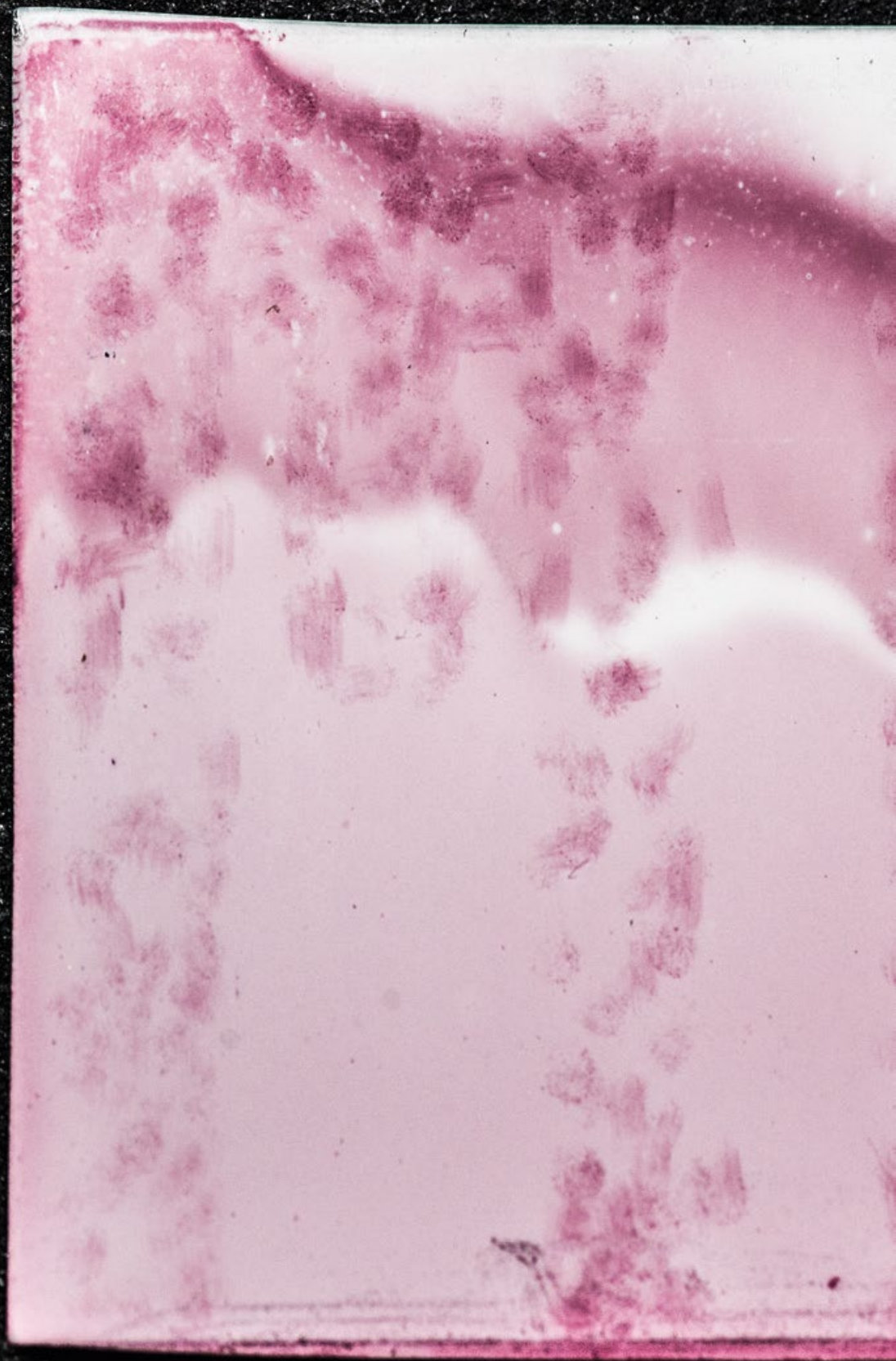
Skylar Tibbits, Lina Kara'in, Joseph Schaeffer, Helena de Puig, Jose Gomez-Marquez and Anna Young

DNA disPLAY



Jose Gomez-Marquez, Anna Young, Lina Kara'in and Skylar Tibbits, Single DNA Drawing, MIT Self-Assembly Lab, 2013
Drawings generated from the Game of Life algorithm were printed with thrombin protein on nitrocellulose paper. The resulting dark spots emerge only as a result of interaction between the printed protein and the washed DNA.

