Barbie Nanoatelier: Open Source DNA-nanotechnology

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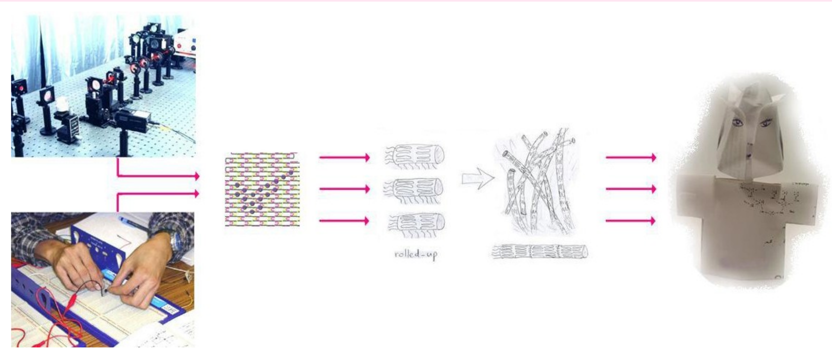
Abstract

We use the DNA origami technique recently developed by Paul Rothemund [1]. The idea is to design a strand of DNA such that it wraps into some meaningful shape. First, the DNA should fold into a two-dimensional rectangular sheet: the *universal DNA-platform* [2]. Secondly, this sheet should wrap itself up into the shape of a short pipe. Third, these little pipes should hook themselves up to each other such that they form one single long pipe. Once the process of DNA folding into 3D structures is understood, shapes can be chosen arbitrarily. We hope it will be possible to maintain molecular sensors, logic gates, and actuators on the surface of 3D DNA-objects, and to reach a swarm behavior of the DNA-origami agents [3].



Introduction

Our DNA-folding project isn't a typical Synthetic Biology project, because we play with 'dead DNA' rather than 'alive DNA' coding proteins. We try to merge the DNA-origami static structures and the dynamic DNA-BioBricks constructs to create living machines. Because we're using DNA-synthesis very actively, it could be called Synthetic Biology or DNA-nanotechnology. The eventual outcome of the project is an Artificial Life and Origami Man. Importantly, we try to add aesthetic principles and rules (symmetry, periodic patterns, recursion, and plasticity) into our future creatures. Crazy? Not at all!

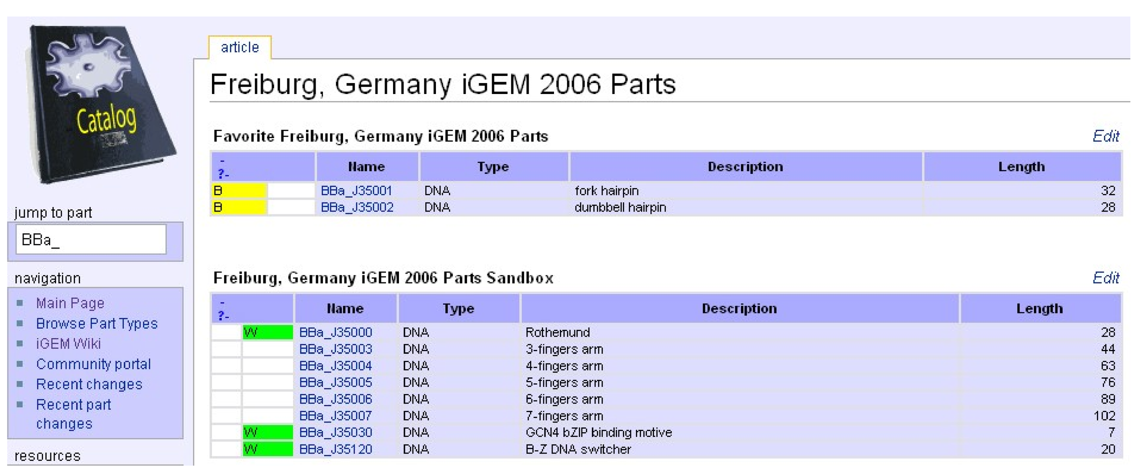
The basic idea is to design DNA so that it folds into DNA-sheet, which we call **the addressable platform with 6 nm scale resolution** [2]. It should be possible to mount some molecules on this DNA-sheet, as if it were graph paper. These molecules could act as sensors, logic gates, and actuators for this nano-platform. We'd attach a specific pattern of catalytic molecules to design synthetic pathways in space, or even to reach an assembly of molecules in the sense of Eric Drexler's assembler. Or we'd organize appropriate molecules, nanoparticles, or quantum dots (qubits) to build a new computer chip. We have a lot of imagination...

