Elucidation of the molecular basis of cell adhesion to the basement membrane

Division of Protein Chemistry, Institute for Protein Research

Professor Junichi Takagi Researchmap https://researchmap.jp/read0105692

Associate Professor Takao Arimori Researchmap https://researchmap.jp/t_arimori

Abstract

Laminins are major component of basement membranes and bind to integrins, cell surface receptors, to attach epithelial cells to the basement membranes (Fig. 1). The binding between laminin and integrin plays important roles in embryonic development, organ formation and homeostasis. We have succeeded in determining the solitary crystal structure of integrin α6β1 and the cryo-EM structure of the integrin α6β1-laminin 511 complex by utilizing of our original Fv-clasp technology (Fig. 2). These structures revealed how integrin recognizes laminin and how integrin transmits signals into the cell.

Background & Results

The surfaces of various organs are composed of epithelial cells attached to a sheet-like structure called basement membrane. Cell adhesion to the basement membrane and the associated signal transduction are essential processes for the normal functioning of each organ. Laminins, major component of the basement membranes, and their receptor integrins play a central role in cell adhesion (Fig. 1). However, the structural analysis of both laminins and integrins is very difficult due to their size and flexibility, and the details of their recognition mechanisms remain unknown.

Small antibody fragments including Fab and single-chain Fv are often used as tools to facilitate structural analysis, because the binding of antibody fragments usually stabilize the structure of target molecules. However, conventional small antibodies sometimes exhibit problems in their productivity and structural rigidity. Therefore, we have previously developed a novel small antibody fragment format "Fv-clasp" that overcomes these problems. In this study, we succeeded in obtaining important structural information for laminin/integrin by applying Fv-clasp to their structural analysis.

In the crystallization of integrin α6β1, crystals were obtained for the first time by using Fv-clasp as a crystallization chaperone, and the structure could be determined (Fig. 2, left). In addition, for the complex of integrin α6β1 and its ligand laminin 511, a very stable complex was obtained in the presence of Fv-clasp, making it possible to determine its structure by cryo-EM single particle analysis (Fig. 2, right). From these structures, we were able to visualize a large conformational change of integrin upon laminin binding. Furthermore, it revealed that the interaction between the C-terminal region of the laminin γ chain and integrin was accompanied by a local conformational change, which triggered the overall conformational change. Those structural observation clarified the molecular mechanism of integrin-mediated signal transduction in detail.

Significance of the research and Future perspective

It is known that integrin α6β1 is abundant on the surface of pluripotent stem cells, such as ES cells and iPS cells, and laminin 511 has already been widely used as a culture substrate to efficiently cultivate these cells. Our findings will greatly help in the development of better culture substrates and contribute to the field of regenerative medicine.

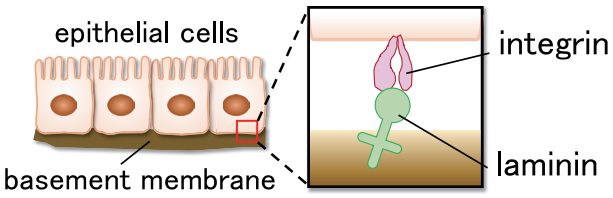

Fig. 1 Adhesion of epithelial cells to basement membranes

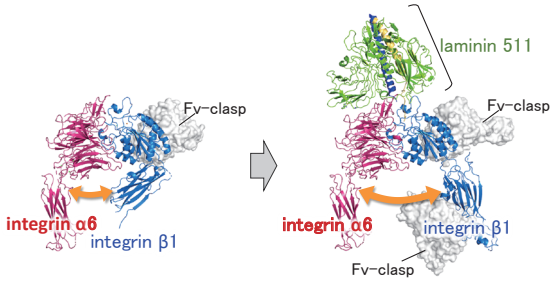

Fig. 2 Crystal structure of the integrin α6β1 (left) and cryo-EM structure of the integrin α6β1-laminin 511 complex (right)

Patent

Treatise Arimori, T et al. Fv-clasp: An artificially designed small antibody fragment with improved production compatibility, stability, and crystallizability. Structure. 2017; 25(10): 1611-1622. doi: 10.1016/j.str.2017.08.011
Arimori, T et al. Structural mechanism of laminin recognition by integrin. Nat. Commun. 2021; 12 (1): 4012. doi: 10.1038/s41467-021-24184-8

URL

Keyword cell adhesion, pluripotent stem cell, small antibody fragment