matrix proteins in the linguliform brachiopod Lingula anatina share almost all detected domains conserved between Laqueus rubellus and Magellania venosa, while the rhynchonelliform brachiopod specific shell matrix proteins were not identified in the shell matrix proteins in Lingula anatina. In addition, the comparative analysis between molluscs and brachiopods showed that Peroxidasin, P-selectin and MSP130 are shared.

Finally, in chapter 4, I discussed the possible molecular mechanisms of brachiopod shell formation, and the evolution of the shell in the ‘Cambrian explosion’ as well as future perspectives based on the results of this study. This study suggests unique mechanisms in brachiopod shell formation and provides a hypothesis of molecular evolution of the shell matrix proteins involved in the independent origin of the brachiopod shells among animals.