"sea of parts"

We started our *Artificial Life Project* with a semi-rational approach [4]. Now we are tuning rationally. We founded *Barbie Nanoatelier* to prove main assembling principles and to design complex DNA-forms. We organized the external BioBricks depository for DNA-nanotechnology. Have a look:

'Dead DNA' (structural DNA)	BBa_J35000 - BBa_J35399
Structures	BBa_J35000 - BBa_J35099
DNA origami	BBa_J35000 - BBa_J35029
protein binding parts	BBa_J35030 - BBa_J35059
aptamers	BBa_J35060 - BBa_J35089
others	BBa_J35090 - BBa_J35099
Devices	BBa_J35100 - BBa_J35199
tweezers	BBa_J35100 - BBa_J35119
nanomechanical switches	BBa_J35120 - BBa_J35139
nanoactuators	BBa_J35140 - BBa_J35159
walking nanomachines	BBa_J35160 - BBa_J35179
others	BBa_J35180 - BBa_J35199
Systems	BBa_J35200 - BBa_J35299
Others	BBa_J35300 - BBa_J35399
'Alive DNA' (protein-coding DNA)	BBa_J35400 - BBa_J35799
Parts	BBa_J35400 - BBa_J35499
Devices	BBa_J35500 - BBa_J35599
Systems	BBa_J35600 - BBa_J35699
Others	BBa_J35700 - BBa_J35799
Other DNA	BBa_J35800 - BBa_J35999

These are just the first examples of *LEGO set of DNA building blocks for Artificial (Synthetic) Life*. They allowed us to run in different directions. Irina pumps aesthetic principles into DNA-creatures. Mona builds the DNA-chip. Andrew used DNA-origami to code images and to design a DNA-nanobot.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Freiburg, Germany iGEM 2006 Parts

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resources

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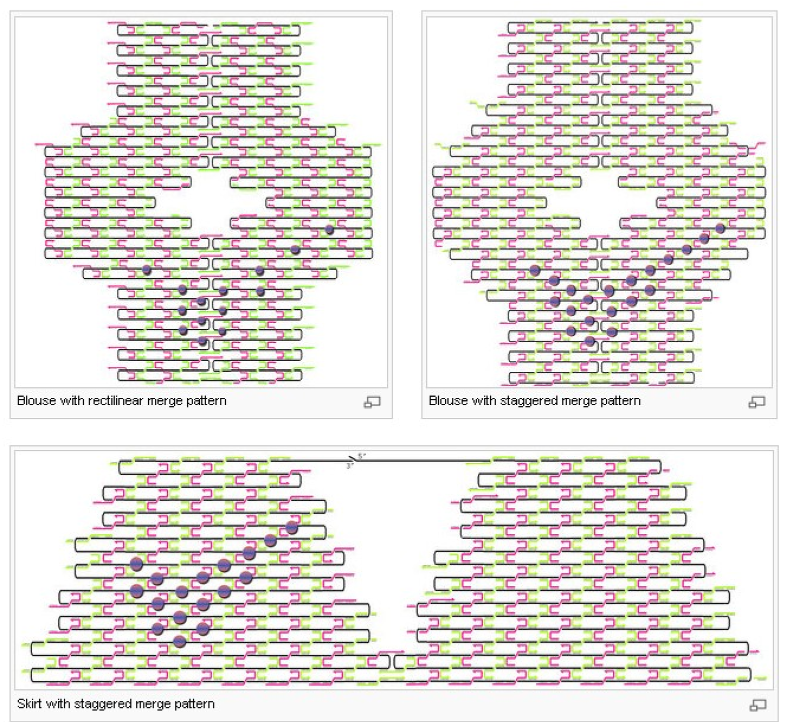
?-		Name	Type	Description	Length
B		BBa_J35001	DNA	fork hairpin	32
B		BBa_J35002	DNA	dumbbell hairpin	28

