

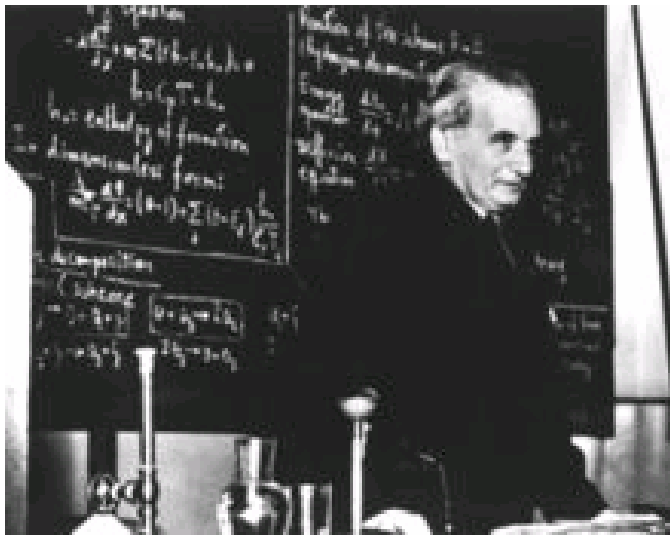


Synthetic Biology

Andrew Hessel

July 4 2007

ahessel@gmail.com



*A Scientist discovers that
which exists; an Engineer
creates that which never was.*

-- Theodore Von Kármán

“Synthetic biology is an emerging area of research that can broadly be described as the design and construction of novel artificial biological pathways, organisms or devices, or the redesign of existing natural biological systems.”

Technology of synthetic biology

It's all about DNA...

DNA is the machine language
program for biochemical processes

The ability to manipulate this one molecule allows virtually anything biological to be engineered!

DNA is to biology as the electron is to computing



Reading code





1980
500 bp/day (manual)



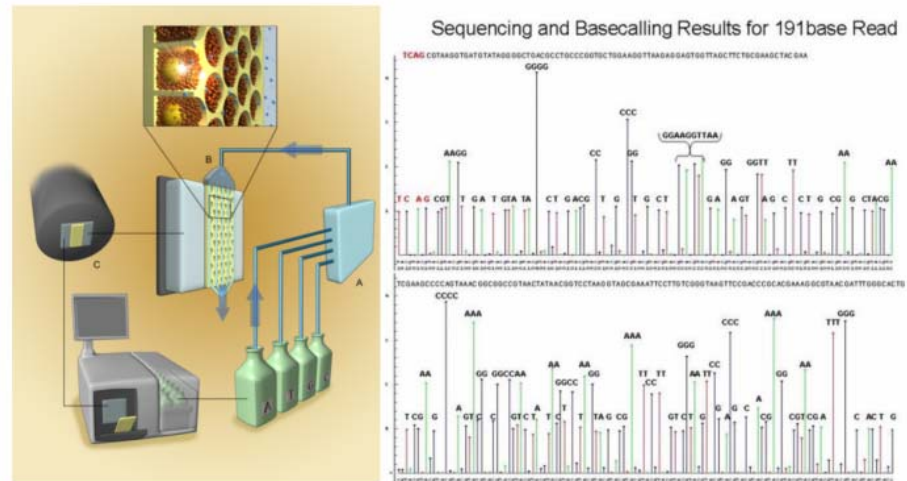
1987
36,000 bp/day (semi-auto)



1995
144,000 bp/day (semi-auto)



1998
500,000 bp/day (automatic)



2007 – Sequencing by Synthesis



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Comprehension



Volume 16 Number 5 1988

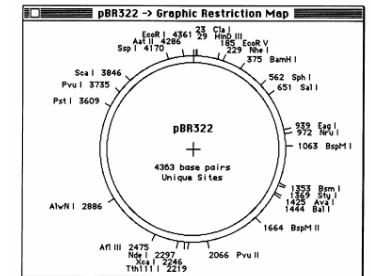
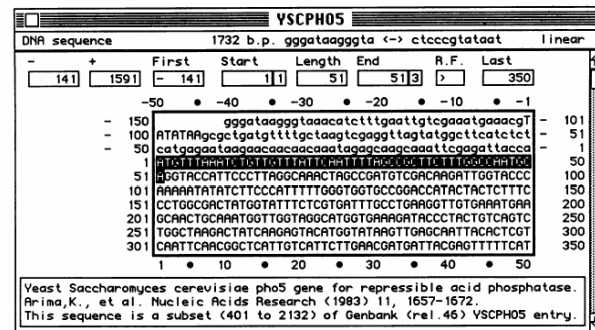
Nucleic Acids Research

'DNA Strider': a 'C' program for the fast analysis of DNA and protein sequences on the Apple Macintosh family of computers

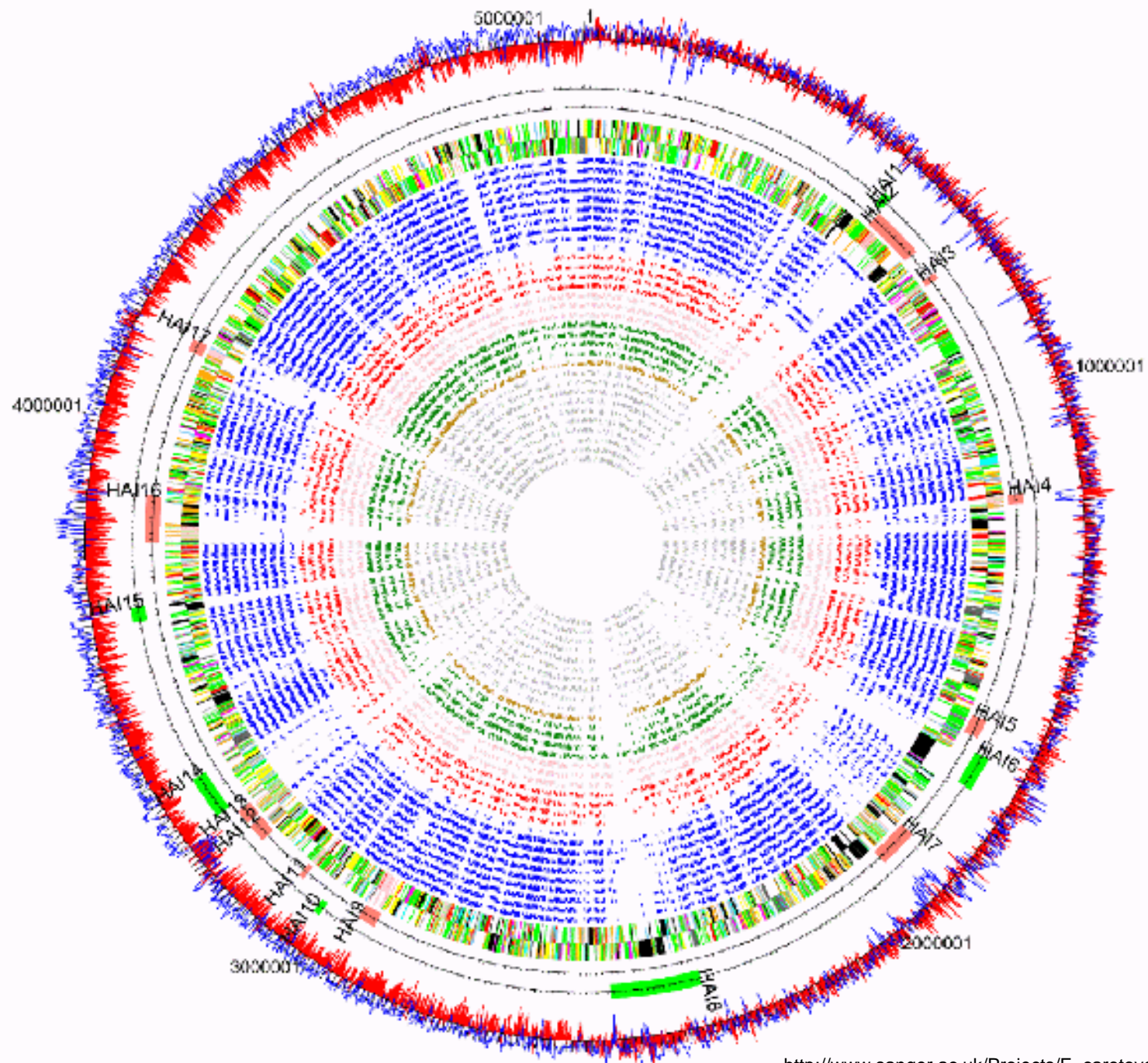
Christian Marck

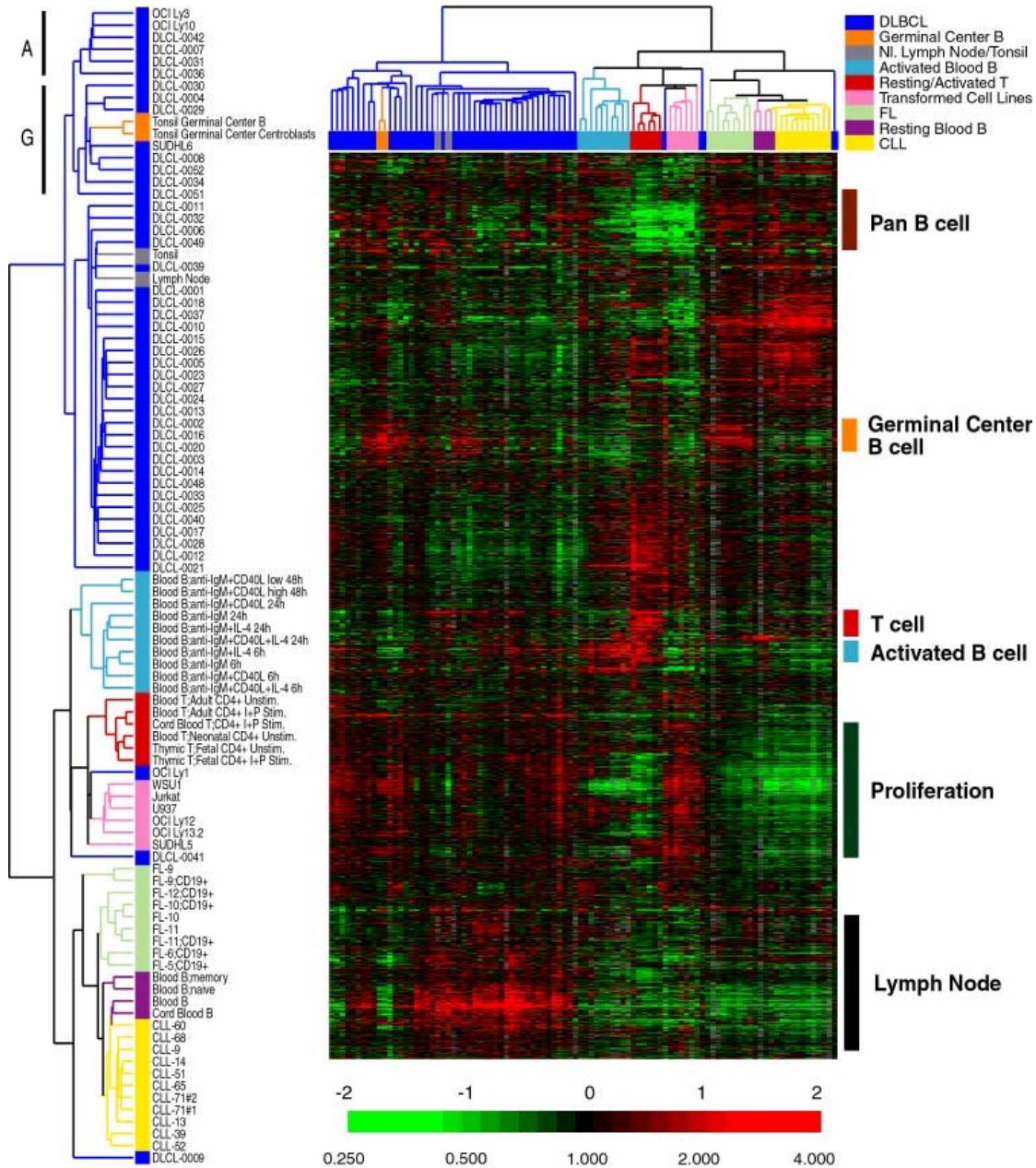
Service de Biochimie, Bâtiment 142, Département de Biologie, Centre d'Etudes Nucléaires de Saclay, 91191 Gif-sur-Yvette Cedex, France