PROGRAMMABLE BIOACTIVE MATERIALS USING CNC PATTERNING

2D, 3D and 4D printing are fertile areas for design and material experimentation, and thus the architectural detail in the future. Here **Skylar Tibbits**, along with his collaborators – Lina Kara'in (MIT Self-Assembly Lab), Jose Gomez-Marquez, Anna Young and Joaquin Navarro (Little Devices Lab, MIT), Lee Gehrke, Helena de Puig and Justina Tam (Gehrke Lab, MIT), and Joseph Schaeffer and Carlos Olguin (Autodesk Inc) – describe the development of DNAdisPLAY: a project that created a physical prototype and design workflow for 2D bioprinting, resulting in the development of software, hardware and printed DNA patterns on paper.

Since the introduction of inkjet printing, two-dimensional printing technologies have democratised our ability to create at-home physical documents. Paper-based printing has become ubiquitously accepted as both a commercial-scale process and a necessary at-home tool. Charles Hull's 1984 invention of 3D printing has followed suit with a new vision for the way we make things from industrial manufacturing to do-it-yourself fabrication.[1] Both of these technologies have challenged and catalysed the design process by transitioning to digital tools and generative processes across software, fabrication and material domains.

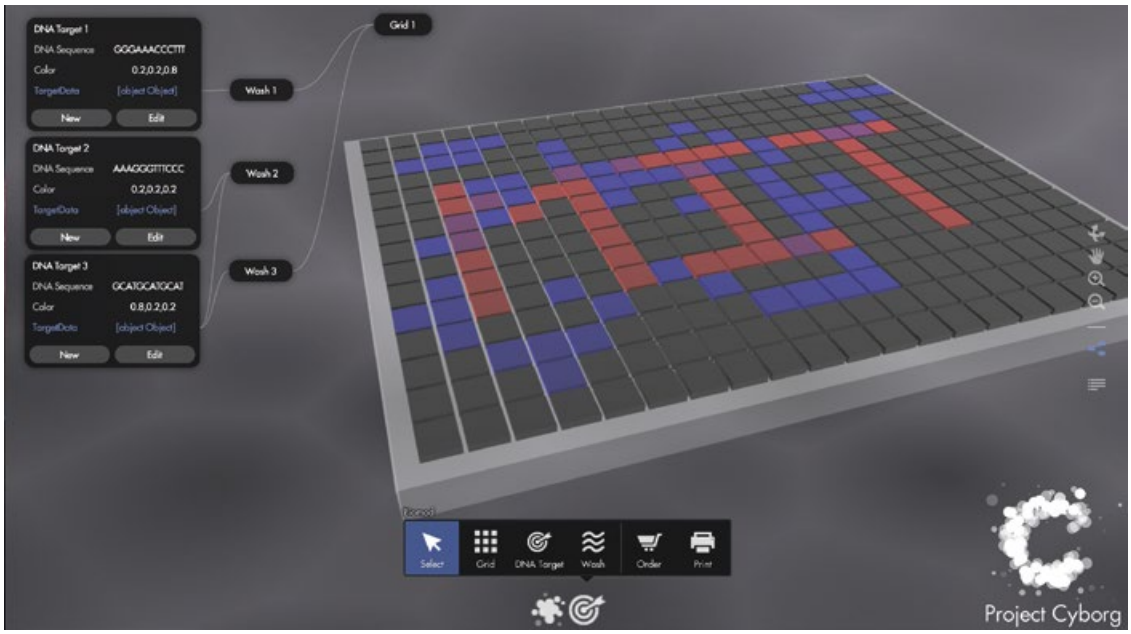
An opportunity has emerged to program materials across length-scales to store information, compute digital logic and change physical state, material property or shape. 4D printing is one recent technique that allows multi-material structures to be printed and self-transform in shape and material property when exposed to water.[2] Similarly, yet at a much smaller scale, 'DNA origami' is a technique where custom sequences of DNA are synthesised and then self-assemble into nanoscale functional objects. These 2D and 3D objects are typically designed using software such as cadnano, developed by the Wyss Institute and Autodesk Inc.[3] At the materials level there is an influx of research around smart materials and self-assembly processes to efficiently manufacture new material properties and fabricate meso-scale structures.[4] Each of these developments points towards a future of programmable materials that can be printed with new physical properties and controllable behaviour, thus once again challenging the design field to invent new tools and applications.