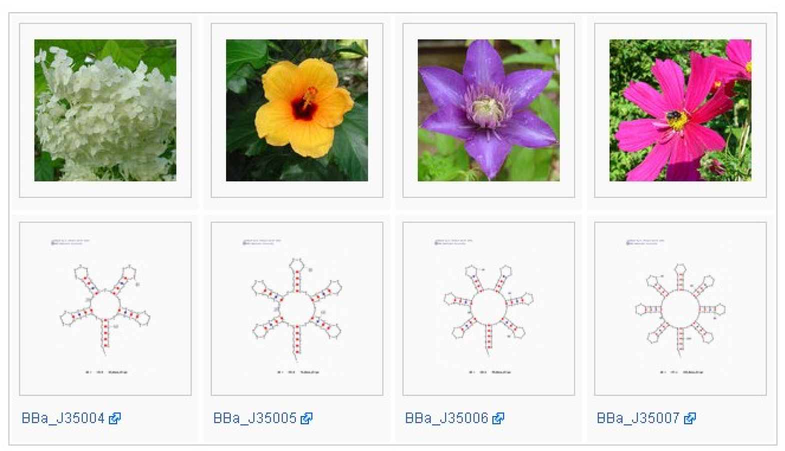
Freiburg, Germany iGEM 2006 Parts Sandbox *Edit*

?-		Name	Type	Description	Length
	W	BBa_J35000	DNA	Rothemund	28
		BBa_J35003	DNA	3-fingers arm	44
		BBa_J35004	DNA	4-fingers arm	63
		BBa_J35005	DNA	5-fingers arm	76
		BBa_J35006	DNA	6-fingers arm	89
		BBa_J35007	DNA	7-fingers arm	102
	W	BBa_J35030	DNA	GCN4 bZIP binding motive	7
	W	BBa_J35120	DNA	B-Z DNA switcher	20

Surrealistic Science

Designing DNA-dresses for an imaginary nano-Barbie doll is the funniest job! We like it, because it requires a great deal of imagination and is really very difficult. None of us could build a bra! - *von Neumann's self-reproducing ... Bra*.





Design Rules

We dramatically simplified Rothemund's scaffold origami method. Now students need only a browser with access to standard bioinformatics tools and a text processor, if they didn't make their design too complex[5]:

Abstraction

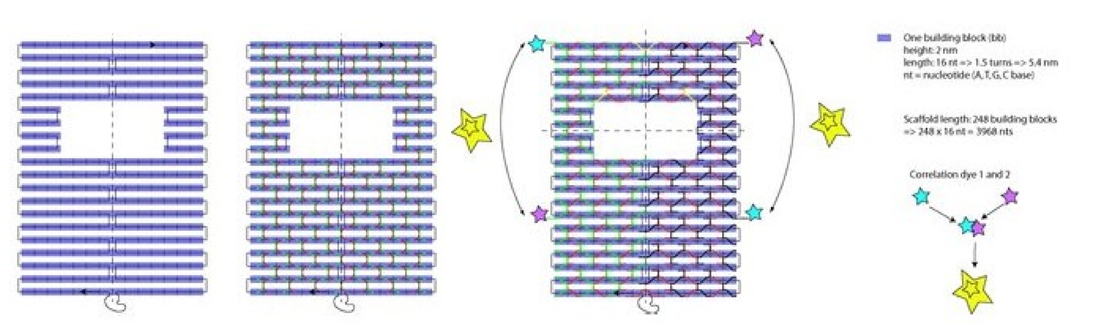
4. Take a sheet of graph paper; 1.5 squares on paper = 1 building block of 16 nucleotides = 1.5 tern DNA = 5.4 nm horizontal and 4 nm vertical.
5. Find a horizontal "snaking" path through the Manhattan skyline geometry of resulting bar graphs, with some vertical turns, and try to exploit symmetry.
6. Starting at one end of the DNA strand, insert a crossover to the strand section above every alternate building block. Add helper strands to bind the scaffold together. As first designed, most staples bind two helices and are 16-mers.
7. Merge helper strands to enhance the scaffold. As second designs, most staples bind three helices and are 32-mers.
8. Fill up the scaffold with letters A, T, G, C, define corresponding staple sequences by complementary mapping from scaffold to valid sequence (A, T and G, C).
9. We now have 1 long scaffold + many shorter staples.