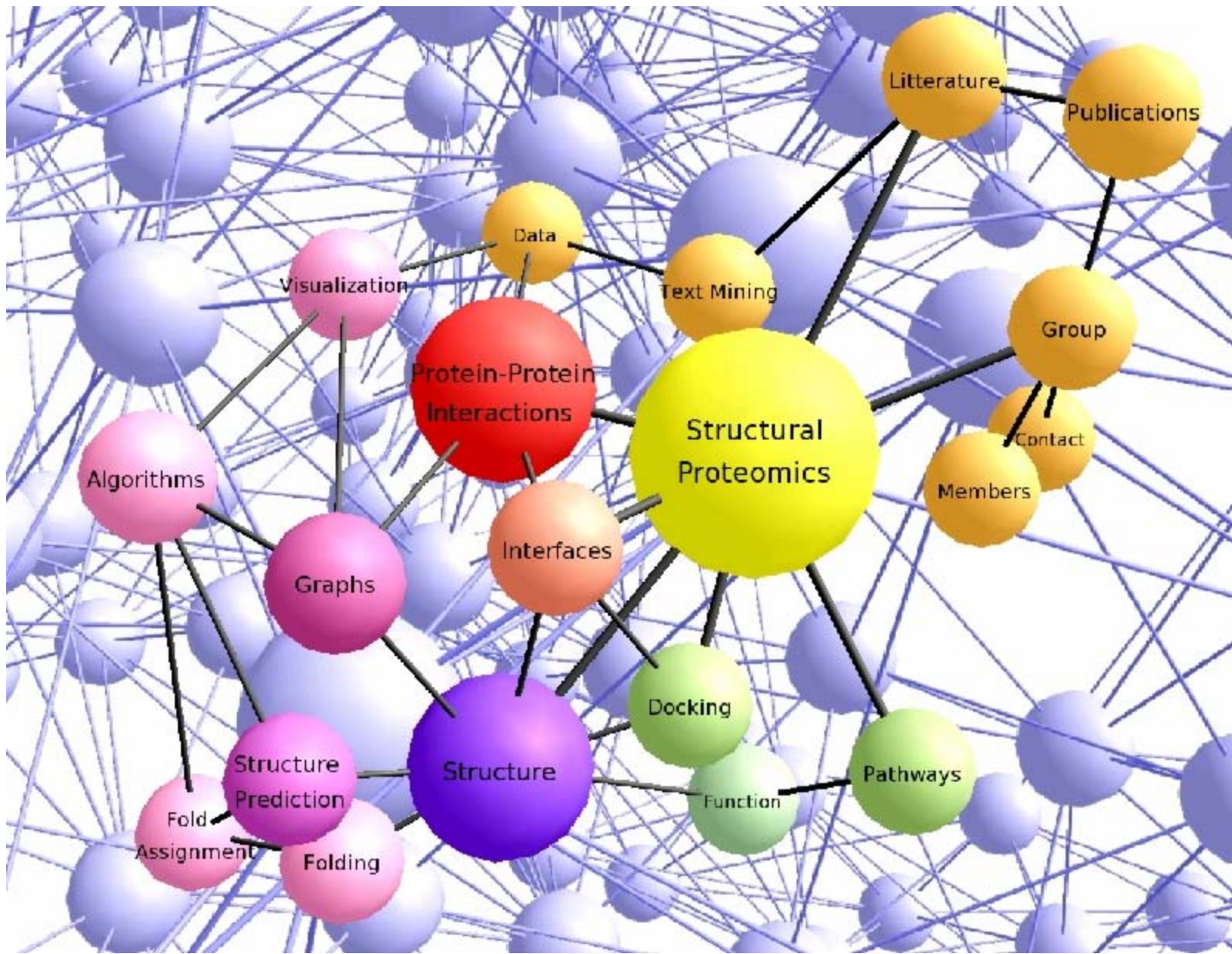
Received August 17, 1987; Revised and Accepted November 15, 1987



280.6 TFLOPS with 131072 nodes

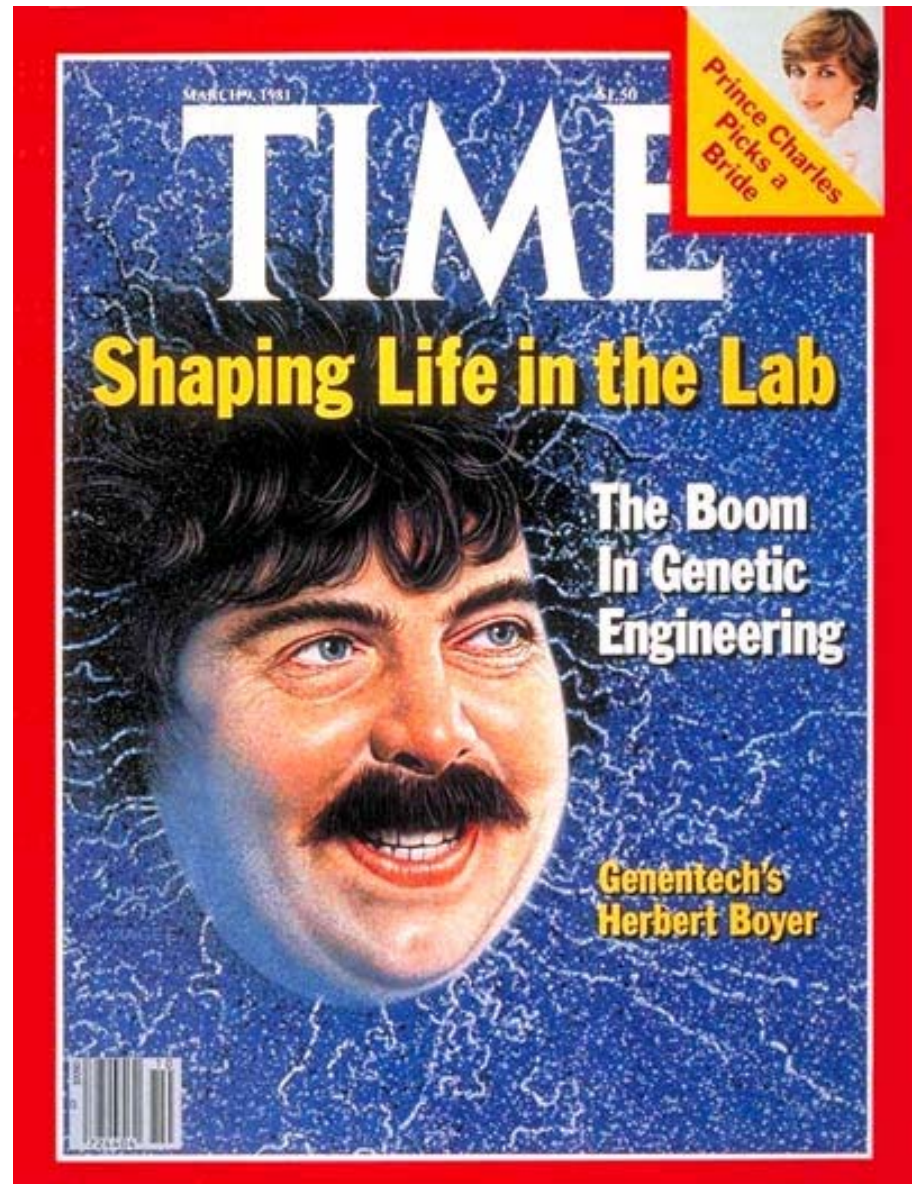
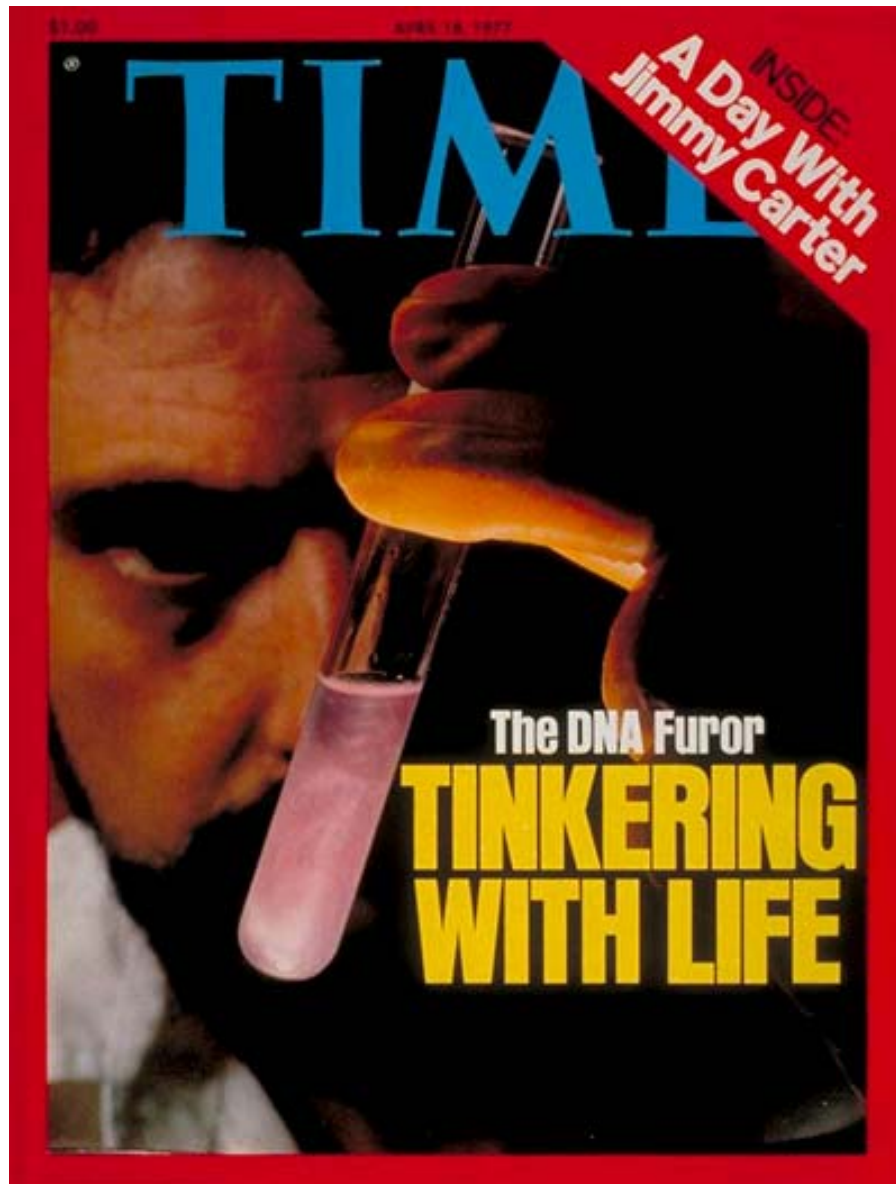






Writing code: synthesis

If we can't build it, we don't understand it.

