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Crystal structure of symbiosis related lectin from octocoral

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Introduction

In marine animals, lectins are believed to contribute as non-self recognition factors to the defense mechanism. Interestingly, it has been theorized that some lectins from marine animals mediate the interaction between symbiont and host. SLL-2 is a D-galactose binding lectin isolated from an octocoral, *Sinularia lochmodes*. It was found that SLL-2 was distributed densely on the surface of symbiotic dinoflagellate *Symbiodinium* sp. cells. Previous report showed that SLL-2 transforms free-swimming stage *Symbiodinium* cells into non-motile stage *Symbiodinium* cells and keep them in their non-motile stage [1, 2]. These results show that SLL-2 is a chemical cue in the symbiosis between dinoflagellates and coral. The three-dimensional structure of SLL-2 will provide information about the symbiosis mechanism.

Result

The SLL-2 protein was purified by Dr. Mitsuru Jimbo and his research group (Kitasato Univeristy). The structure of SLL-2 was determined by the molecular replacement method using atomic coordinates of *Helix pomatia* agglutinin (HPA lectin, PDB code: 2ccv) as a search model. The SLL-2 monomer (Fig.1 left) is composed of six stranded antiparallel β-sandwich consisting two three-stranded β–sheets (β1, β3, β6 and β2, β4, β5), and extended one β-strand (βN). In the monomer of SLL-2, there are two intramolecular disulfide bonds between Cys8 and Cys93, and Cys17 and Cys21. Three monomers are located around a non-crystallographic pseudo three-fold axis to form a tight trimeric globular form. The β3 and β4 from adjacent molecule extend the anti-parallel β-sheet interactions across the interface of two molecules resulting in a continuous 6-stranded large bending cleft composed by β1, β6, β3, and β4', β5', β2' (apostrophes mean the adjacent molecule). Two trimers of SLL-2 form an intertwined, dumbbell-shaped hexameric molecule (Fig. 1 right), with dimensions of 105 Å in length by 45 Å in diameter, which two globular domains are connected by a three pairs of extended βNs with β-strand like hydrogen bonding interactions using the residues 1-5. The intertwining structure in SLL-2 contributes to produce a material of strength and stability with many interchain hydrogen bonds.

The sites of N-glycosylation (N-site) and sugar binding (site 1) were identified clearly in the cleft made of two monomers. In addition, a large electron density, which appears for a part of oligosaccharides but was not enough to ensure the bound species and its orientation, was observed (site 2; a galactose molecule is tentatively fitted in Fig.1). In the SLL-2 hexameric molecule, two of the six "site 1"s possess galactopyranoside derivative that might come from the N-glycosylation site, three contain the precipitant molecule, and the remaining one accommodate a water molecule. Crystals from low GalNAc concentration and GalNAc-rich environment are also obtained. The SLL-2 hexameric molecule in the former crystal holds three GalNAcs and three precipitant molecules in its "site 1"s. The crystal structure of SLL-2-GalNAc complex from sugar-rich environment indicated that GalNAc molecules bind to all "site 1"s. These observations reveal that SLL-2 can maintain both unsymmetrical and symmetrical hexameric molecule stably across various environments. The unsymmetrical structure might be a key to understand the function of SLL-2 in the symbiosis between dinoflagellates and coral.

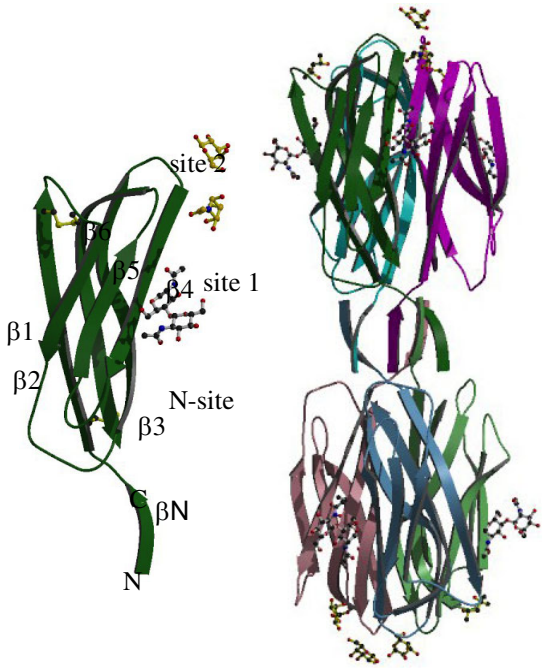

Fig. 1. Structure of SLL-2

References

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