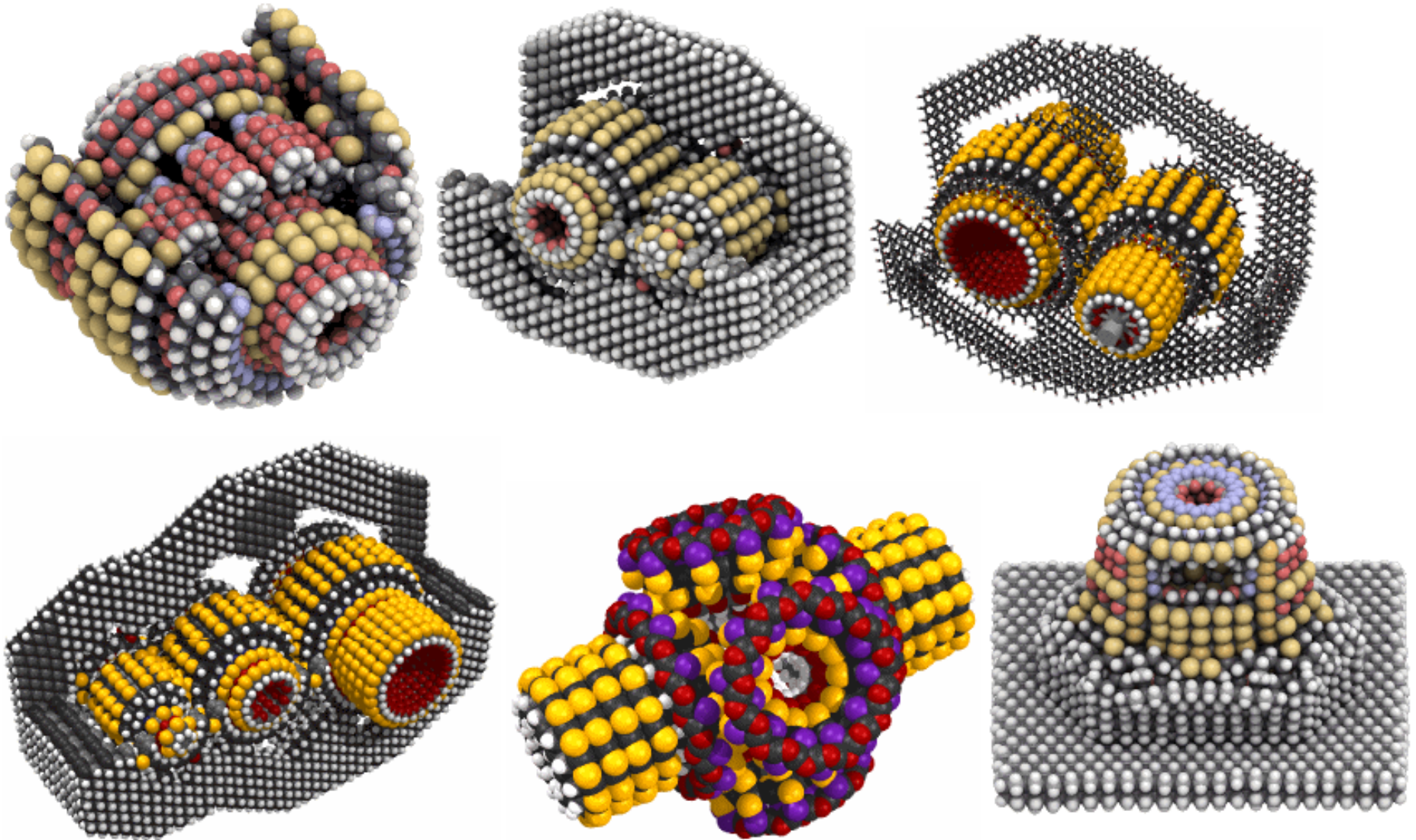


# A low-cost, open-source DIY DNA Synthesizer Project

# Nanoengineer

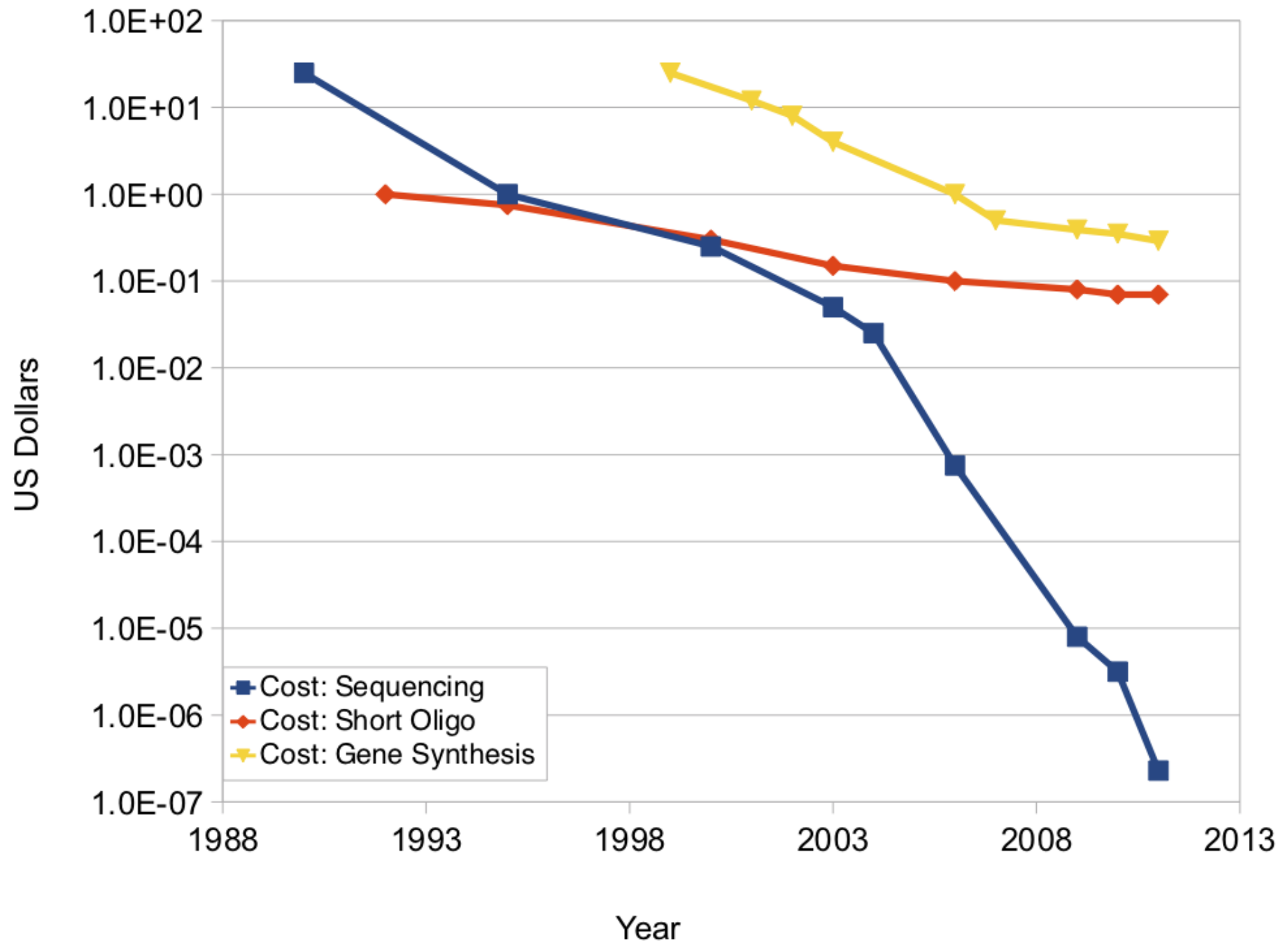


# Nanoengineer

<https://youtube.com/watch?v=vEYN18d7gHg>

# Cost Per Base of DNA Sequencing and Synthesis

Rob Carlson, June 2011, [www.synthesis.cc](http://www.synthesis.cc)



# History of DNA Synthesizers

- Biology is largely about the manipulation, maintenance and transport of DNA.
- Oligonucleotide synthesis is too complex and tiresome to make genomes by hand.
- So, machines were created.
- (Fun patent wars happen here; patents mostly expired by now.)
- We still mostly have these same designs and machines to this day.