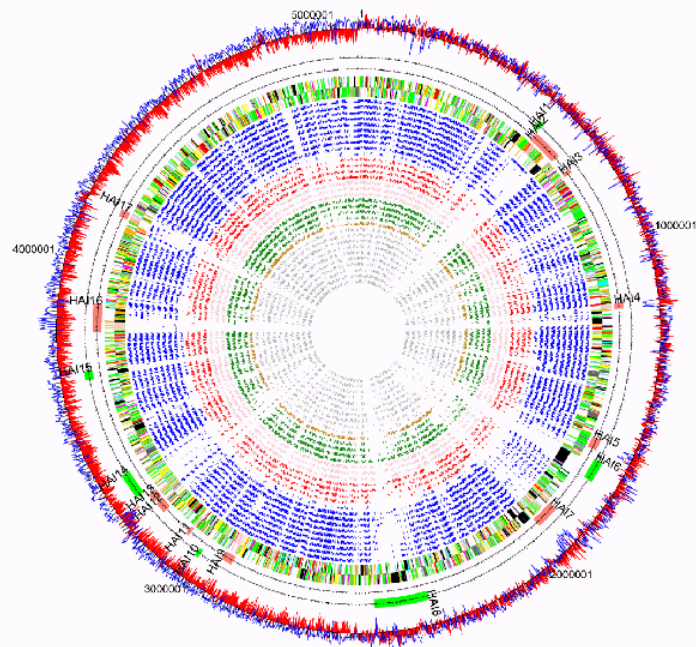




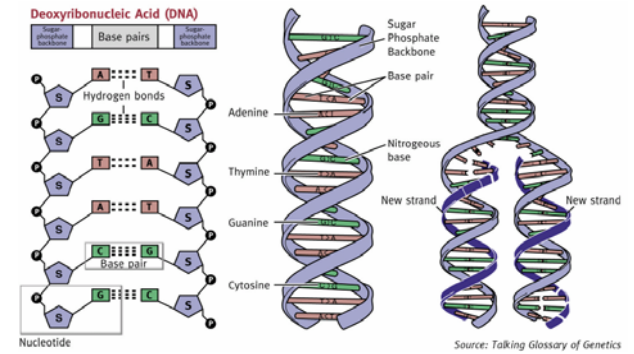
if you can **W**RiTe **D**Na,

You **'rE** **nO** **LONGER** **liMi****TED**

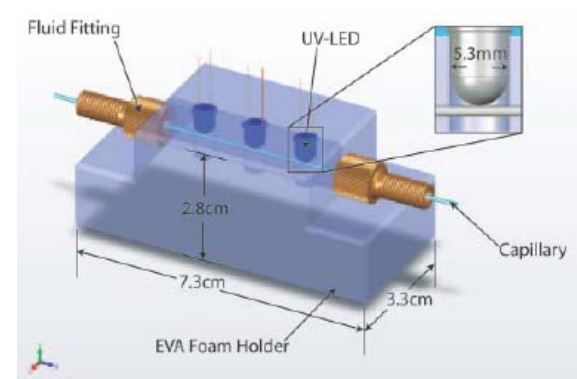
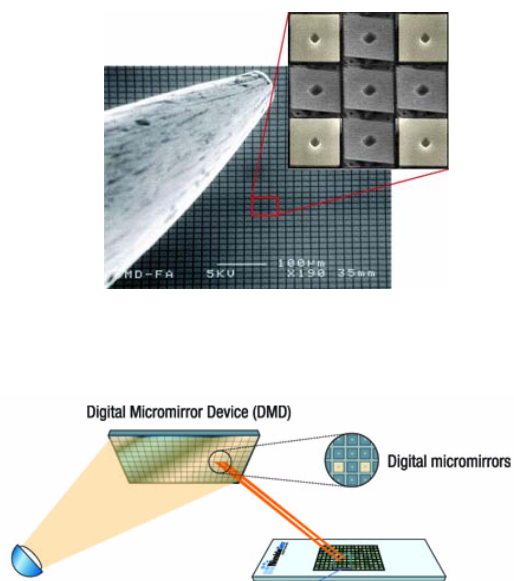
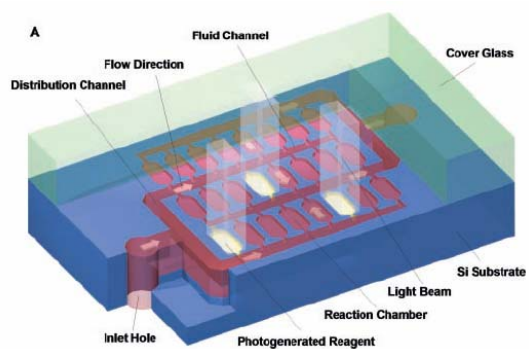
to " **IS** " but **TO** **what** you **could** **MAKE** •

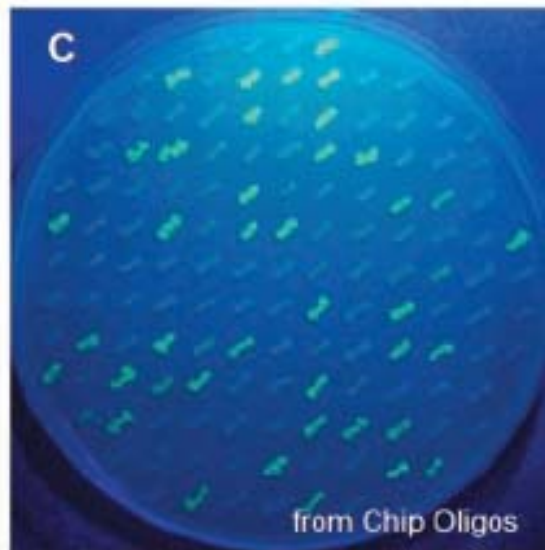
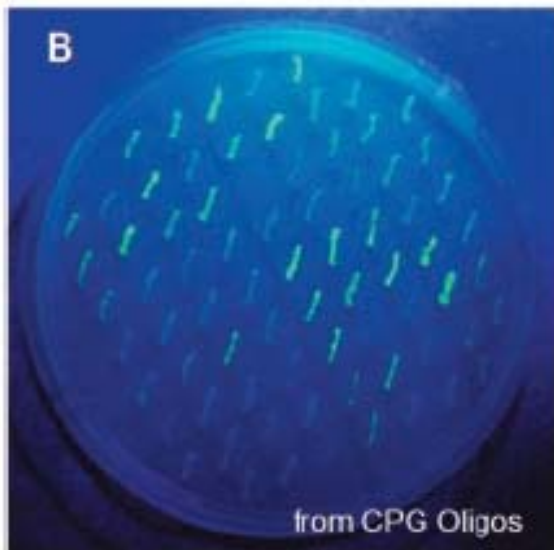


Digital DNA “design”



