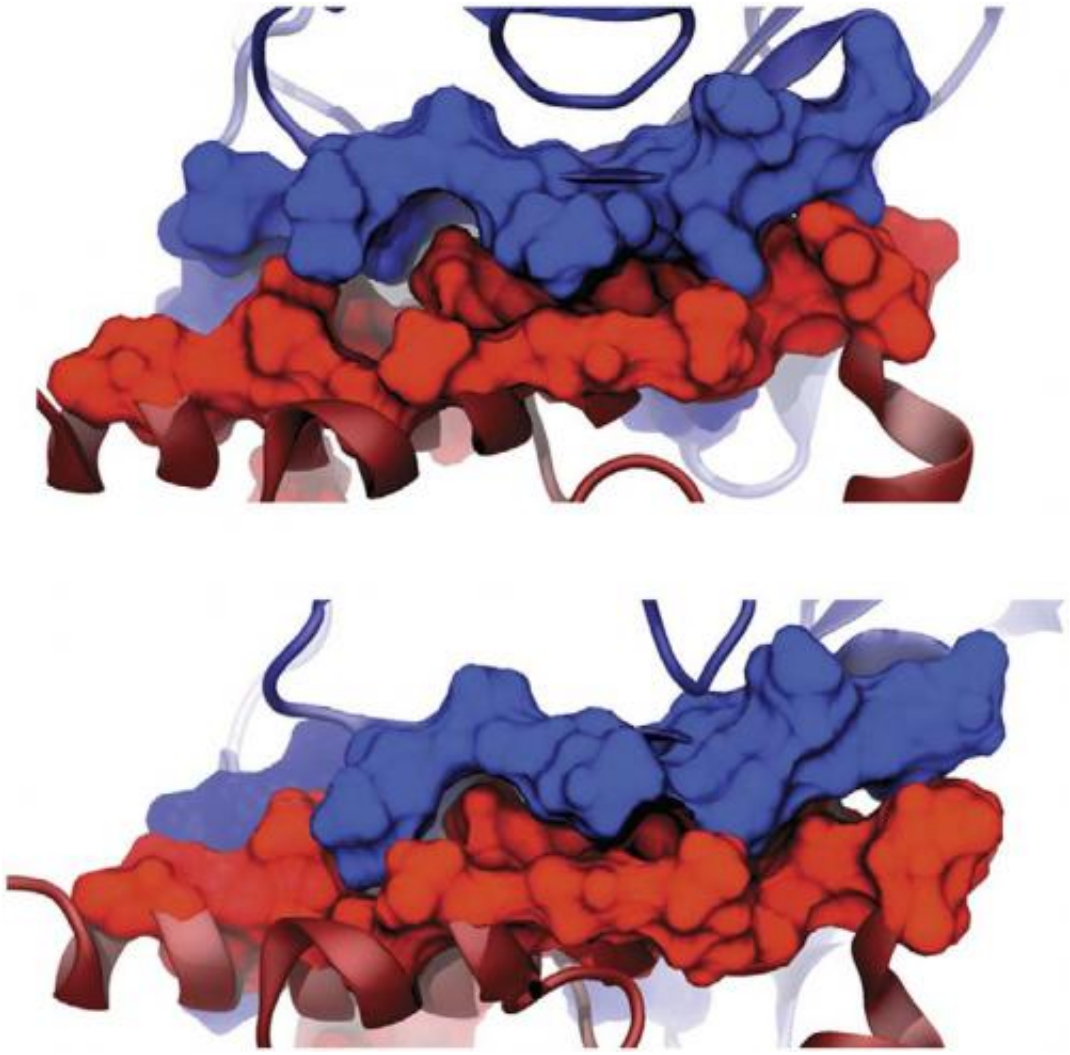
Supercomputers help solve puzzle-like bond for biofuels

March 16 2015, by Jorge Salazar



The proteins Cohesin (blue) and Dockerin (red) rearrange their surfaces to lock onto each other on contact (lower). Credit: Dr. Klaus Schulten

One of life's strongest bonds has been discovered by a science team researching biofuels with the help of supercomputers. Their find could boost efforts to develop catalysts for biofuel production from non-food waste plants.

Renowned computational biologist Klaus Schulten of the University of Illinois at Urbana-Champaign led the analysis and modeling of the bond, which behaves like a Chinese Finger Trap puzzle. "What's new is that we looked at the system very specifically, with the tools of single molecule force spectroscopy and molecular dynamics, computing it for the first time," Schulten said.

The researchers published their results in the journal *Nature Communications* in December of 2014. The biomolecular interaction binds at about half the strength of a covalent bond pieces of a finger-like system of proteins called cellulosomes used by bacteria in cow stomachs to digest plants. The main finding of the study identified the nature of the adhesion complex of cellulosomal proteins, which show extreme resistance to applied force.

