Parallel computing to tame biofuel

THE 21ST CENTURY MAY BELONG TO BIOSCIENCE AND THE POWER OF COMPUTING, WRITES
S ANANTHANARAYANAN

In a warming world, biological processes would be the viable way to provide food as well as materials for non-food applications. And it is with the help of increasing computing ability that the world may be able to manage the growing complexity of demand and supply while keeping the drain on earth's resources under control.

A team at the Oak Ridge National Laboratory, Tennessee, has pressed into service the massive computing potential of the supercomputer Titan, with the capacity of 10 petaflops, or ten million times a billion decimal calculations every second, to understand the mechanics of a roadblock in one of the most urgent and promising solutions that bioscience may be able to provide to replace fossil fuels.

The solution in question is the production of ethanol, or alcohol, from biomass or plant tissue to power all kinds of machinery and transport, which now depend on petroleum, and possibly for generating electricity by replacing coal as the fuel for power plants. And the roadblock is that the capacity to get ethanol out of biomass is severely limited by *lignin*, a natural constituent of plant material itself.

The most common method of getting ethanol from plant material is by fermentation of the sugars in fruit or sugars derived from starch in grain. Ethanol is produced for industry from cane sugar or from corn that is grown for this very purpose. While cane sugar can be fermented directly, the starch in corn is first converted to sugars by the action of enzymes and the sugars are then fermented, the runoff distilled to get the required grade of alcohol. The commercially attractive proposition, however, is not from cane or from corn but from the inedible, cellulose material in plant tissue.

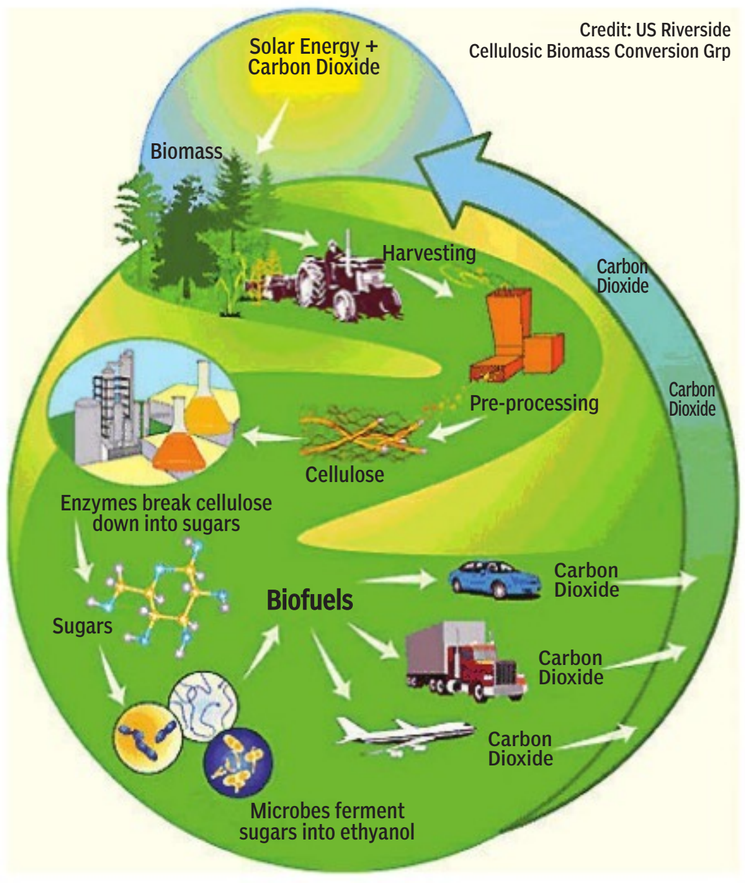
The advantage with cellulosic ethanol is that cellulose is abundant without having to be specially cultivated, so that its use does not affect the land available for growing food-grain. A major negative is the extra cost in the pretreatment and special enzymes that are needed to break cellulose into fermentable sugars, but the economics are still in its favour. The table shows the advantage of ethanol from corn or cellulose over gasoline, along with the case of what happens if land is used exclusively to grow cellulose.

We can see that there is positive saving of GHG emissions with the use of ethanol. This despite the negatives in the last column, because cellulose actually does not need to be cultivated specially as it is abundant in unused plant waste, paper and packaging waste and wood shavings and in good quantity from woody forest waste. There is more cellulose, in fact, than any other organic molecule on earth!