Printing with DNA

3D printing remains far from the realities of construction and architectural materials that rely heavily on two-dimensional assemblies of sheet-based goods and complex user, material and environmental constraints. Thus there is an opportunity for a resurgence of 2D printing with the addition of programmable materials, offering limitless applications in architectural sheet materials, wall finishes, responsive materiality, environmental adaptation and many others.

Advances in DNA nanotechnology offer an opportunity to use printed DNA as a smart architectural material. DNA's four nucleotides can form specific bonds: adenine (A) binds to thymine (T), and guanine (G) binds to cytosine (C). This logic allows a programmable substrate to build computational components such as logic gates, motors and sensors as well as self-assembling 2D and 3D objects with defined structural properties and bioreactive patterns.[5] DNA can be used to store arbitrary information into custom-synthesised DNA strands that can then be sequenced to read out the exact digital information from biological material.[6] These properties make it extremely well suited as a programmable printed material that can sense internal and external conditions, compute information and visually transform.

2D bioprinting is a mature field generated from microarray and high-throughput multiplexed analysis platforms.[7] Self-assembly at the nanometer scale using additive and subtractive processes employs expensive photolithographic and atomic force microscopy tools.[8] Similarly, static stamping of protein patterns onto substrates has given way to a number of computer-controlled dynamic patterning deposition options such as contact spotting, non-contact spotting, inkjet and piezoelectric deposition systems.[9] However, all of this equipment remains an expensive niche domain, often costing in excess of $100,000. The software layer that runs these machines is often complex and optimised for parallelisation and combinatorics, varying the intensity and reagents within a matrix. Unfortunately, the deposition systems available are difficult to use and do not lend themselves to complex DNA patterning on a variety of material substrates.



Carlos Olguin and Joseph Schaeffer, Project Cyborg, Bio/Nano/ Programmable Matter Research Group, Autodesk Inc, 2013
Autodesk's Project Cyborg is a software tool under development for the generative design and simulation of biological information. The displayed patterns emerge only after simulating the washing of complimentary DNA strands.

The DNA Printing Workflow

DNA disPLAY was developed as a collaboration between the Massachusetts Institute of Technology (MIT) Self-Assembly Lab, Little Devices Lab, Gehrke Lab and Autodesk as a physical prototype and design workflow including software, hardware and printed DNA patterns on paper.[10] The project attempts to take on the challenge of utilising DNA as a new programmable design medium by making it both physical and visual for the built environment. The aim was to allow anyone to design and print with DNA, eliminating the expensive and difficult step of DNA imaging and opening up possibilities of biological printing for architectural surfaces. Proteins were CNC-printed on nitrocellulose paper and then washed with DNA and gold nanoparticles to reveal custom patterns. The workflow has four main steps: design and software; custom biomaterial; CNC printing; and programmable drawings.

Design and Software

The first step in the DNA printing workflow is a new software tool for generative design and simulation of biological information. A custom application was developed and built on top of Project Cyborg, the code name of a new design platform being developed by Autodesk Research's Bio/Nano/ Programmable Matter Research Group. This software tool allows one to design and map patterns with their associated biological information. It was designed to embed domain-specific knowledge and allow users to easily work at a high level, designing patterns, interactions and dynamic transitions for DNA printing. Each printed droplet represents a biological pixel in the software, and can be embedded with known biological sensors and organised into complex patterns reacting to internal or environmental triggers. After an arrangement of pixels has been designed, populated with biological sensors and simulated, the custom proteins and DNA sequences can be ordered directly. Future work will attempt to link the designed digital structure in Project Cyborg directly with hardware platforms, allowing them to be printed directly on various substrates.