# Milligen Cyclone Plus



01/01/00 | MAIN MENU | 00:02:45

MILLIGEN/  
BIOSEARCH  
CYCLONE™ PLUS  
DNA SYNTHESIZER

MAIN MENU

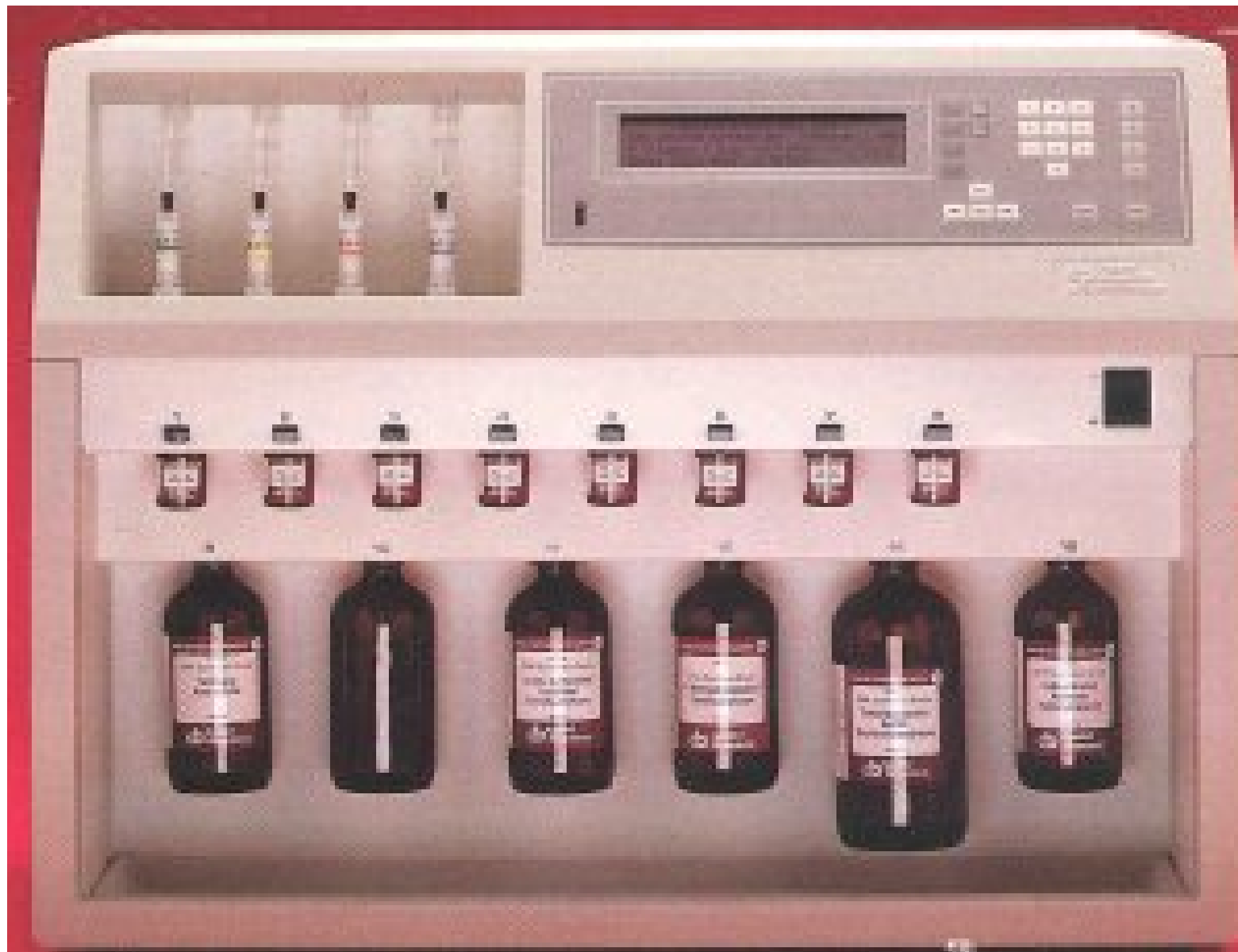
RUN

SEQ

PRIME

-ETC-

# ABI 394





# ABI 381A



eBay: \$599 (not working)