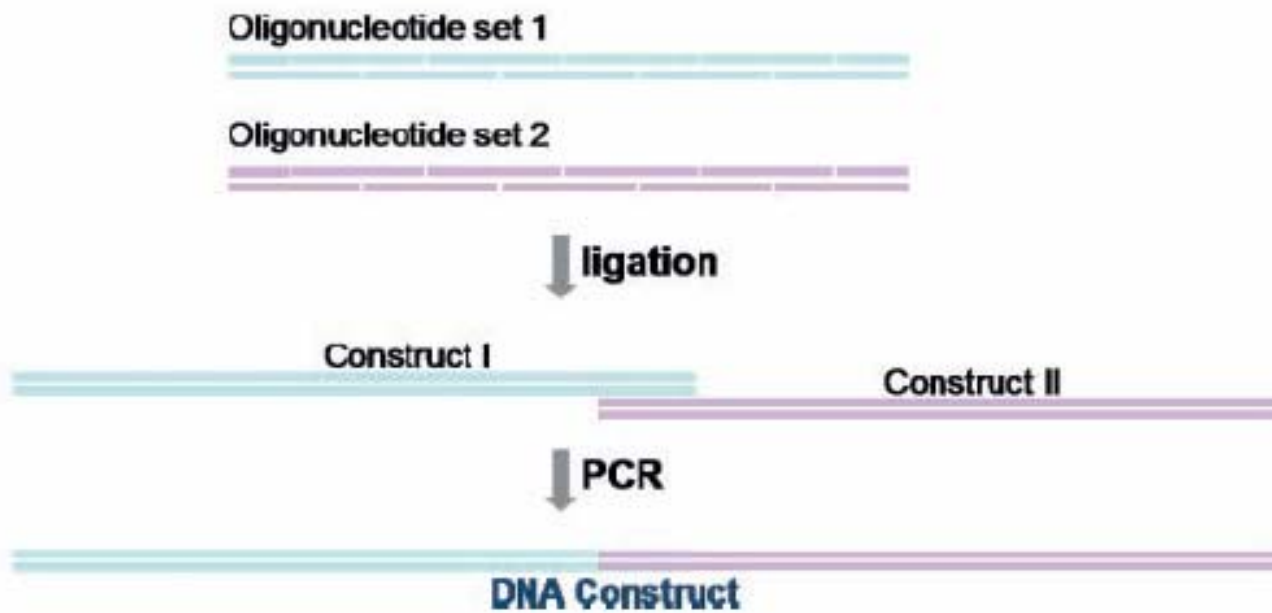
Physical DNA and outputs



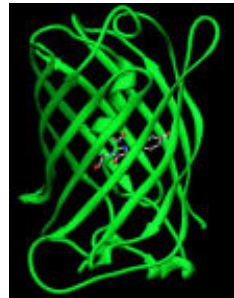


EGFP gene 714 bp

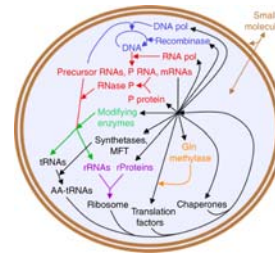
D



Applications dependent on synthetic capabilities

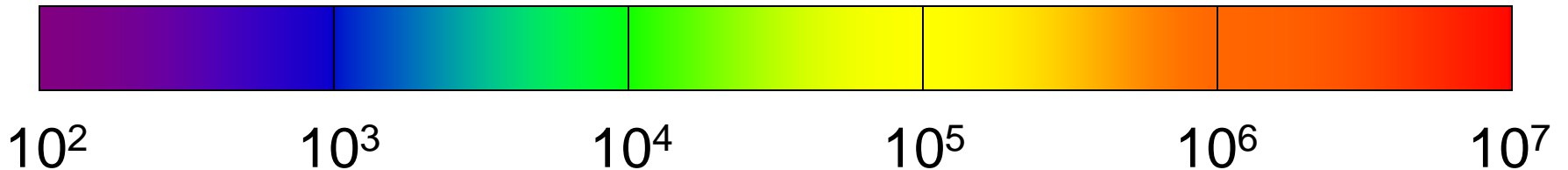


single genes*



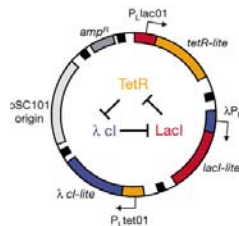
minimal life

base
pairs



genetic circuits, viruses, GEMs

Engineered organisms



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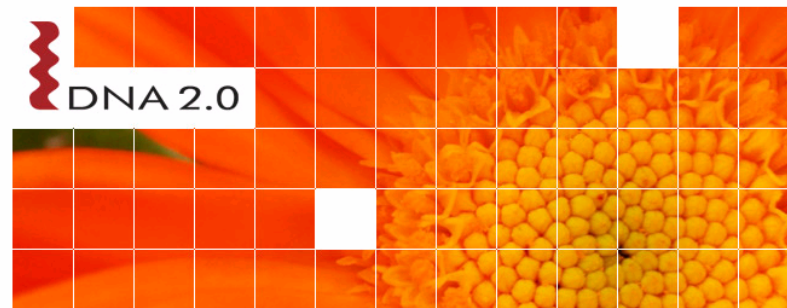
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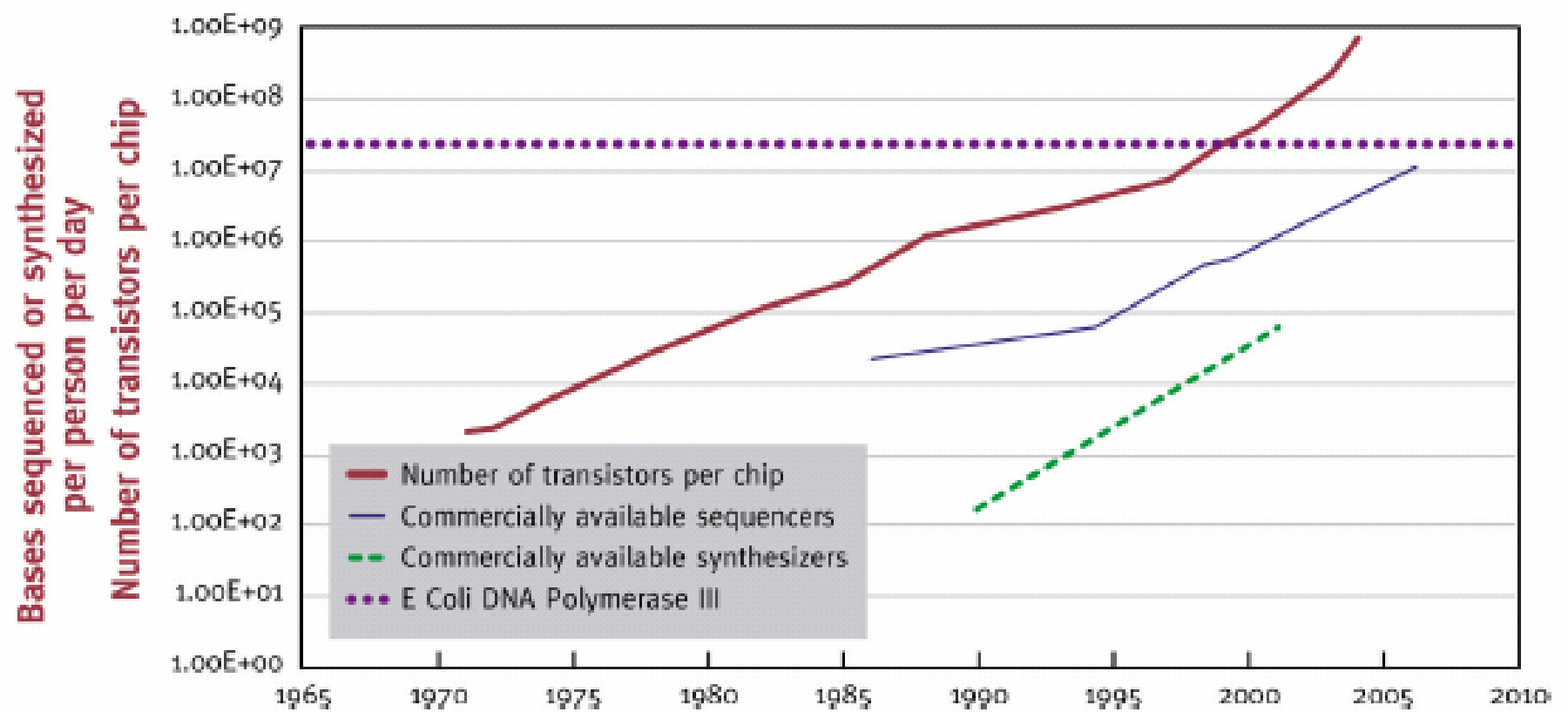


THE BUILDING BLOCKS OF LIFE. BUILT FOR YOU.



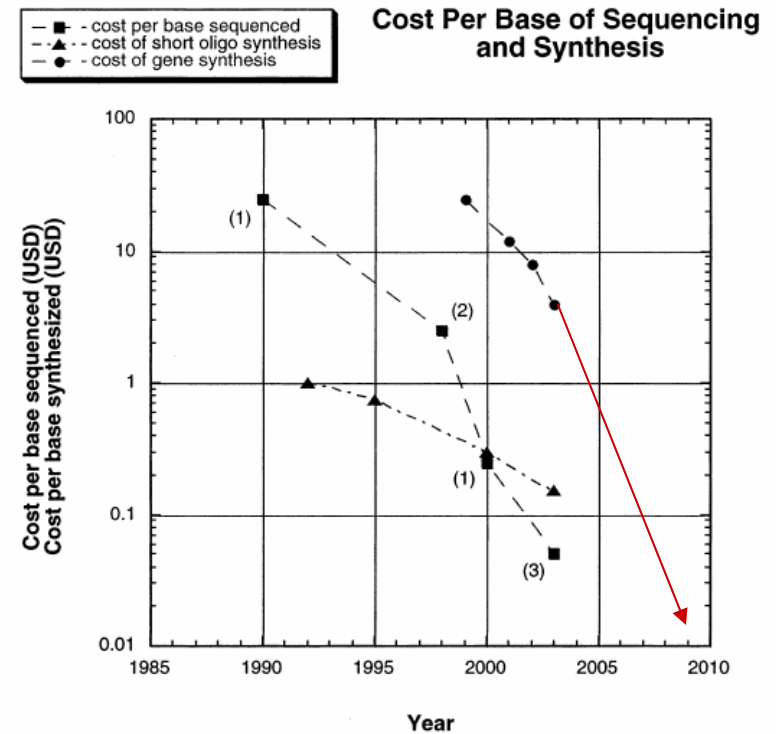
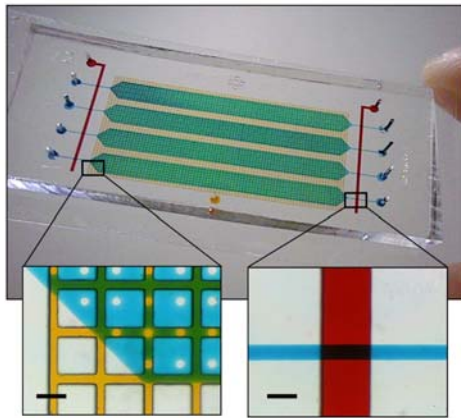
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Source: R. Carlson, Bio-era.

- -5 years: 0.5 - 5kb, \$10-\$15/bp
- 0 years: 50 - 500kb \$0.50-\$1/bp
- +5 years: 5mb - 5gb <\$0.0001/bp

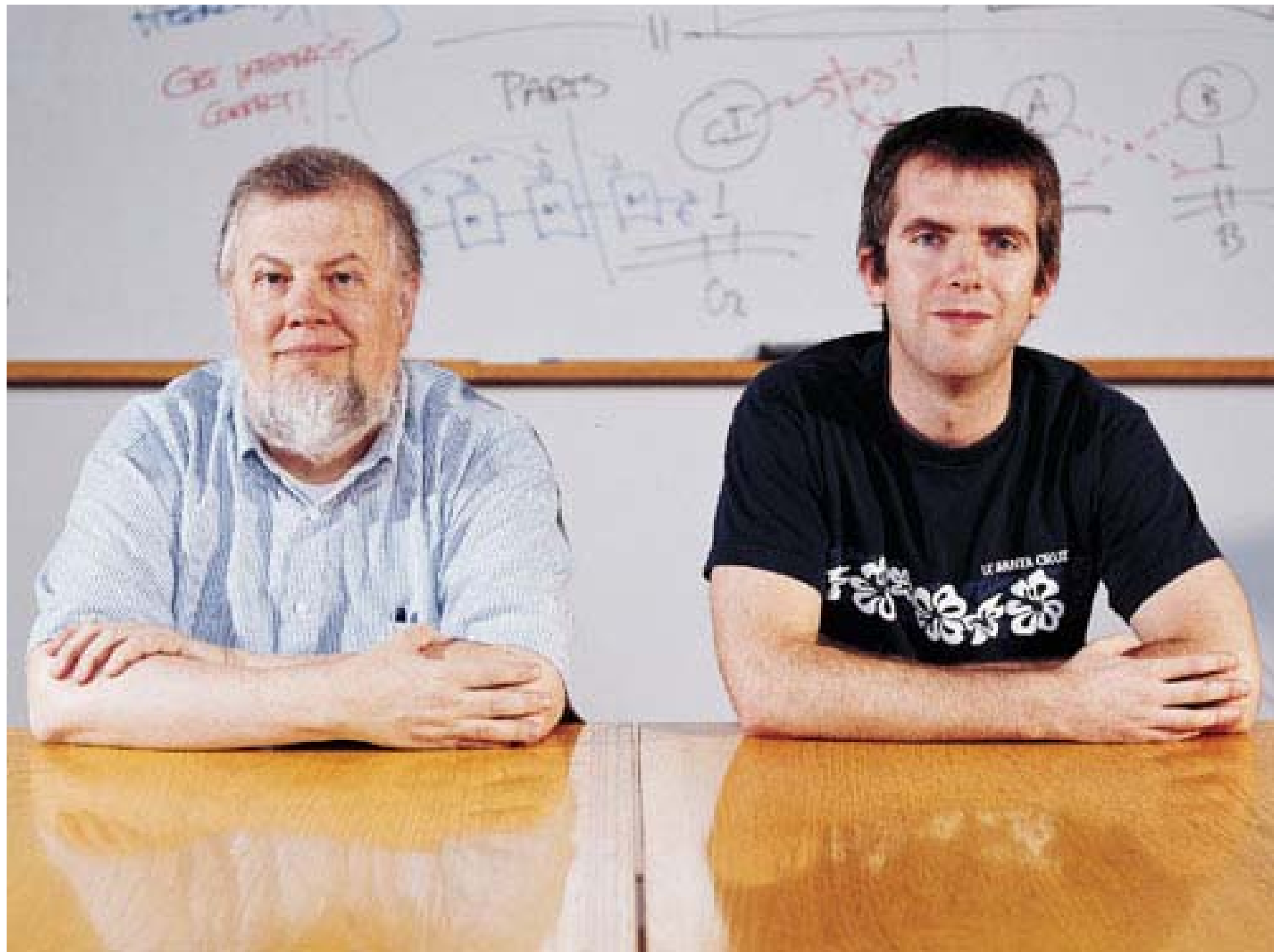


Carlson, R. (2003) The Pace and Proliferation of Biological Technologies

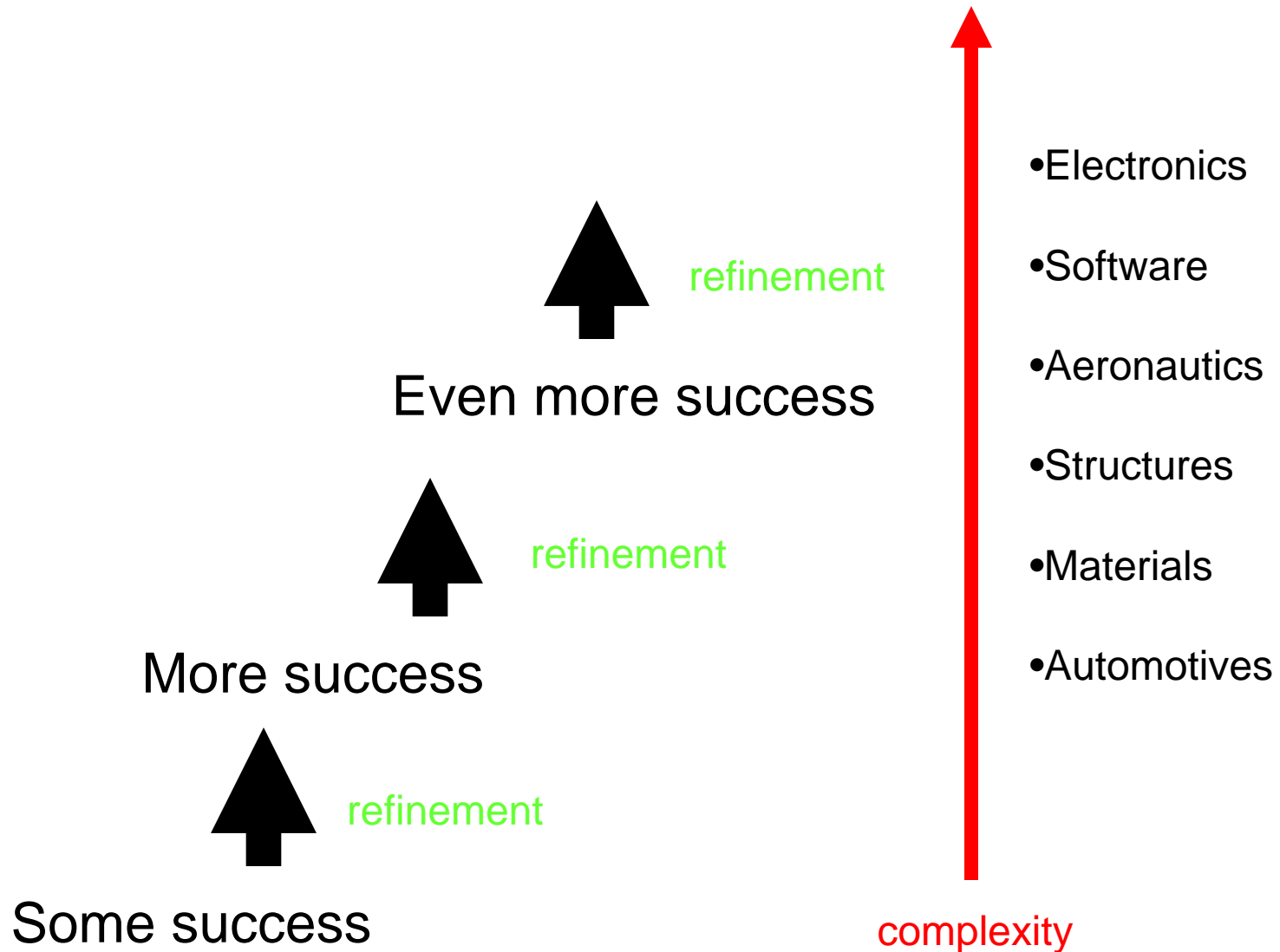


Sources: Source: R. Carlson, G. Epstein, A. Yu (2005)

Engineering philosophy



Engineering process...