Custom Biomaterial

In previous work, the research team printed multiple patterns and symbols with ligands on paper to create a diagnostic imaging system. As the team moved towards an easier approach to 2D biopatterning, a convenient biological structure was sought to serve as a conceptual framework. Thrombin protein and its aptamer (thrombin binding aptamer/TBA) were selected due to their well-characterised interaction with binding affinities in the nanomolar range. One of the first therapeutic DNA aptamers to be isolated was with human thrombin, a key protein that helps regulate the formation of blood clots.[11] The DNA aptamer 5' GGTTGGTGTGGTTGG 3' self-folds in a G-quadruplex secondary structure in order to bind to thrombin.[12] The aptamer was bound to a gold nanoparticle solution and the protein remained in a separate solution, ready for printing. The binding allowed the printed thrombin patterns to emerge only after encountering the correct DNA sequence.

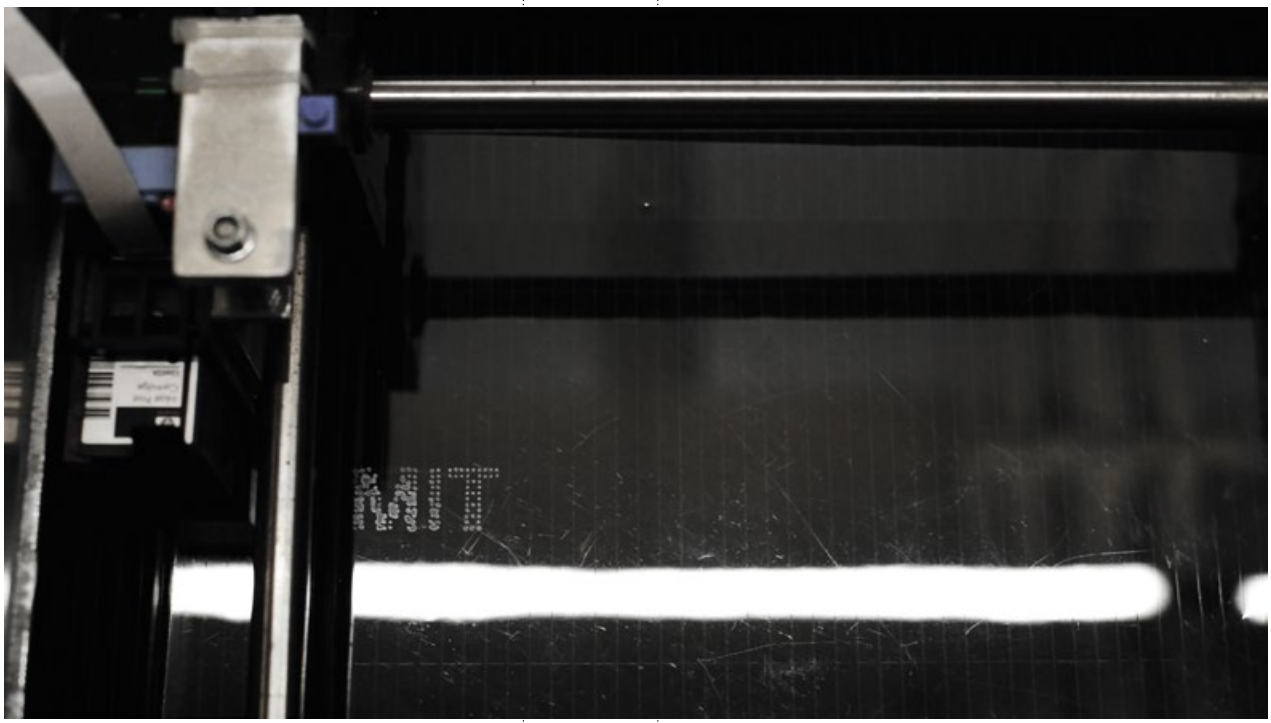
CNC Bioprinting

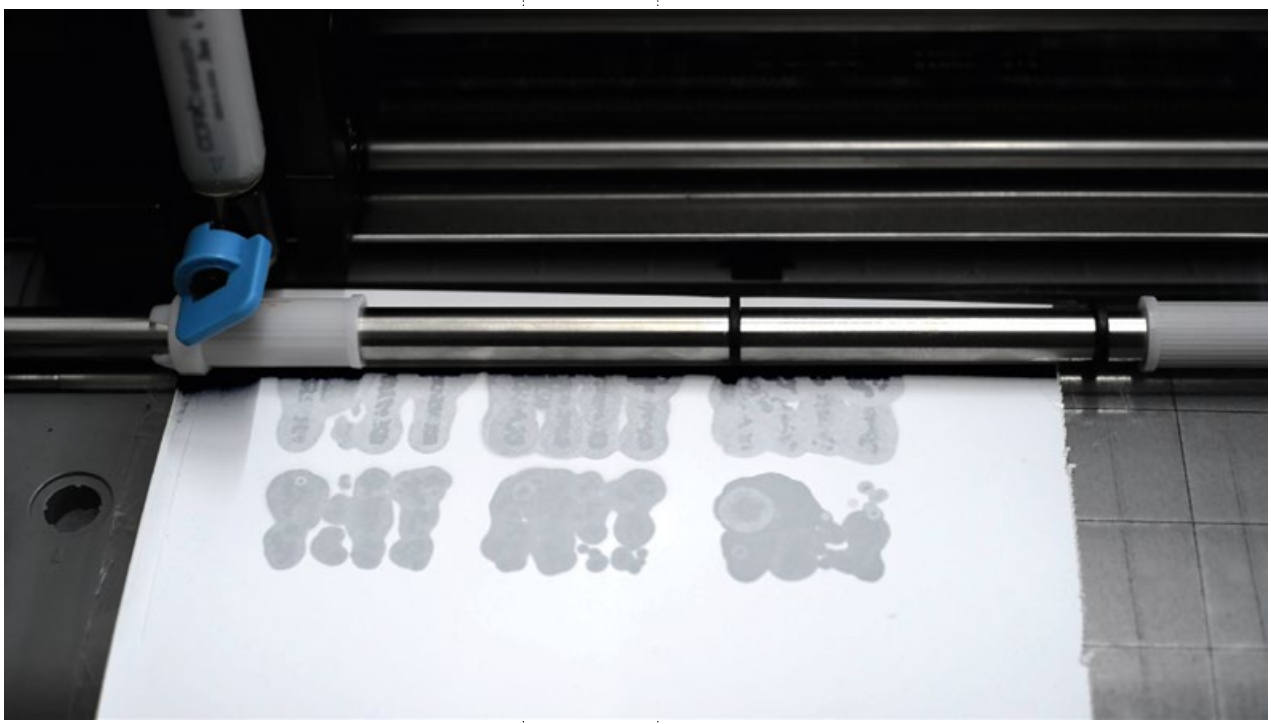
After generating patterns, simulating interactions and ordering custom biomaterials, the physical structures were then printed on various substrates. Three main hardware platforms were tested, with corresponding opportunities and drawbacks. First, a custom CNC printing machine was built and used to deposit DNA and protein markers with HP inkjet cartridges. This platform allowed for complete control over resolution and quantity of droplet size, however it required a specialised software workflow that increased workflow complexity to transform from a standard image into a DNA printed output. The same approach was tested on a simple desktop printer, easing the design workflow but

