Edward Bayer

Yoav Barak
David Nakar
Alon Karpol
Jonathan Caspi
Ilit Noach
Ely Morag
Linda J.W. Shimon

The high-affinity cohesin-dockerin interaction as a model for protein-protein biorecognition

Department of Biological Chemistry

Tel. 972 8 934 **2373** **Fax.** 972 8 934 **8256**
E-mail: ed.bayer@weizmann.ac.il
Web page: www.weizmann.ac.il/Biological_Chemistry/scientist/Bayer/*BayerHome.html

The cellulosome

Many cellulolytic microorganisms produce intricate multi-enzyme complexes called cellulosomes that efficiently degrade cellulose — the most abundant organic polymer on Earth. The cellulosomes are composed of a conglomerate of subunits, each of which comprises a set of interacting functional modules. A multi-functional integrating subunit (called scaffoldin) is responsible for organizing the cellulolytic subunits into the multi-enzyme complex. This is accomplished by the interaction of two complementary classes of domain, located on the two separate types of interacting subunits, i.e., a cohesin domain on scaffoldin and a dockerin domain on each enzymatic subunit. The high-affinity cohesin-dockerin interaction defines the cellulosome structure (Fig. 1). The scaffoldin subunit also bears a cellulose-binding domain (CBD) that mediates attachment of the cellulosome to its substrate.

The cohesin-dockerin interaction

We have cloned and expressed individual cellulosomal domains and have analyzed their structure-function relationship via biochemical, molecular, and structural studies. One approach has been to determine the crystal structures of the important domains, e.g., the cohesins and the CBD. In an alternative, complementary bioinformatics-based approach, we have analyzed the cohesin-dockerin interaction by site-directed mutagenesis and "progressive gene swapping" (Fig. 2). The native interaction itself was found to be one of the strongest protein-protein interactions in nature. By comparing the cohesin-dockerin interaction between two different dockerin species, we were able to identify the important residues on both the cohesin and dockerin modules that contribute to interspecies specificity.

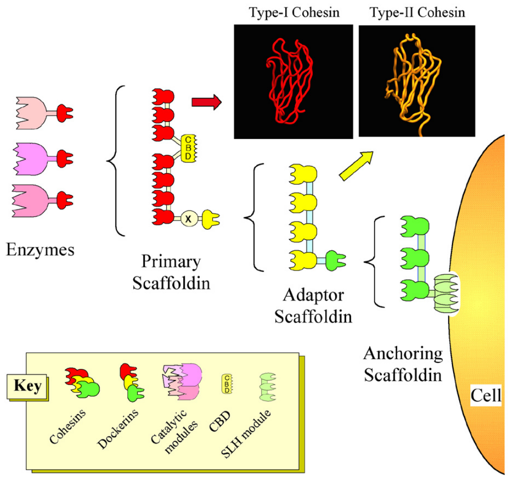

Fig. 1 *Schematic representation of the supramolecular architecture and disposition on the bacterial cell surface of a representative cellulosome system. Dockerin-containing enzymes are incorporated into a primary scaffoldin interaction with the resident type-I cohesins. The type-II cohesins of an adaptor scaffoldin interacts with the dockerin of the primary scaffoldin, and its own dockerin binds to the cohesins of an anchoring scaffoldin, whose SLH module serves to attach the entire cellulosome system to the cell surface. The respective specificities of the different cohesin-dockerin pairs are color-coded. A single CBD in the primary scaffoldin targets the complex and the entire cell to the cellulose substrate. Crystal structures for type-I and II were determined in collaborative studies. The figure implies the enormous diversity inherent in the Lego-like strategy of the cellulosome system.*

Designer cellulosomes - nanosomes

Designer cellulosomes comprise recombinant chimaeric scaffoldin constructs and selected dockerin-containing enzyme hybrids, as a conceptual platform for promoting synergistic action among enzyme components. This approach enables the precise incorporation of complementary dockerin-containing components into a designer cellulosome by simply mixing them in solution together with the chimaeric scaffoldin, thus controling the composition and architecture of the resultant complexes. This approach will eventually be appropriate for general use as a molecular Lego for application in biotechnology and nanotechnology.

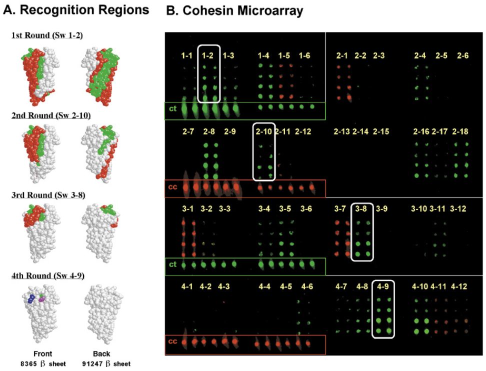

Fig. 2 *lucidation of recognition residues by progressive gene swapping. A. Mapping the recognition regions on the surface of the cohesin structure during the progressive rounds of swapping. B. Protein microarray showing the binding preference of the resultant library of chimaeric cohesins to either of the two dockerin species (red spots versus green spots).*

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