F1760

Sender Device

B0015

terminator

Name: B0015

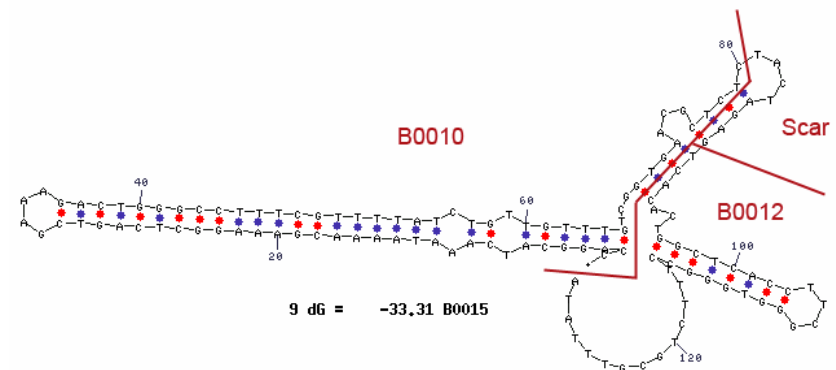
Type: Double terminator

Length 129 bp

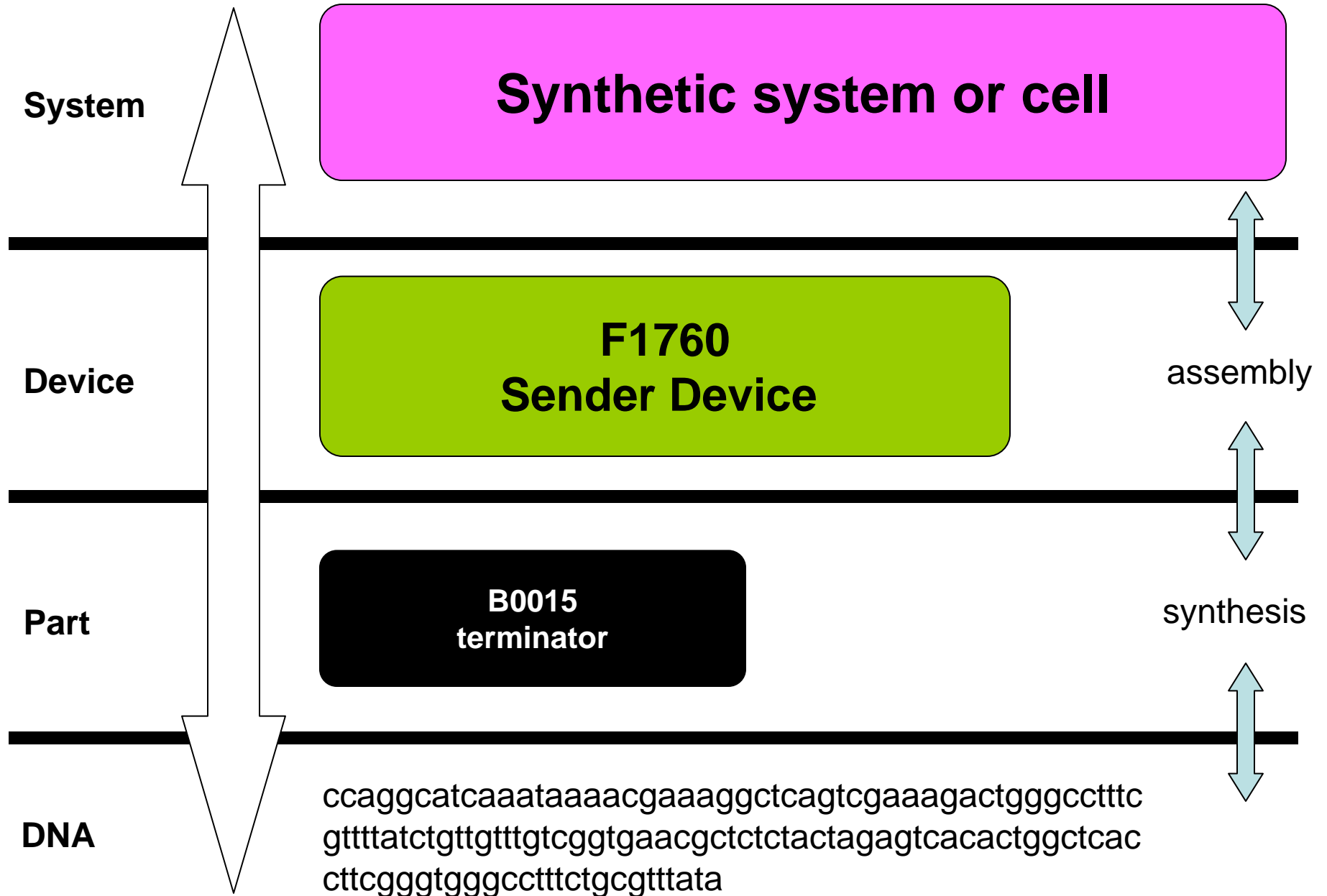
Designed by: Reshma Shetty

Forward efficiency: 0.984

Reverse efficiency: .295



STANDARDIZED DATA





jump to part

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article

Transcriptional Regulators

Available repressible regulators (normally ON) -?-

[Show 0 more parts](#)

[Edit](#)

-?-	Name	Description	Direction	Control -?-	Output Low High	Length
A W	BBa_I14032	promoter P(Lac) IQ	Forward			37
A W	BBa_R0040	promoter (tetR, negative)	Forward	aTc, tetracycline		54
A W	BBa_R0051	promoter (lambda cl regulated)	Forward	lambda cl		49

Available inducible regulators (normally OFF) -?-

[Show 0 more parts](#)

[Edit](#)

-?-	Name	Description	Direction	Control -?-	Output Low High	Length
A	BBa_I12007	Modified lambda Prm promoter (OR-3 obliterated)	Forward	cl		82
A	BBa_R0062	Promoter (luxR & HSL regulated -- lux pR)	Forward	luxR, HSL		55
A	BBa_R0079	Promoter (LasR & PAI regulated)	Forward	PAI		157
A	BBa_R0080	Promoter (AraC regulated)	Forward	araC		149

Available other regulators

[Show 172 more parts](#)

[Edit](#)

-?-	Name	Description	Direction	Control -?-	Output Low High	Length
A W	BBa_I0500	Inducible pBad/araC	Forward	araC, arabinose		1210
A W	BBa_I13453	Pbad promoter				130
A W	BBa_J13002	TetR repressed POPS/RIPS generator	Forward	ATc		74
A W	BBa_J13023	3OC6HSL+LuxR dependent POPS/RIPS generator				117
A W	BBa_J23100	constitutive promoter family member				35
A W	BBa_J23101	constitutive promoter family member				35
A W	BBa_J23102	constitutive promoter family member				35
A W	BBa_J23103	constitutive promoter family member				35
A W	BBa_J23104	constitutive promoter family member				35
A W	BBa_J23105	constitutive promoter family member				35
A W	BBa_J23106	constitutive promoter family member				35
A W	BBa_J23107	constitutive promoter family member				35
A W	BBa_J23108	constitutive promoter family member				35
A W	BBa_J23109	constitutive promoter family member				35
A W	BBa_J23110	constitutive promoter family member				35
A W	BBa_J23111	constitutive promoter family member				35

<http://parts.mit.edu>

BBa_F2620

3OC₆HSL → PoPS Receiver

http://parts.mit.edu/registry/index.php/Part:BBa_F2620



Authors:
Barry Canton [bcanton@mit.edu]
Anna Labno [alabnoa@mit.edu]

Last Update: 15 January 2007

Description

A transcription factor (LuxR, BBa_C0062) that is active in the presence of cell-cell signaling molecule 3OC₆HSL is controlled by a TetR-regulated operator (BBa_R0040). Device Input is 3OC₆HSL. Device output is PoPS from a LuxR-regulated operator. If used in a cell containing TetR then a second input signal such as aTc can be used to produce a Boolean AND function.

Characteristics

Input Swing: 1E-9 to 1E-6 M 3OC₆HSL, exogenous

Output Swing: 0±1 to 503±1 GFP molecules cfr⁻¹ s⁻¹

Switch Point: 7±1 nM 3OC₆HSL, exogenous

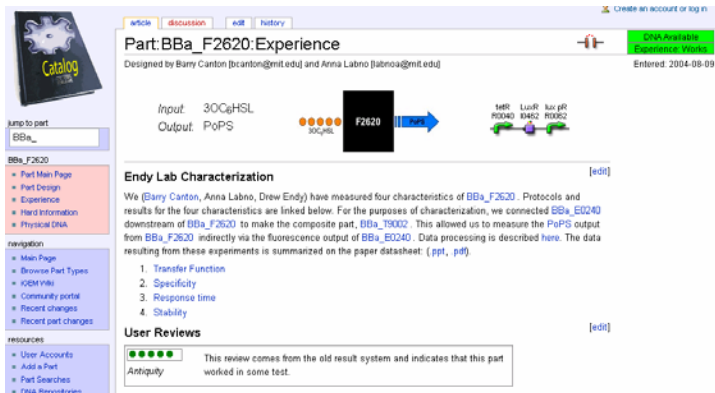
LH Response: 9 min (t_{50%}), 27 min (t_{90%})

Key Parts

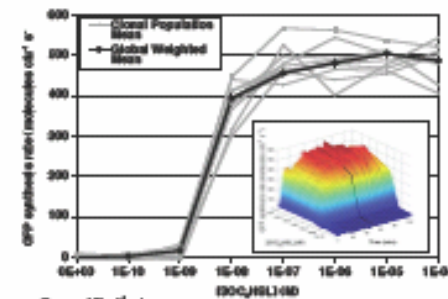
BBa_R0040: TetR-regulated operator

BBa_C0062: luxR ORF

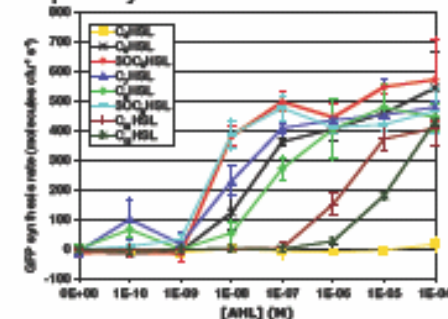
BBa_R0002: LuxR-regulated operator



Transfer Function*



Specificity*



Demand (low/high input)

Translational: 256/8048 ribosomes cfr⁻¹
3.8E3/1.2E5 charged tRNA cfr⁻¹ s⁻¹

Compatibility

Chassis: Compatible with MC4100, MG1655, and DHSα

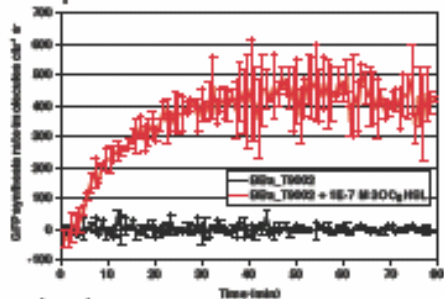
Plasmids: Compatible with pSB3K3 and pSB1A2

Devices: Compatible with E0240, E0430 and E0434

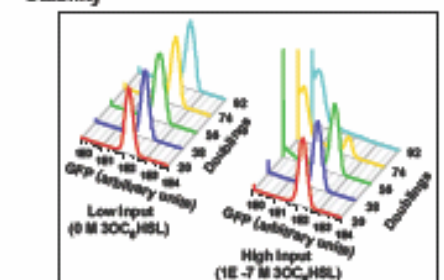
Crosstalk: with systems containing TetR (C0040)

Signaling: Crosstalk with input molecules similar to 3OC₆HSL

Response Time*



Stability**



Stability (low/high input)

Genetic: >92/74 replication events**

Performance: >92/74 replication events**

Conditions (abridged)

Output: Indirect via BBa_E0240

Vector: pSB3K3

Chassis: MG1655

Culture: Supplemented M9, 37°C

***Equipment:** PE Victor3 plate reader

****Equipment:** BD FACScan cytometer

Registry of Standard Biological Parts

making life better, one part at a time

License: Public

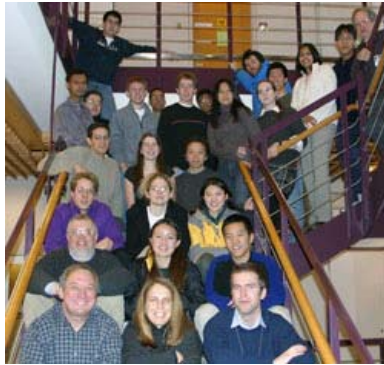
Signaling Devices



Shares:

- DNA parts
- DNA code
- Protocols
- Experience
- Publications





2006 Jamboree – 400 gengineers

Designs on life

Earlier this month, students from around the world locked horns in competition. Their challenge was to build functioning devices out of biological parts. Erika Check finds out how they got on.

