学 位 論 文 内 容 の 要 旨

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学 位 論 文 題 名

Functional modification of mussel adhesive proteins for environmental applications
(イガイ接着タンパク質の機能改変と環境分野への応用)

Researchers persistently try to overcome the complexities of underwater environments, such as water pressure, temperature, salinity, and contaminants, in order to develop durable and effective underwater adhesives capable of establishing strong bonds even in submerged conditions. Marine mussels produce special foot proteins to adhere to rocky surfaces in wet conditions, they produce a strong underwater adhesive material called mussel adhesive proteins (MAPs). These MAPs can attach to the surface of various inorganic and organic materials in physiological conditions. Scientists have been focusing on MAPs due to their unique and remarkable adhesive properties. Low toxicity, low antigenicity, and biodegradability make them promising bio-adhesive materials in the field of biomedical, environmental, and industrial applications. However, the productivity of MAPs by mussels is very low, and the extraction of MAPs without affecting their structure and functionality requires careful techniques. Therefore, extraction and purification of MAPs in large quantities is challenging. Achieving cost-effective and large-scale production methods of MAPs is still under research. Recombinant protein production is a potential method followed by scientists to restore the natural properties of MAPs However, production of MAPs by recombinant method usually results in the aggregation of proteins and reducing the adhesive properties. These problems can be solved by functionalizing MAPs. In this study, MAPs are functionalized to improve solubility and control adhesive properties and are studied for applications in the environmental field.

Chapter 1 presents the research background, introduction to MAPs, their adhesion mechanism, research scope, and the objective, originality, and significance of the research were discussed.

In Chapter 2 literature review of research was carried out in the field of recombinant MAPs and their functionality modifications according to application or property demands was discussed.

In Chapter 3, we successfully developed a method to control the adhesion of MAPs using a fusion protein technique. Specifically, we fused Foot protein 1 (Fp1), a type of MAP, with a highly water-soluble protein derived from the C-terminal domain of ice-nucleation protein K (InaKC). The fusion protein was designed with a protease cleaving site separating Fp1 and InaKC. This innovative approach resulted in a fusion protein with reduced adhesion properties. While exhibiting enhanced solubility and stability. the adhesive property of Fp1 could be restored by removing the InaKC moiety through protease cleavage. This restoration of adhesive capability was evaluated and confirmed by observing the agglomeration of magnetite particles in water. The MAPs showed adhesion towards magnetite in the preliminary studies in our laboratory. The ability to control adhesion and agglomeration makes MAPs highly promising for the development of bio-based adhesives. The findings are significant to produce MAPs on a larger scale, allowing for controlled adhesion properties.

In Chapter 4, we employed the InaKC-Fp1 fusion protein to recover microplastics from underwater environments. Since Fp1 can adhere to a variety of surfaces underwater, we utilized this property to form complexes of different types of materials, that is, magnetite (inorganic) and plastic (organic) microparticles. The formed complex can be separated by the magnetic field, leading to the recovery of microplastics underwater. As a model, we used polystyrene (PS) microbeads as microplastic for this study. Agglomerate of PS beads with magnetite nanoparticle formed underwater efficiently by strong adhesive property of Fp1 recovered after removing the InaKC soluble tag from the fusion protein InaKC-Fp1. The controlled agglomeration of magnetite with PS beads was assessed, and microplastics were successfully separated from magnetite after removal. This study demonstrated that the InaKC-Fp1 fusion protein has the potential to serve as an effective adhesive for the controlled recovery of microplastics underwater.

Finally, Chapter 5 summarized all the results obtained in each chapter and provides prospects for future works in the above areas.