Implementation

- Send your request to a DNA synthesizing company such as *febit* in Heidelberg. You will get 2 bottles: 1 with the scaffold DNA, the other full of staples in 1xTAE (pH 7-8.4) buffer with 10 mM MgAc.
- Get the following equipment: pipettes, gradient thermocycler, AFM, mica.
- Mix the scaffold and staple DNAs in 1/10 (M/M) proportion (2 x 50 µl),
- Warm to 92°C and program the cooling down to room temp 20°C, over 16 hours
- Cleave the mica and place 5 µl droplet on the mica. Image with AFM, landsay eureka!

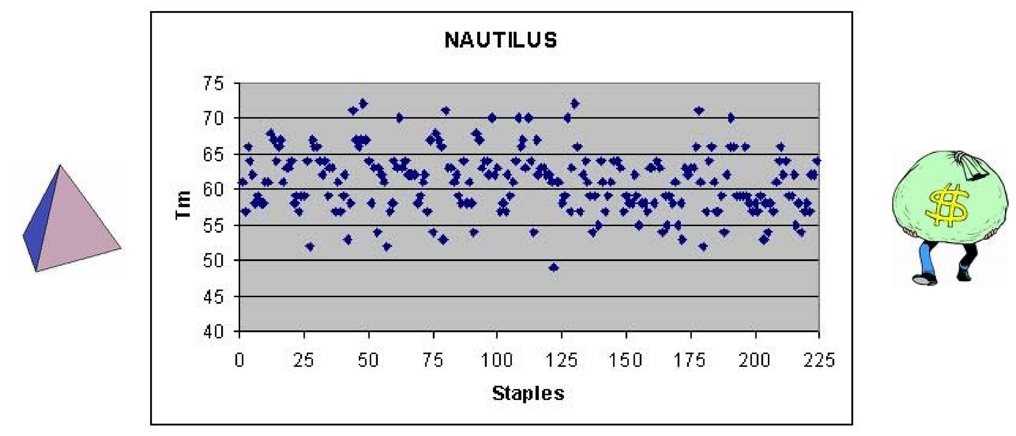
Methods of analysis

- DNA folding (electrophoresis in the polyacrylamide gel)
- 2D structures
  - transmission electron microscopy
  - atom force microscopy
- 3D structures
  - nanoparticle trap
  - quenching of fluorescence
  - *fluorescence correlation spectroscopy/microscopy*



Nanobot NAUTILUS

The tetrahedron on about a 50 nm scale was designed as a derivate of the short pipe to be *a main building block for quaternary structures* (like protein complexes). We hope to use this building primitive to create smart materials and even *a nanoswarm*.



Conclusions

- We designed a lot of creatures from DNA. You'll see!
- We realized *the DNA-synthesis is a bottle-neck* of DNA-nanotechnology.
- We weren't able to create *"self-replicating" staples* and Artificial Life was not created this time. We'll try again.
- We'll won't design the Artificial Life in a tube, rather in the *DATABASE*. We'll only manipulate DNA by a mouse, next by modeling, and then we'll bring it into the lab…

Future projections

Unconventional computing, cryptography, nanoelectronics, nanooptics, nanosensors, drug delivery systems, and smart nanomaterials all are the potential applications for near future.

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4. *Kuznetsov A., Schmitz M., Mueller K.* On Bio-Design of Argo-Machine // GWAL-7: 7th German Workshop on Artificial Life, Jena, Germany, 26-28 July 2006. P. 125-133.
5. *Olga Soboleva, Daniel Hautzinger, Marc Wilnauer, Andrey Kuznetsov, Svetlana Santer, Christian Mueller, Albrecht Sippel, and Jan Korvink* T-shirt from DNA // ibid [2]

Acknowledgements

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