The problem

The function of cellulose, which forms the bulk of the plant cell wall, is to provide both protection and a rigid framework for the plant to stand and receive air and sunshine and for channels that allow movement of nutrients. The cellulose fibre alone, however, does not have the necessary rigidity and it derives strength from lignin, a complex chain molecule that forms the scaffolding on which cellulose makes up the cell wall. Hardier plant tissue, like the bark of trees, is particularly rich in lignin, which is resilient and helps plant tissue combat corrosion and weathering. The trouble is that when plant tissue is made use of for the non-plant purpose of being broken down to sugars for fermentation, its lignin contents continue to exercise their protective role.

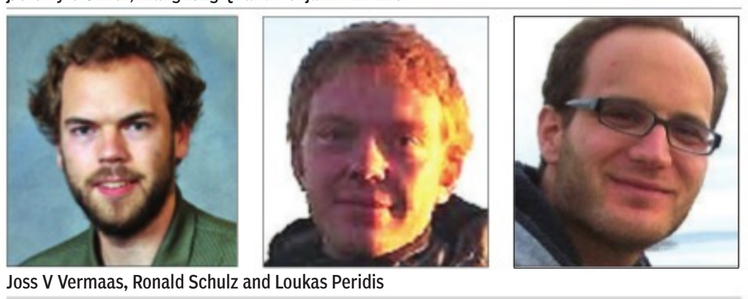
The first step in processing biomass is heating with dilute acid to remove most of the non-cellulose components of biomass, which is generally successful, except for the lignin content that persists. The objective is to increase the access to cellulose of the enzymes that convert cellulose to sugars. But even after this treatment, the conversion remains incomplete and the suspicion has been that it is the presence of lignin that is responsible. Some ways of how this comes about could be cellulose associating with lignin in a way that the enzyme is prevented from acting on cellulose or lignin binding with the enzyme to disable its action, says a paper by Jeremy C Smith, professor at the University of Tennessee and director of the Oak Ridge National Laboratory Center for Molecular Biophysics, with Josh V Vermaas, Loukas Petridas, Xianghong Qi, Ronald Schulz and Benjamin Lindner, in the journal, *Biotechnology for biofuels*. Evidence for the hypothesis or about the mechanism of its action, however, has not been available, the paper says.



Computer simulation

As both cellulose and lignin are polymers, or molecules where a pattern repeats hundreds of times, and the chain winds and unwinds millions of times


jJeremy C Smith, Xianghong Qi and Benjamin Lindner


Joss V Vermaas, Ronald Schulz and Loukas Peridis

a second in the ceaseless motion at the atomic scale, experimental investigation into how lignin actually inhibits the action of enzymes is not practical. The team at the ORNL, hence, created a computer simulation to represent the interactions to try out and analyse the billions of orientations of atoms.

For a fair representation of the problem, the simulation needed to consider as many as 23.7 million atoms and the computer had to take into account the orientations and interactions of the mechanical and electrical forces that act in this huge complex of cellulose, lignin and the added enzymes. The modelling was limited to basic forms of the cellulose chains and the commonly encountered lignin molecules, and other parameters were based on available experiential data.

Established algorithms, or computer procedures to mimic the interactions, and available software packages were then used to analyse the huge data arising from simulation of even a few seconds of the atomic scale process.

As each instant of the process involved computation of millions of atoms, the processing was carried out with the help of Titan, the mega, parallel processing computer system at the Oak Ridge laboratory. Dealing with huge computations, as one that involves millions of atoms, can take years if the operations are done one after another. Titan, hence, splits the work into independent sections and carries them out simultaneously with the help of effectively more than 18,000 separate computers working together.

As can be imagined, this also needs software to separate the parallel procedures in the virtual model of the atomic system and then to put things together, for each instant of the interaction being modelled. Each of the runs of the simulation hence covered very short stretches of the actual process, the longest being 45 nanoseconds and the total over the course of a year has come to 1.3 microseconds. This much time, however, represents a viable collection of information of a process that occurs very rapidly and it has been possible to piece together a useful picture of how lignin affects the interaction of cellulose and enzymes.

The results of the analyses were to confirm the suspicions, but with detail at the atomic level that lignin forms bonds with cellulose at the very places where the enzymes would need to dock. This by itself impedes or limits the action of enzymes. But lignin goes further — it forms bonds with the enzyme at the places in the structure of the enzyme that are needed for binding with cellulose. In this way, the action of the enzyme is hit twice — the targets are blocked and where they may be free the enzyme itself is disabled.