Even if you're thinking big, you usually have to start small. Especially, as a group of Swiss students found, when big means counting to infinity. The team was drawing up a blueprint for the world's first counting machine made entirely of biological parts. Although they had their sights on lofty numbers, they opted to go no higher than two. If the plan worked, it would be a proof-of-principle for a much larger tallying device.

The group, from the Federal Institute of Technology (ETH) in Zurich, was one of 17 teams unveiling their projects at the first international Intercollegiate Genetically Engineered Machine (iGEM) competition, held at the Massachusetts Institute of Technology (MIT) in Cambridge on 5 and 6 November. The event attracted students from all over the world to design and build machines made entirely from biological components such as genes and proteins. They drew up grand designs for bacterial lach-2-shakes, phorbol-1-shakes, thermometers and sensors. And if none of the designs succeeded completely, that was more because of the limitations of the nascent science of synthetic biology than any lack of enthusiasm, creativity or hard work.

Synthetic biology aims to merge engineering approaches with biology. Researchers working at the most basic level are copying simple biological processes, such as the production of a protein from a gene. They break the process down into its component elements, such as a gene and the pieces of DNA and other molecules that control its activity. They then string these elements together to build a module they know will behave in a particular way — say, oscillate between producing and not producing a protein, or produce a protein that can switch another module on or off.

It is these kinds of components — oscillators and switches — that engineers order from suppliers and link together to build more complex electronic circuits and machines. Synthetic biologists are trying to develop a similar array of biological components, dubbed BioBricks, that can be inserted into any genetic circuit to carry out a particular function. Scientists at MIT have established a Registry of Standard Biological Parts, a catalogue of BioBricks that theoretically



Bidding for glory: teams from the ETH Zurich (top), Cambridge, UK, (bottom right) and Massachusetts at the first international Intercollegiate Genetically Engineered Machine competition.

selection of designs. Students from the University of Cambridge, UK, tried to make a circuit that could control the movement of *Escherichia coli* bacteria. They aimed to engineer the bacteria to contain a switch governing their sensitivity to the sugar maltose. With the switch off, the microbes would ignore the sugar. Tipping

the switch on would cause the bacteria to move toward the sugar.

Competition: Much of the student iGEM competition is about building a machine that can perform a specific task.



COVER STORY

By Carl Zimmer

Scientist of the Year

Jay Keasling is developing ways to program DNA as easily as people program computers.



BRIEF COMMUNICATIONS

Engineering *Escherichia coli* to see light

These smart bacteria 'photograph' a light pattern as a high-definition chemical image.

We have designed a bacterial system that is switched between dormant states by red light. The system consists of a synthetic sensor kinase that allows a lawn of bacteria to function as a biological film, such that the projection of a pattern of light on to the bacteria produces a high-definition (about 100 megapixels per square inch), two-dimensional chemical image. This spatial control of bacterial gene expression could be used to 'print' complex biological materials, for example, and to investigate signalling pathways through precise spatial and temporal control of their phosphorylation state.

Plants and some bacteria use a class of protein photoreceptors known as phytochromes to control photomorphogenesis and the production of protective pigments¹. Photoreceptors are not found in eukaryotes, such as *Escherichia coli*, so we created a light sensor that functions in *E. coli* by engineering a chimera that uses a phytochrome from a cyanobacterium.

A phytochrome is a two-component system that consists of a membrane-bound, extracellular sensor that responds to light and an intracellular response-regulator². The response-regulator of most phytochromes do not have DNA-binding domains and do not directly regulate gene expression, so we fused a cyanobacterial photoreceptor to an *E. coli* intracellular histidine kinase domain (Fig. 1a, and see supplementary information). This design was based on the well-studied *E. coli* EnvZ-OmpR two-component system, which normally regulates porins expressed in response to osmotic shock³. The EnvZ histidine kinase domain has been used for the construction of functional chimeras^{4,5}, and a plant phytochrome has

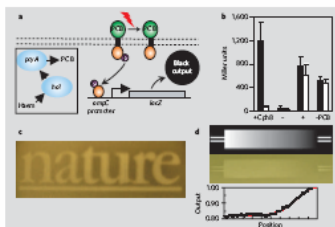


Figure 1 Light imaging by using red fluorescent cells. **a**, The chromatic light receptor Cph1 contains the photoreceptor from *C. crescentus* (Cph1) and the histidine kinase and response-regulator from *EnvZ*-OmpR (for simplicity, conversion of EnvZ to phytochrome (PCB), which forms part of the photoreceptor). Red light drives the sensor to a state which strongly phosphorylates the response-regulator, turning off gene expression. For details of genes, see text. **b**, Miller assay showing that Cph1 is activated by red light (red bar) in the presence of PCB and inactive in the light (white bar). There is a light-dependent activity in the absence of Cph1 (—) which is a constitutive activity when the bacteria have been grown in the presence of PCB. **c**, When an image is projected on to the bacterium, the EnvZ reporter is expressed. **d**, Transient function of the circuit. As the intensity of the light is increased, the circuit output gives a graded response.

enzymatically produces a black compound. The part of the photoreceptor that responds to light, phytochrome, is not naturally produced in *E. coli*. We therefore introduced two phytochrome-like domains (the histidine kinase and response-regulator) but converted them into phytochromes (parts 1, 1150A, B, 1150B, MIT Registry of Standard Biological Parts) (Fig. 1a, inset). Individual Cph1-EnvZ chimeras were then fused at 37 °C for 4 h with broad-spectrum light and assayed for expression of lacZ reporter. The chimera Cph1-1150B produced a particularly strong response to light (Fig. 1b).

For bacterial photography, we grew a lawn of bacteria on agar. The lacZ reporter was activated by addition of 5-gal (5,4-cyanoheptatriene-β-D-galactopyranoside). LacZ types the formation of a stable, insoluble, X precipitate from 5-gal. Light exposed a response in the bacteria, giving a 6-contrast replica of the applied image on

the biological film, as appeared light and dark (Fig. 1c, and see supplementary information). The lacZ activity shows the resulting light image in the brightest light (Fig. 1c, inset). Our creation of a non-invasive imaging of the power and accurate methods available in the synthetic biology. The light regulation should be applied to and used in various applications in synthetic biology.

Andrew Linker, Aaron J. Silver, Zachary Smith, Lawrence, Matthew Lowy, Alexander Sorensen, and Edward M. Marcovitz, *E. coli*



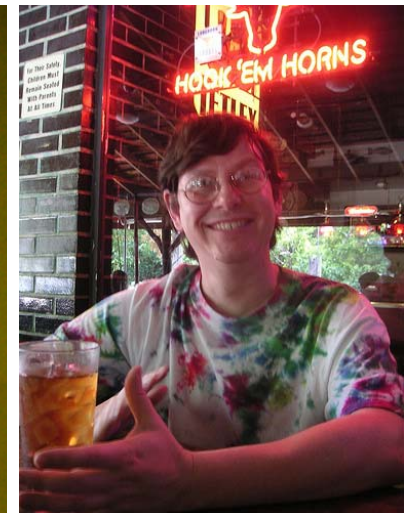
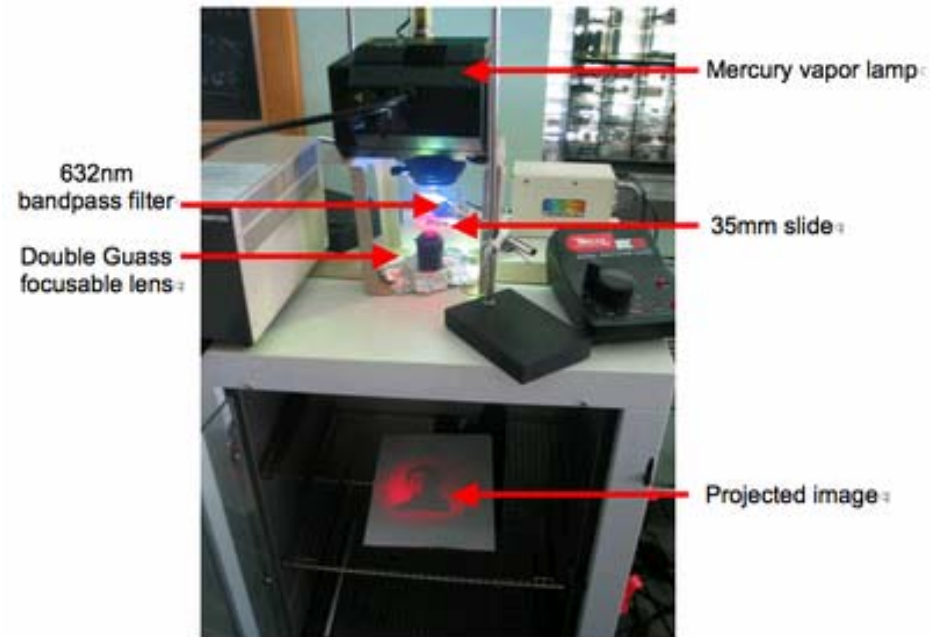
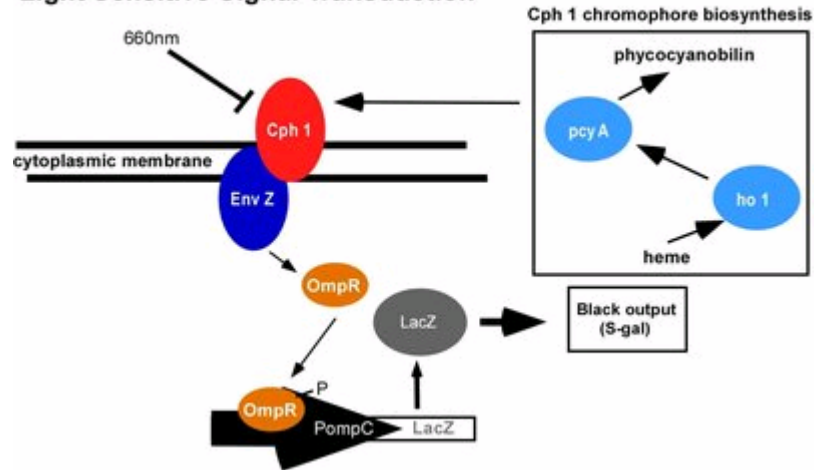
Genetic 'Jamboree' draws innovators