# Expedite 8909





# Azco Biotech Oligo 800 (\$34,000)



# Old Synthesizers Still in Circulation

- They show up on eBay, labx, liquidations, auctions and from refurbishers.
- IDT has someone on staff they pay to build DNA synthesizers, instead of buying new/refurbished equipment.
- IDT also has a “buy back” program where in exchange for your synthesizer they give credit towards their services.
- Cheaper to spend \$4k to pay a synthesis company, than it is to buy a \$50k synthesizer. But the machines don't really cost \$4k in the first place.

# But.

- Per base-pair synthesis costs are projected to drop, but haven't.
- Machine prices haven't changed significantly.
- \$40 million per genome isn't that useful to me, but thanks Craig.
- Intuitively, my cells can make copies of my genome all day long for \$0.00001/genome. This shows that lower costs are at least imaginable.
- So, a better solution might exist.

# Building low-cost DNA synthesizers

- Our project has multiple phases, but the most interesting phase is the microfluidic chip.
- Microscopic channels guide the direction of fluids and reagents.
- Version 1: phosphoramidite chemistry.
- Version “the next one”: ligation chemistry from on-chip 4,096 droplet library (synthesis by short oligo ligation)
- Version “the Future”: enzymatic synthesis by direct control of DNA polymerase.

# Microfluidic Technology

The mess of hoses and tubing is reduced to microscopic traces

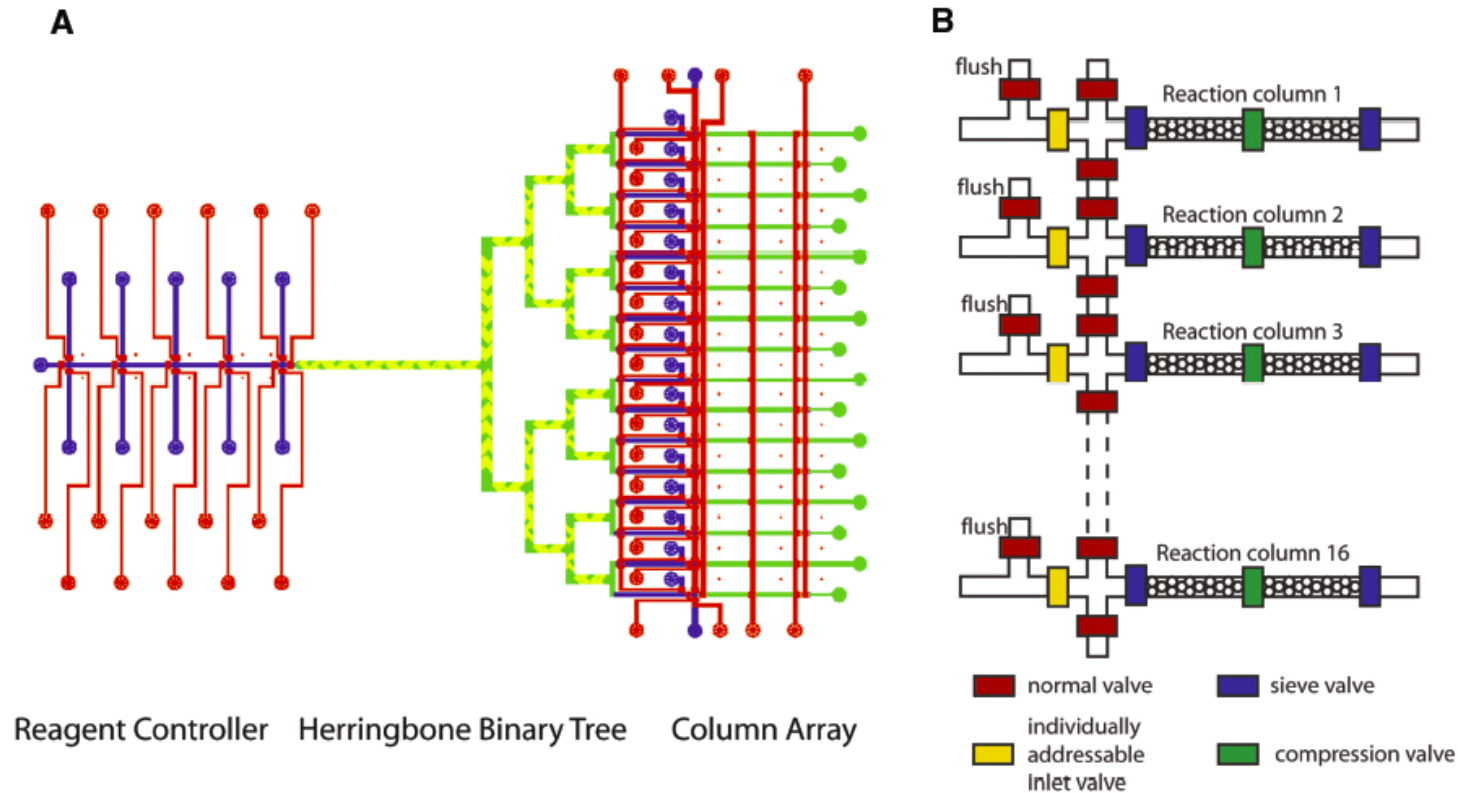
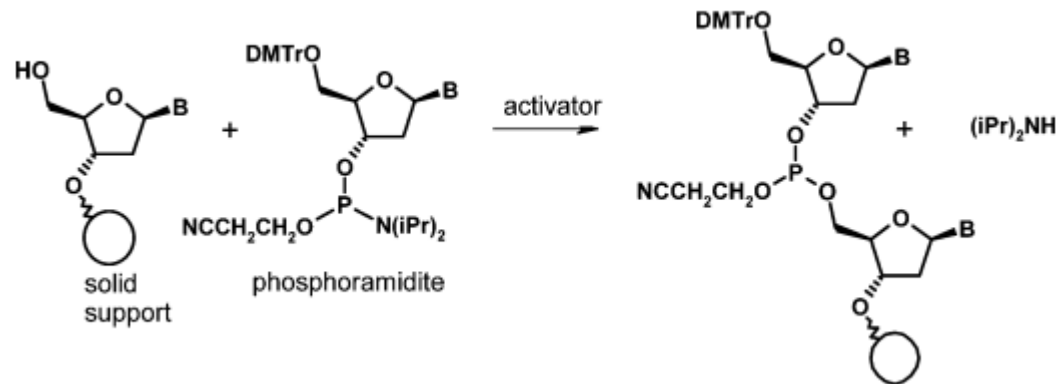
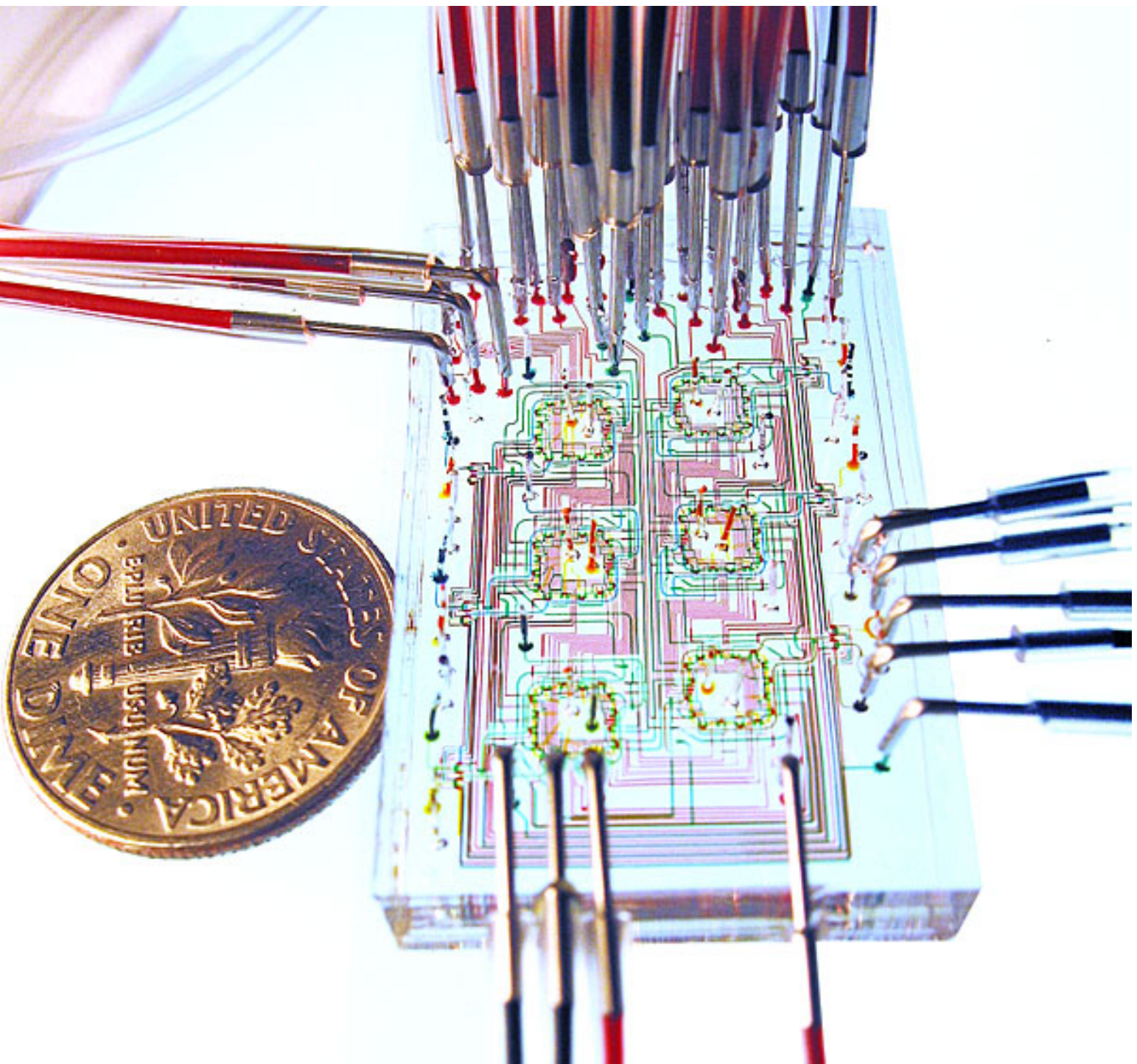
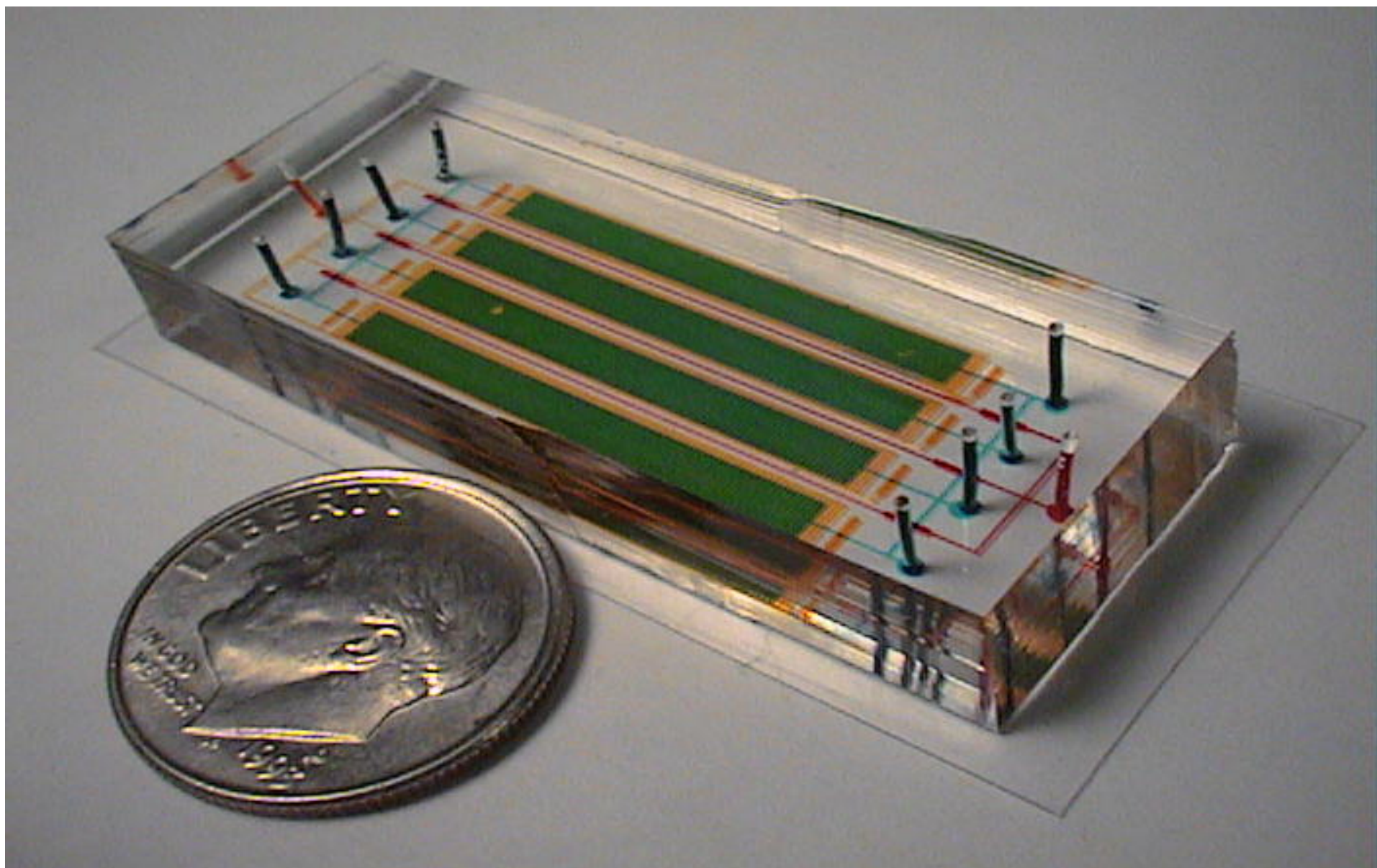