Another detail found through the simulation was that lignin formed bonds more readily with a crystalline form of cellulose than with a non-crystalline form. The knowledge of the molecular mechanism of lignin action and the preference for the crystalline form could lead to processes, on the one hand, for engineering the form of lignin so that it bonds less with cellulose and, on the other, to favour production of non-crystalline cellulose during pretreatment. "Eventually, we would like to construct a simple model of a plant cell wall that we could process in silico, or via computer simulation, and see how it changes during pretreatment," says Loukas Petridis, staff scientist at the Center for Molecular Biophysics. "We're trying to reach the complexity that is found in nature and industrial conditions."

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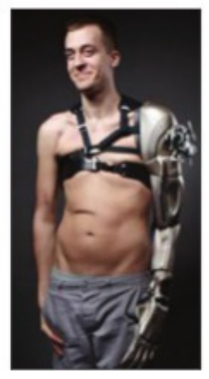
CO_2 released for every kilo-watt-hoir of energy produced

Fuel type	CO_2 released	Reduction of GHG by use of ethanol	Including use of reduction of GHG land for cultivator by use of ethanol	
Gasoline	331 grams		331 grams	
Corn ethanol	266 grams	20%	637 grams	-93%
Cellulosic ethanol	101 grams	70%	497 grams	-50%

PLUS POINTS

Hi-tech synthetic arm

The job advertisement was highly specific: applicants had to be passionate about computer games and live in the UK. Oh, and they also had to be amputees who were interested in wearing a futuristic prosthetic limb. James Young knew straight away he had a better shot than most. After losing an arm and a leg in a rail accident in 2012, the 25-year-old Londoner had taught himself to use a video-game controller with one hand and his teeth. "How many amputee gamers can there be?" he asked himself. In the end, more than 60 people replied to the ad, which was looking for a games-mad amputee to become the recipient of a bespoke high-tech prosthetic arm inspired by Metal Gear Solid, one of the world's best-selling computer games. Designed and built by a team of 10 experts led by London-based prosthetic sculptor Sophie de Oliveira Barata, the £60,000 carbon-fibre limb is part art project, part engineering marvel. For Young, who unveiled the new prosthetic on 20 February at BodyHacking Con 2016, a conference in Texas devoted to "human augmentation", the synthetic limb is likely to be life-changing, both in terms of its functionality and the levels of attention it will bring him. "I'll be on stage in Texas talking about it," he said before boarding his flight to the USA. "That will be a different level of attention — I'll have to get used to it."



The limb is fitted with a 3D-printed hand that is controlled by sensors that detect minute muscle movements in Young's back. Designed by Bristol firm Open Bionics, it is substantially more dextrous than the rudimentary NHS prosthetic he acquired following his accident.

RICHARD JINMAN/THE INDEPENDENT

Timely flood alarms

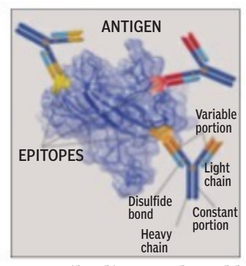
Communities in flood-prone areas often do not have access to information about flood forecasting. As a result, they do not have sufficient time to evacuate and put their cattle and belongings in a safe area. The new Community-Based Flood Early Warning System, the International Centre for Integrated Mountain Development's new initiative, now allows communities downstream to access almost real-time information about the water level upstream.

The system is cheap (at $1,000) and the technology simple. A solar-charged transmitter has a flood gauge set up on a river and a receiver has a control unit installed in a household on the bank. As the water rises, the electronic sensors produce an alarm that is communicated to the receiver through a wireless device. A caretaker observes the risk level and sends a flood-warning message via mobile phone. The message is relayed to the focal person living in a flood-prone downstream village and the project team and the district disaster management authorities further disseminate the information to vulnerable communities further downstream. Those living along some 45 flood-prone villages downstream of the Jiadhal and Singora rivers in India are already benefitting from the service.

SCIDEV.NET

Better reagents

Facing problems of inconsistent, time-consuming, and costly antibody production, some researchers are turning to alternatives to target specific proteins of interest in the lab and in the clinic. Antibodies are large proteins, weighing in at about 150 kDa. Four polypeptides — two heavy chains and two light chains — are linked by disulfide bonds to form a Y-shape molecule. The amino acid sequences at the tips of the short ends of the Y vary greatly between antibodies produced by different B cells, while the rest of the molecule is relatively consistent. The variable portion of the antibody binds in a specific region (epitope) on a foreign protein (antigen) and signals the immune system to the presence of an invader.