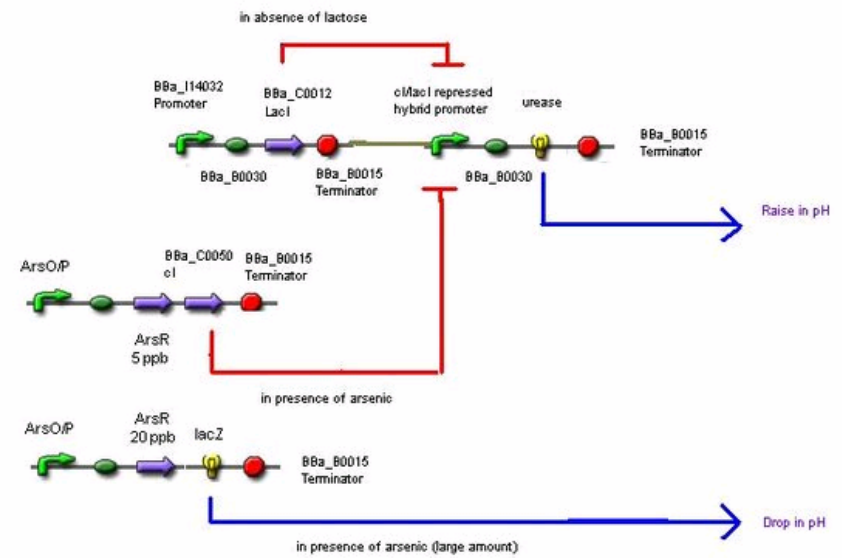
Science students the world over share research



At MIT's International Genetically Engineered Machine Competition yesterday, the audience listened to a presentation on synthetic biology. (John Tlumacki/Globe Staff)

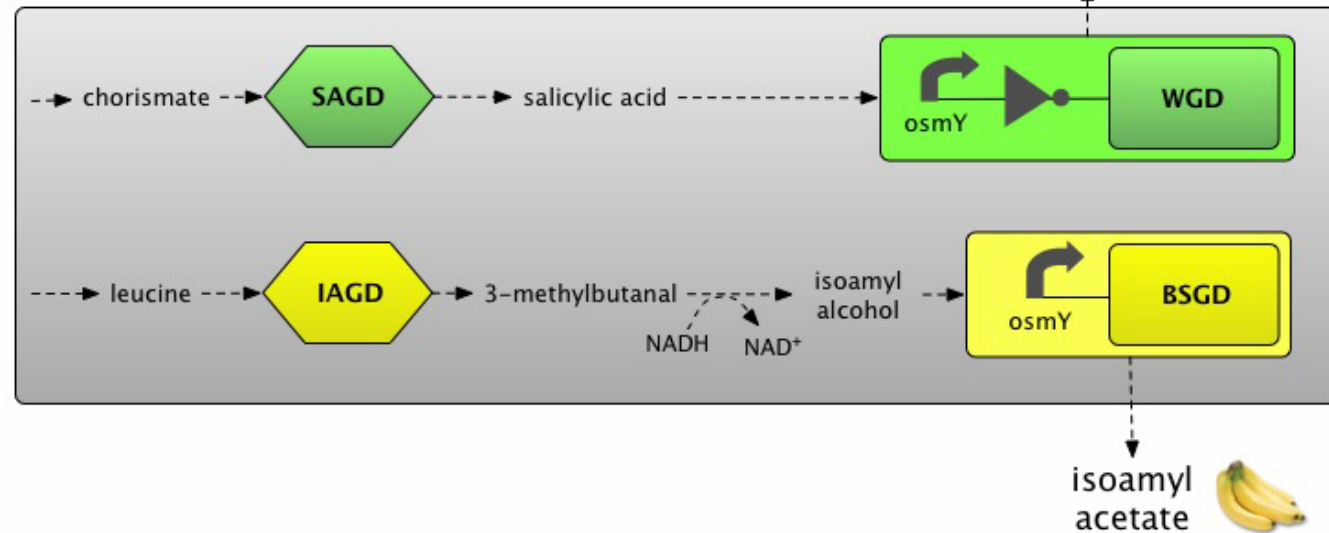
Light Sensitive Signal Transduction







indole deficient tnaA5⁻ chassis



iGEM 2007

- 57 teams – 20 countries
- USA (26)
- Scotland (3)
- Colombia
- Italy (2)
- Mexico
- Taiwan
- Russia
- Germany
- South Africa
- Middle East
- Canada (6)
- Japan (2)
- Australia
- England
- Switzerland
- China (4)
- Spain
- India
- France
- **Slovenia**

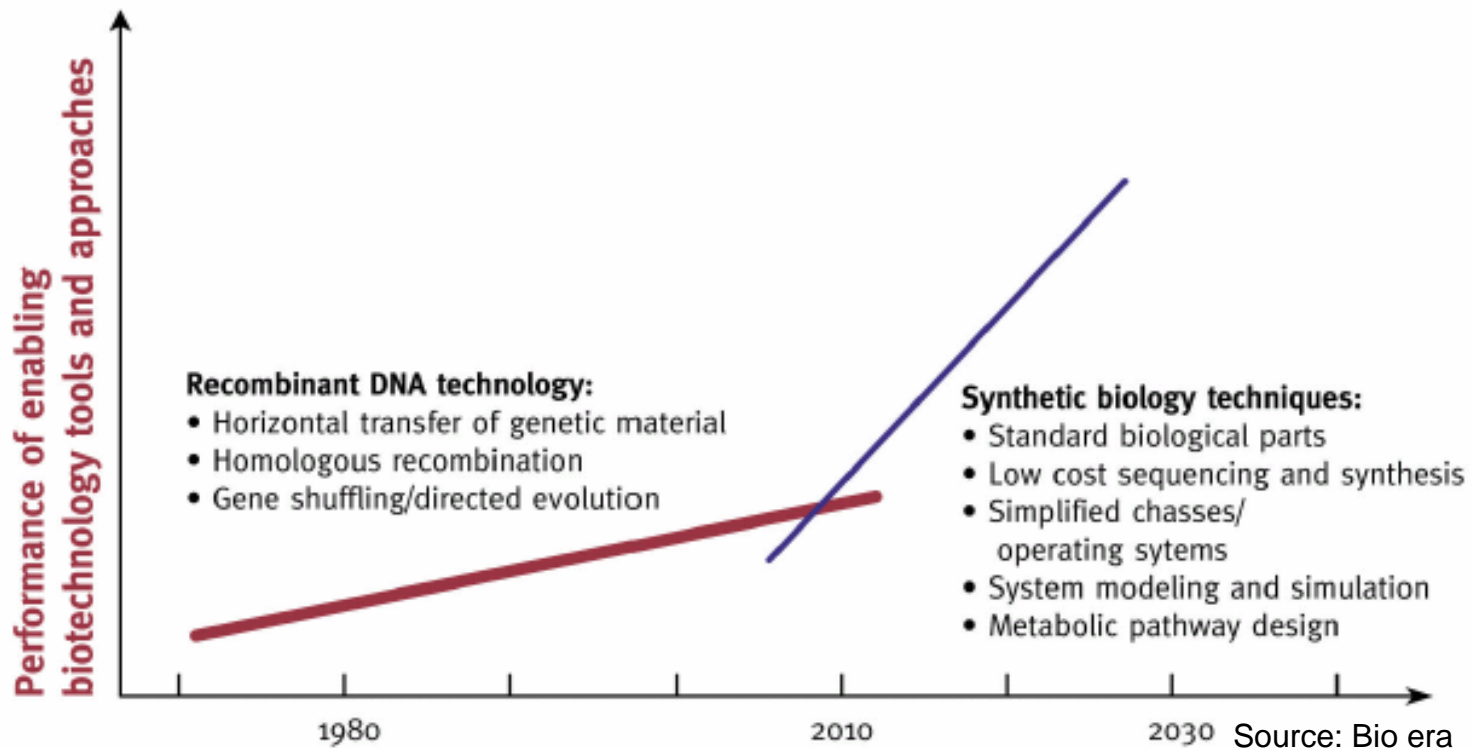
U of A iGEM Team

- Biobutanol project – “*Plan B*”
- Moving metabolic pathway from *Clostridium* into *E. coli*



Opportunities for Alberta

Synthetic biology is going to grow fast!



PIMP MY GENOME!

SYNTHETIC BIOLOGY:
A PLAN FOR ENGINEERING BIOLOGY



Wednesday April 4, 2007

University of Alberta, Telus Centre, Room 150
111 Street and 87 Avenue
Edmonton, AB

Doors open and refreshments - 3:00 PM

Presentation - 3:30 PM

*Complimentary return bus transportation will be provided
to guests from the University of Calgary.*

Join Drew Endy, a leader in synthetic biology from MIT, for an engaging look at how biological engineering is changing. Find out how the latest advances in this new era of biology are helping make R&D faster, cheaper and easier.

Drew is a fellow in the Department of Biology and the Biological Engineering Division at MIT. He co-founded the MIT Synthetic Biology working group and the Registry of Standard Biological Parts. He is also co-founder of iGEM, the International Genetically Engineered Machine competition, Codon Devices Inc., a venture-funded startup that is working to develop next-generation DNA synthesis technology, and the BioBricks Foundation, a not-for-profit organization that is working to develop legal and economic strategies needed to support open biotechnology. Drew's work has been featured in *The Economist*, *Forbes*, *Wired*, *Schematic American* and the *New York Times*.

For registration and more information,
please visit www.albertaingenuity.ca.



The Alberta Ingenuity Fund supports science and engineering research of the highest calibre to create a prosperous future for the province. It draws funding from a \$7 billion endowment established and managed by the Government of Alberta to build the capacity for innovation, especially in areas with long lasting social and economic impact.

Engineering synthetic biological constructs will become the foundational technology of the 21st century Tom Knight, MIT, SB3

- Biology > Physics (\$, staff, discoveries)
- Biology is more important than physics, as measured by its economic outputs, ethical implications, and effects on human welfare
- Alberta already *has* a vibrant bio-economy
- Well-positioned to become a global leader in synthetic biotechnologies *is we act quickly and decisively*

Start education programs...



A JOINT PROGRAM OF THE CALIFORNIA INSTITUTE FOR QUANTITATIVE BIOMEDICAL RESEARCH (QB3)
AND LAWRENCE BERKELEY NATIONAL LABORATORY (LBNL)

The California Institute for Quantitative Biomedical Research (QB3) and Lawrence Berkeley National Laboratory (LBNL) have joined forces to accelerate the growth of synthetic biology, a new field that promises major new advances in preventing and treating disease, generating new energy sources, and preventing and mitigating environmental threats.

Opening in spring 2005 in a spacious, modern building in west Berkeley, the Berkeley Center for Synthetic Biology gives renowned scientists and engineers the chance to pool their talents and collaborate in new ways with enormous potential benefits for California's advances in biomedicine and economic growth.

the control and design of
id new organisms to solve a
energy, and environmental
solved using naturally
as. The inherently



QB3 and LBNL scientists occupy lab space in a building renovated in 1997 for biotech research, previously leased by Bayer, featuring large labs, viral suites, and tissue culture rooms. UCSF Mission Bay and numerous biotech firms are nearby.

MIT establishes groundbreaking biological engineering major

February 17, 2005

The Massachusetts Institute of Technology faculty yesterday approved a new course of study for undergraduates, in biological engineering, the first entirely new curriculum established at the Institute in 29 years.