The research team, in particular Rafael Bernardi of the University of Illinois at Urbana-Champaign, used the computational resources of XSEDE, the Extreme Science and Discovery Environment, a single virtual system funded by the National Science Foundation (NSF) that allows scientists to interactively share computing resources, data and expertise.

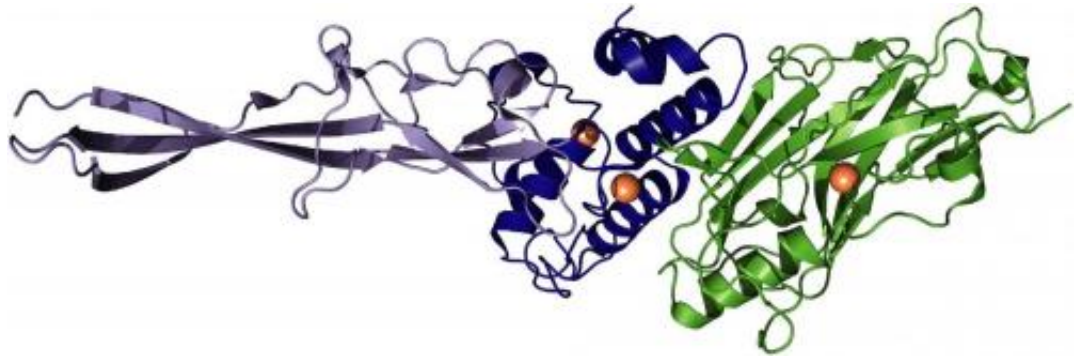
"XSEDE allowed us to employ one of the fastest supercomputers to, in parallel, perform the simulations that helped us to reveal how the building blocks of the cellulosomes become ultrastable in harsh environments," Bernardi said. "The massive amount of computer time necessary to perform our study, and the fact that fast supercomputers are necessary to have a fast iteration with experimentalists, makes a study

like this simply not feasible without support from public available supercomputers like Stampede."

Simulation and analysis of the bimolecular bond was conducted on the Stampede supercomputer of the Texas Advanced Computing Center and on the Blue Waters supercomputer of the National Center for Supercomputing Applications at the University of Illinois at Urbana-Champaign. Grants from the NSF, the National Institutes of Health (U.S.), the European Research Council, and the Excellence Cluster Center for Integrated Protein Science Munich provided most of the study's funding.

The bond behaves like a Chinese Finger Trap, a puzzle made of a grippy tube of woven bamboo. Two fingers inserted into the ends of the puzzle become stuck when one tries to pull them out.

"Analogs are never perfect," Schulten said. "But in a way, it's just like it...The word we use in biology is called catch bond, a bond that is sort of soft, but once you pull on it, you get caught."



Crystal structure of the XMod (purple)-Doc(blue):Coh(green) complex. Ca2+ ions are shown as orange spheres. Credit: Klaus Schulten

What's bonded together are two proteins, Cohesin and Dockerin. The bacteria *Ruminococcus flavefaciens*, which live inside the stomach compartment of cows, take Cohesin and Dockerin and piece them together to form a finger-like system of proteins called the cellulosome. Bacteria connect the cellulosome they assemble outside on their cell wall.

The point of all this machinery is to have scaffolding that hangs on to enzymes needed for bacteria, and ultimately the cow, to digest the variety of grass, wood chips, etc...that the animal finds to eat. "Just imagine (the cellulosome) like a hand, where the tips of the fingers contain different enzymes that can digest plant cell walls," explained Schulten. "The bacteria need to build those cellulosomes and those enzymes according to whatever plant material they encounter."

The rumen of a cow is a tough place to hang on to anything—there's enormous mechanical work being done in the form of contraction, expansion, and flows of liquid.