To produce antibodies, researchers immunise lab animal with a protein of interest. The animal's B cells then generate antibodies that bind to different regions, or epitopes, on the protein. The diverse antibodies that bind to the target protein can then be isolated and purified for use. Because these bind numerous epitopes, they are called polyclonal antibodies. Alternatively, the immunised animals' B cells can be isolated from the spleen or lymph nodes and fused with a tumor cell to generate immortal hybridoma lines. Cell lines that produce the desired antibody against a specific epitope of the target protein can then be grown in large bioreactors to scale up production.

THE SCIENTIST

PROGRESSING IN PHASES

SEVERAL CRITICAL TRANSITION POINTS CONTROL THE SERIES OF EVENTS DURING CELL DIVISION CYCLE, WRITES **TAPAN KUMAR MAITRA**

The control system that regulates progression through the cell cycle must accomplish several tasks. First, it must ensure that the events associated with each phase of the cell cycle are carried out at the appropriate time and in the right sequence. Second, it must make sure that each phase has been properly completed before the next one is initiated. Finally, it must be able to respond to external conditions that indicate the need for cell growth and division.

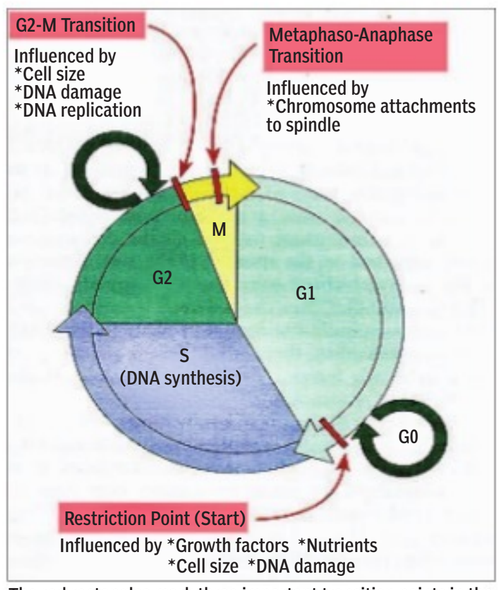
The preceding objectives are accomplished by a group of molecules that act at key transition points in the cell cycle. At each of those points, conditions within the cell determine whether or not it will proceed to the next stage of the cycle.

The first such control point occurs during late G1. This is the phase that varies most among cell types, and mammalian cells, which have stopped dividing, are almost always arrested during G1. For example, in cultured cells one can stop or slow down the process of cell division by allowing them to run out of either nutrients or space or by adding inhibitors of vital processes such as protein synthesis. In all such cases, cycle is halted in late G1, suggesting that progression from G1 into S is a critical control point. In yeast, this control point is called start — it must have sufficient nutrients and reach a certain size before they can pass through start. In animal cells, the comparable control point is called the restriction point. The ability to pass through the restriction point is determined to a large extent by the presence of extracellular growth factors, which are proteins used by multi-cellular organisms to stimulate or inhibit cell proliferation.

Cells that have successfully passed through the restriction point are committed to S phase, whereas those that cannot do so enter into GO and reside there for variable periods of time, awaiting a signal that will allow them complete the process. A second important transition point occurs at the G2-M boundary, where the commitment is made to enter into mitosis. In certain cell types, the cycle can be indefinitely arrested at the end of G2 if division is not necessary — under such conditions, the cells enter a non-dividing state akin to GO. The relative importance of controls exerted during late G2 or late G1 in regulating the rate of division by transiently halting the cell cycle varies with the organism and cell type. In general, arresting the cell cycle in late G1 (at the restriction point) is the more prevalent type of control in multi-cellular organisms.

A third key transition point in the cell cycle occurs at the junction between metaphase and anaphase, where the commitment is made to segregate the chromosomes into two new daughter cells and exit from mitosis. Before cells can pass through this transition point and begin anaphase, it is important to have all the chromosomes properly attached to the spindle. If the two chromatids that make up each chromosome are not properly attached to opposite spindle poles, the cell cycle is temporarily arrested to allow spindle attachment. In the absence of such a mechanism, there would be no guarantee that each of the newly forming daughter cells would receive a complete set of chromosomes.

Cell behaviour at the various transition points is influenced by successful completion of preceding events in the cycle and by factors in the cell's environment.