Figure 1. (A) Schematic diagram of a 16-column microfluidic DNA synthesizer. *Nucleic Acids Research*, 2010, Vol. 38, No. 8 2515



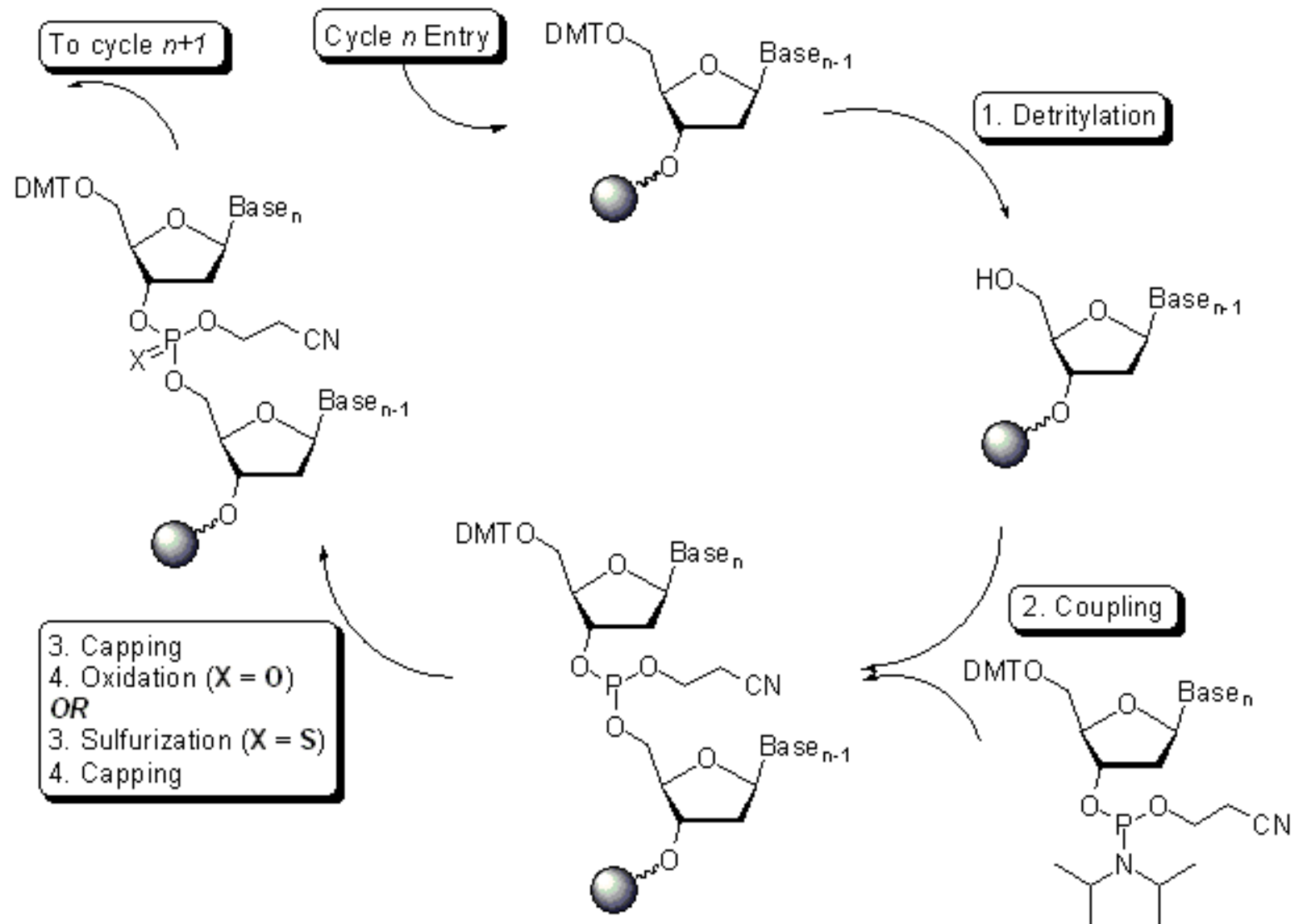








# Phosphoramidite Chemistry



# Some Estimated Costs (Reagents)

|   |       |         |
|---|-------|---------|
| dA-CE (deoxy A - beta cyanoethyl)<br>phosphoramidite                          | 1g    | \$50    |
| dC-CE phosphoramidite   | 1g    | \$50    |
| dG-CE phosphoramidite   | 1g    | \$50    |
| dT-CE phosphoramidite   | 1g    | \$50    |
|   |       |         |
| Activator - 0.45M Tetrazole in<br>Acetonitrile                                | 450mL | \$200   |
| Diluent - Acetonitrile, Anhydrous (Sigma)                                     | 1L    | \$89.7  |
| Cap Mix A - THF/Pyridine/Ac <sub>2</sub> O                                    | 450mL | \$72    |
| Cap Mix B - 16% Melm in THF   | 450mL | \$96    |
| Oxidizing Solution - 0.02M I <sub>2</sub> in<br>THF/Pyridine/H <sub>2</sub> O | 450mL | \$72    |
| Deblocking Mix - 3% TCA/DCM   | 2L    | \$144   |
| Glen UnySupport 500 Angstrom CPG  | 1g    | \$95    |
|   |       |         |
| DNA synthesis reagent cost  | Total | \$768.7 |
|   |       |         |
| Misc solvents, PDMS, Argon  |       |         |
| Mineral Oil, Enzymes, Clean-up kits   |       |         |
| Shipping  |       |         |
|   | Total | <\$1500 |

# Other Costs

- Stanford Foundry – order chips for \$250-\$450/mold (per different mask or prototype), \$50-\$75 per chip

- 100 prototypes => \$45,000

Or ... one time costs:

- CO2 laser cutter: \$10,000 - \$15,000
- Bluray diode laser cutter: \$5,000
- Photolithography setup

# Future Considerations

- Technical feasibility has been proven by already existing commercial technologies, as well as next-generation platforms from academia.
- Material costs of these devices - under \$5,000. Traditional synthesizers could be built cheaper.
- Mail-order DNA synthesis companies have constructed a shared “harmful sequences” database.
- What happens at \$10/genome? \$1/genome? What about \$0/genome?
- Why DRM doesn't work
- Interested in your thoughts.