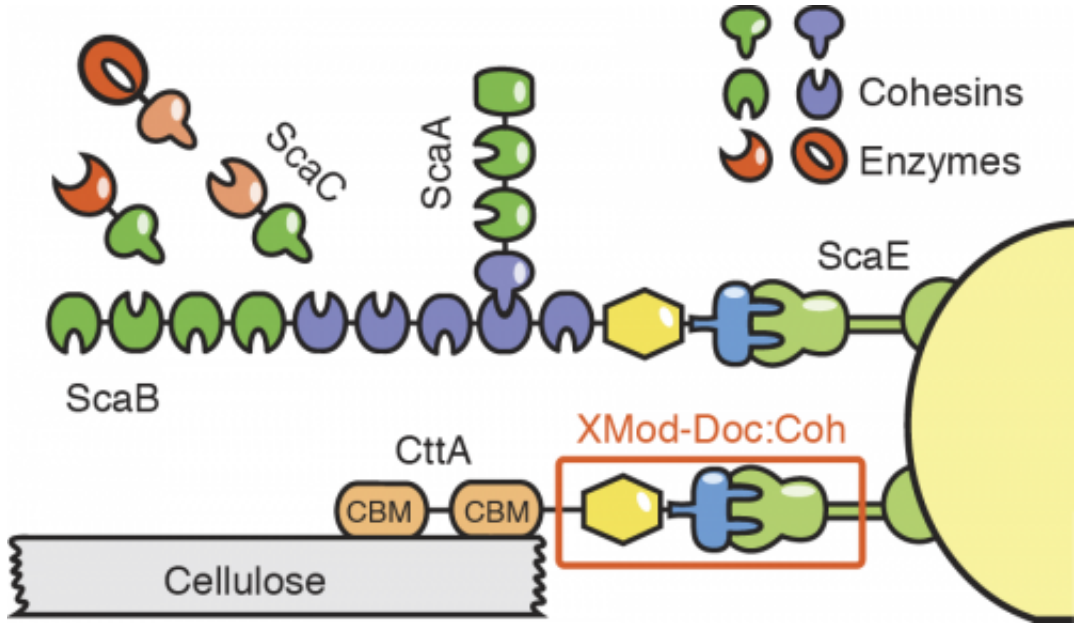
"There is some kind of puzzle, namely to piece the cellulosome together from its parts," said Schulten. That's because during construction, the forces holding together the pieces must not be very strong in order to permit flexible assembly and disassembly.

"But once the cellulosome starts to work and force is exerted on it, then the cohesion forces become very strong," he continued. "They become in fact almost as strong as complete chemical bonds that are real molecular connections between molecules. They can bond into a very strong connection, and you need to use very strong forces to break it."

Schulten's study co-author Hermann Gaub and his group at the University of Munich did just that, building and then stretching apart an XMod-Doc:Coh complex, the building block of the cellulosome. From

that the scientists measured the force extension curve, the force needed to stretch a certain extension.

"That gives you information, not the detail that tells you what physical process is going on and that permits you to explain the physical properties," Schulten said. "For that you need to simulate them."



Schematic of selected components of the R. flavefaciens cellulosome. The investigated XMod–Doc:Coh complex responsible for maintaining bacterial adhesion to cellulose is highlighted in orange.

The challenge of using molecular dynamics to simulate the Cohesin-Dockerin system was its size, which ranged in Schulten's and Bernardi's simulations between 300,000 and 580,000 atoms. What's more, they had to simulate the computationally long timescales of half a microsecond. "That is impossible for us to reproduce. But we wanted to get as close as possible to it," Schulten said.

Schulten used the NAnoscale Molecular Dynamics program (NAMD) to characterize the coupling between Cohesin and Dockerin. His group developed the widely-used scalable parallel molecular dynamics code in 1995, which he continues to update and is available for free. What's more, his group also developed Visual Molecular Dynamics (VMD), also freely available and widely used to analyze and animate bimolecular systems in 3D.

"This achievement that our program works in the most optimal way would only be possible because of the availability of staff helping us test programs, giving us advice, and also in the broad scope of the available resources in Texas, not only the Stampede computer, but also the graphics computer (Maverick) that is available there," Schulten said. "It was real teamwork that made it possible, and the staff at TACC was always absolutely outstanding."

Schulten's work champions computationally-guided research, and among his accomplishments are solving the structure of the AIDS virus with the Blue Waters supercomputer. "Only the simulations give us a view," Schulten said. "The simulations are a kind of computational microscope that tells engineers this is what is happening. This is why it works. This is how we can improve it."

Schulten said this research on the cellulosome has fertile ground for application to benefit society. Biofuels made from non-food plant such as corn stalks or straw are currently too costly to produce at scale because the enzymes needed to digest the tough material are expensive. Engineered bacteria might one day be able to lower that cost by more efficiently delivering enzymes to plant cellulose.

"Understanding cellulosomes might actually allow us one day to design our own cellulosomes that we make just for the purpose of stable, long-time digestion of plant materials and other toxic materials. And in

particular, you learn to take advantage of the modularity of a cellulose-type system that we engineer, particularly also making such strong bonds as we investigated in the experiments and in the simulations that would give us an incredibly versatile, mechanically stable, long-lived, robust tool in the hands of engineers. This is very important for sustainability, for cleaning the environment, etc...There is a really long perspective in this kind of work."

More information: *Nature Communications*, www.nature.com/ncomms/2014/141 … full/ncomms6635.html

Provided by University of Texas at Austin