The red rectangles mark three important transition points in the eukaryotic cell cycle where control mechanisms influence whether or not the cell will continue to proceed through the cycle. That determination is based on chemical signals reflecting both the cell's internal state and its external environment. The two circular, dark green arrows indicate locations in late G1 and late G2 where the cell can exit from the cycle and enter a non-dividing state.

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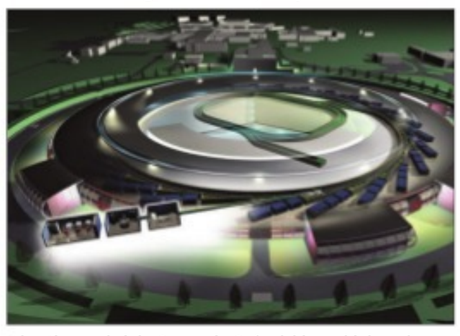
Diamond Light Source

STEVE CONNOR REPORTS ON HOW A GIANT MICROSCOPE COULD HELP FIND OUT HOW BACTERIAS ARE ABLE TO RESIST ANTIBIOTICS

A new generation of antibiotics could emerge from a study revealing the hidden Achilles' heel of many drug-resistant superbugs, according to scientists. Powerful light beams many millions of times brighter than the sun have for the first time taken detailed images of the complex, molecular structure that helps to build the protective cell wall of disease-causing bacteria. Researchers believe that understanding how this molecular machinery is arranged at the atomic scale within the bacterial cell wall holds the key to developing new kinds of antibiotic drugs that can breach the outer defences of resistant microbes.

The study revealed how the structure, known as the Beta-barrel Assembly Machinery, is made up of five protein sub-units that work together to build the protein gateways of the bacterial cell wall, which allow vital nutrients to flow into the microbial cell. The Bam machinery is found in all "gram-negative bacteria" — a class of microbes that causes a range of potentially lethal infections from pneumonia and meningitis to blood sepsis and food poisoning. Antibiotic resistance is a particular problem with gram-negative bacteria.

By mapping the three-dimensional positions of the five Bam sub-units, scientists believe they have finally found a way of understanding how to block the machinery that builds the cell wall's gateways, and in doing so starve the microbe of vital nutrients. They said the findings paved the way for a new generation of drugs that kill superbugs by tearing down their defensive walls rather than attacking the bacteria itself. This approach might even prevent the development of further drug-resistance, they added.



The Diamond Light Source in Oxfordshire, a high-tech research centre, produces highly focused X-ray, infrared and ultra-violet beams.

"Bacterial multi-drug resistance, also known as antibiotic resistance, is a global health challenge. Many current antibiotics are becoming useless, causing hundreds of thousands of deaths each year. The number of superbugs is increasing at an unexpected rate," said Professor Changjiang Dong of the University of East Anglia in Norwich, who led the study published in the journal *Nature*.

"Gram-negative bacteria are one of the most difficult ones to control because they are so resistant to antibiotics. All gram-negative bacteria have a defensive cell wall. Beta-barrel proteins form the gates of the cell wall for importing nutrition and secreting important biological molecules," Professor Dong said. "The Bam is responsible for building the gates — the beta-barrel proteins — in the cell wall. Stopping the Beta-barrel Assembly Machine from building the gates in the cell wall cause the bacteria to die," he said.

Unravelling the mechanism could also help to understand human cell dysfunctions linked to disorders such as diabetes, Parkinson's and other neurodegenerative diseases, he said, as similar molecular machinery is also found in the cell membranes of human mitochondria — the microcopic "power packs" of the cells. "In human mitochondria, a similar complex called Sorting and Assembly Machinery is responsible for building the outer membrane proteins in the outer membrane of mitochondria."

Intense light beams, some 10 billion times brighter than the sun, produced by the Diamond Light Source in Oxfordshire were used by the scientists to explore the atomic detail of the bacteria's machinery for making the vital gateways of its own cell walls. The study showed how Bam's five sub-units – BamA, BamB, BamC, BamD and BamE — were assembled into a working, sub-microscopic machine for inserting protein gateways in the outer wall.

"Our research shows the whole Bam structures in two states — the starting and finishing states. We found that the five sub-units form a ring structure and work together to perform outer membrane protein insertion using a novel rotation and insertion mechanism," Professor Dong said.

The Diamond Light Source in Harwell, Oxfordshire, is one of the most advanced scientific machines in the world.

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