California Institute for Quantitative Biomedical Research



2/1/2007 - BP awards \$500 million bioenergy grant

Global energy firm BP has selected UC Berkeley, in partnership with Lawrence Berkeley National Laboratory and the University of Illinois, to lead an unprecedented \$500 million research effort to develop new sources of energy and reduce the impact of energy consumption on the environment. The funding will create the Energy Biosciences Institute, which initially will focus its research on biotechnology to produce biofuels. QB3 helped coordinate the research proposal and will help administer the project. [More](#) >



6/26/2007 - Bay Area partnership to host DOE bioscience center

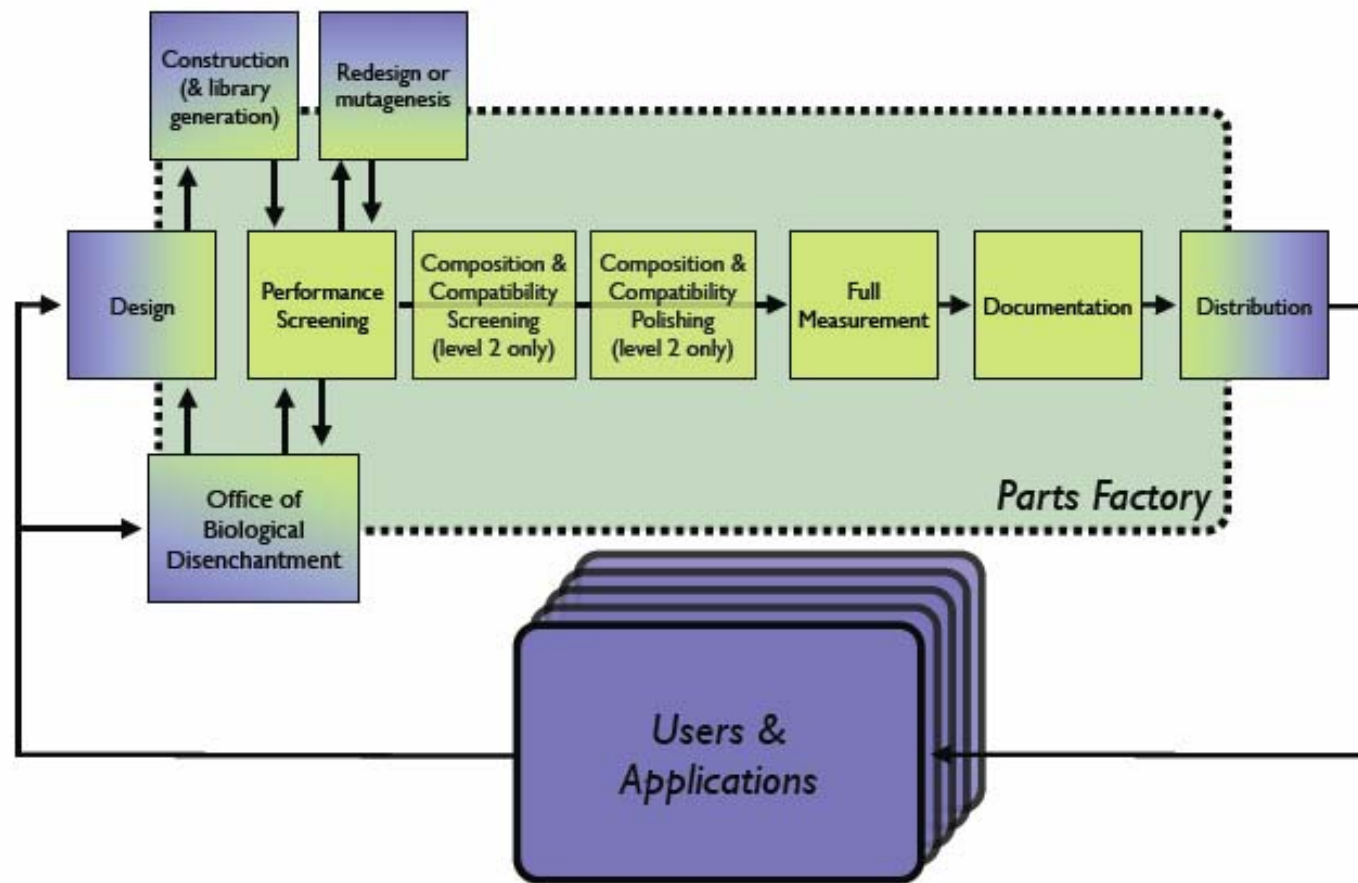
The U.S. Department of Energy has announced the creation of a new bioenergy research center, with UC Berkeley and Lawrence Berkeley National Laboratory as two of its six collaborating institutions. The Joint BioEnergy Institute – to be headquartered in the East Bay and led by Jay Keasling – will receive approximately \$125 million in DOE funding over five years. [More](#) >



8/3/2006 - New center poised to transform biotech

Aided by a \$16 million NSF grant, QB3 has launched the Synthetic Biology Engineering Research Center at UC Berkeley, with collaborators at UC San Francisco, MIT, Harvard, and Prairie View A&M University. Researchers hope to make it as quick and easy to engineer biology as it now is to assemble microprocessors, hard drives, and memory chips into a computer. [More](#) >

Build a bio-fab to support research community and next-gen companies





Monday, April 09, 2007

DNA Factories

Cheaply churning out made-to-order DNA could revolutionize molecular biology.

By Emily Singer



LS9, INC.

the renewable petroleum company™





Realizing the Promise of Synthetic Biology

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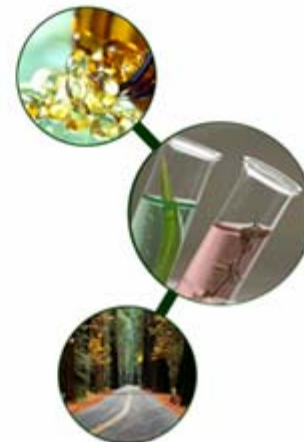
[NEWS](#)

[CAREERS](#)

Welcome



Amyris Biotechnologies is translating the promise of synthetic biology into solutions for real-world problems. Building on advances in molecular, cell and systems biology, we are engineering microbes capable of producing high-value compounds to address major global health and energy challenges. We are employing these living chemical factories to produce novel pharmaceuticals, renewable fuels, and specialty chemicals.



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- *Applications (health, biofuels, bioproducts)
- Software: Metabolic and genomic design tools
- Hardware: Advanced synthesis hardware, biological test and measurement devices
- Ethics and social policy of synthetic biology
- Educational program development
- Next-generation biotechnology company development



MAY 3, 1982

\$1.50

TIME

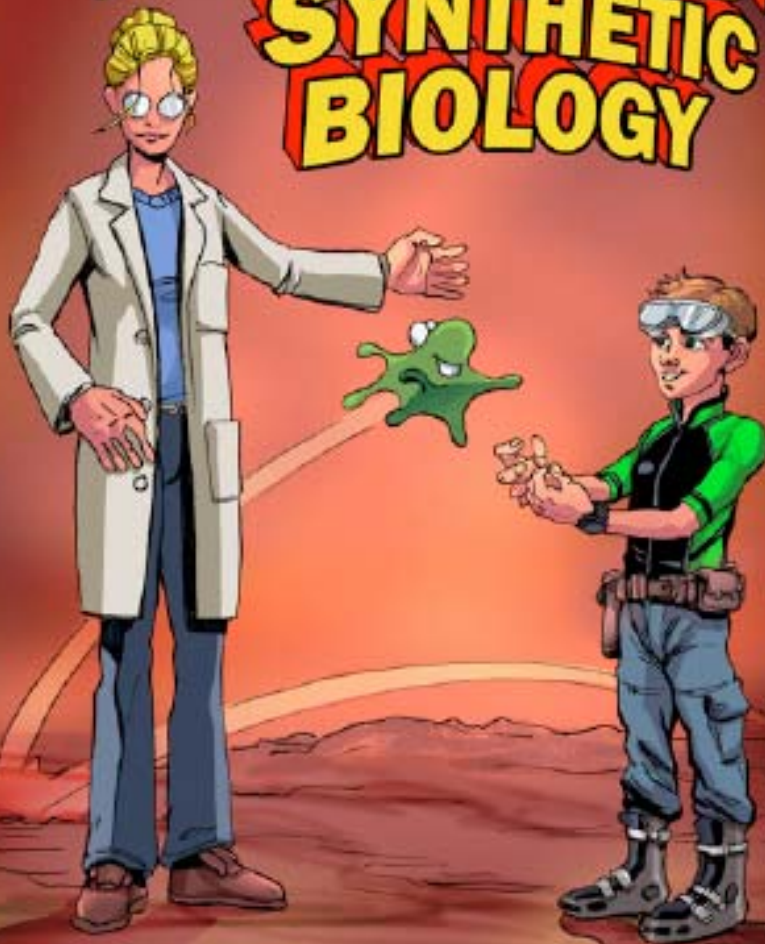
COMPUTER GENERATION

A New Breed of Whiz Kids



-3%
INFLATION VANISHES!
At Least for
A Month

ADVENTURES IN SYNTHETIC BIOLOGY



STORY: DREW ENDY ISADORA DEESE
THE MIT SYNTHETIC BIOLOGY WORKING GROUP
ART: CHUCK WADEY WWW.CHUCKWADEY.COM

ENGINEERED GENETIC DEVICES

LET ME INTRODUCE YOU TO A FRIEND OF MINE. IT'S CALLED AN INVERTER DEVICE.

IT COULD BE THE ANSWER YOU'RE LOOKING FOR.

SEE, THANKS FOR TELLING ME AHEAD OF TIME!

WHAT THE HECK IS AN INVERTER?!

I KNOW BACTERIA BALLOONS COULD WORK—
—IF ONLY THERE WAS SOME WAY TO STOP THEM FROM GROWING UNTIL THEY EXPLODE!

OK, PAY ATTENTION! AN INVERTER IS A COMBINATION OF BASIC DNA PARTS THAT—

—WORKING TOGETHER, TURN SOMETHING UPSIDE DOWN.

ON BECOMES OFF, LOW BECOMES HIGH, AND SO ON.

Parts of an Inverter

1. **Ribosome Binding Site (RBS)** - Basic elements that start the process of protein synthesis.
2. **Repressor** - A gene that encodes a particular type of protein that will bind DNA sites in a specific Operator part and cause changes in the rate of gene expression.
3. **Terminator** - Special elements that decrease the flow of RNA polymerase along DNA, sometimes to zero!
4. **Operator** - Stretches of DNA that contain Repressor protein binding sites and RNA polymerase binding and initiation sites. With a Repressor protein, the Operator part will be turned OFF. Without a Repressor protein, the Operator part will be turned ON, allowing RNA polymerase to bind and initiate a HIGH output signal.

YOU COULD HAVE USED AN INVERTER DEVICE TO HELP PREVENT BUDDY'S UNFORTUNATE ACCIDENT.

UHM... WHY'S IT CALLED A DEVICE?

IT'S ENOUGH YOU'RE A KNOW-IT-ALL, YOU DON'T HAVE TO RUB IT IN.

FOR EXAMPLE, HERE'S SOME DNA CODE—

WE CALL AN INVERTER A DEVICE IN ORDER TO HIDE ALL THE DETAILS OF HOW IT WORKS.

—NOW YOU TELL ME WHAT IT DOES!

YOU'D PREFER THING— ANAJISSY?

HEY! WATCH IT!

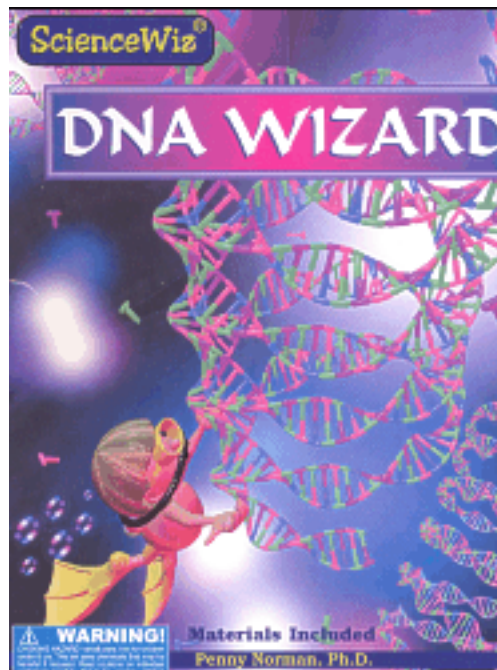
I HAVE NO IDEA, OK? WHAT IS IT?

DON'T FEEL BAD. MY POINT IS, YOU SHOULDN'T HAVE TO MEMORIZE EVERY LAST PIECE OF DNA.

WE'RE GOING TO HIDE ALL THESE DETAILS INSIDE THE DEVICE.

PHEN—

HOW DID YOU DO THAT?



Projects with DNA

For ages 8 and up
Adult Supervision Required

Materials included except for the items listed.
Through play, hands-on projects, patterns and puzzles
this book and kit explores the amazing DNA story.

Extract DNA

Heat SHOCK!

Decode the code of life

Build a DNA ladder.

Grow glowing cells

Is it a boy or girl?

Ooey, Gooley, DNA!

Dress up for sterile techniques.

Solve the chromosome puzzle.

Quality time, quality learning, quality play.