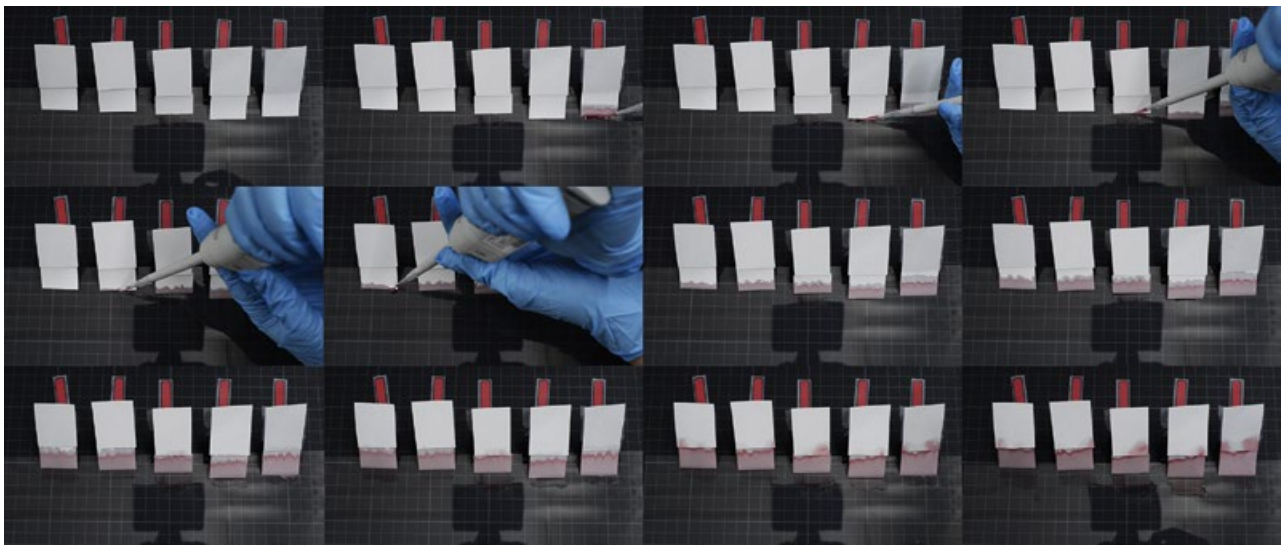
Jose Gomez-Marquez, Anna Young, Lina Kara'in and Skylar Tibbits, CNC DNA Printer, MIT Self-Assembly Lab, 2013
top: A custom CNC printer was developed to deposit synthesised DNA onto clear film in the precise letters 'MIT'.

Jose Gomez-Marquez, Anna Young, Lina Kara'in and Skylar Tibbits, DNA Pen Plotter, MIT Self-Assembly Lab, 2013
bottom: A pen-plotter and vinyl cutting machine were used for DNA and protein deposition. This hardware provided an easy interface and precise control over droplet resolution. A felt-tip pen was loaded and used to deposit thrombin protein on nitrocellulose paper.

Jose Gomez-Marquez, Anna Young, Lina Kara'in and Skylar Tibbits, DNA Washing, MIT Self-Assembly Lab, 2013
opposite: After depositing thrombin protein on nitrocellulose paper, a custom-synthesised DNA sequence with gold nanoparticles was washed across the paper. The blank paper then revealed custom-designed DNA patterns emerging from the programmed interaction of the protein and DNA strands.







adding constraints in substrate handling and control of droplet size. The final machine, and the most successful thus far, was a vinyl cutter and pen-plotter. This achieved a fast workflow and full droplet size control. A felt-tipped pen, filled with thrombin protein, was mounted on the machine instead of a cutting blade. Droplet size was achieved by varying the impact height of the pen. In effect, an affordable computer-controlled contact-printing system was created. A series of six unique drawings were printed with thrombin protein using this machine and were then washed with DNA to reveal custom patterns.

Programmable Drawings

Printed DNA patterns were designed using principles from Conway's Game of Life, an early model of visual computing that has three fundamental rules: overcrowding, under-population and reproduction.[13] These rules were propagated from an initial pattern spelling the letters 'MIT'. A series of generations were printed showing a progression of the letters dissolving into complex dynamic patterns that emerged only after being washed with complementary DNA. The Game of Life, created with the material of life, was specifically chosen as a case study due to its proven capability of universal computing, its visual aspect and rule-based patterns of local interaction. These dynamic patterns can be extrapolated as a strong example for programmable DNA printing since they argue for a visual output that could dynamically transition between patterns based solely on local DNA computing.

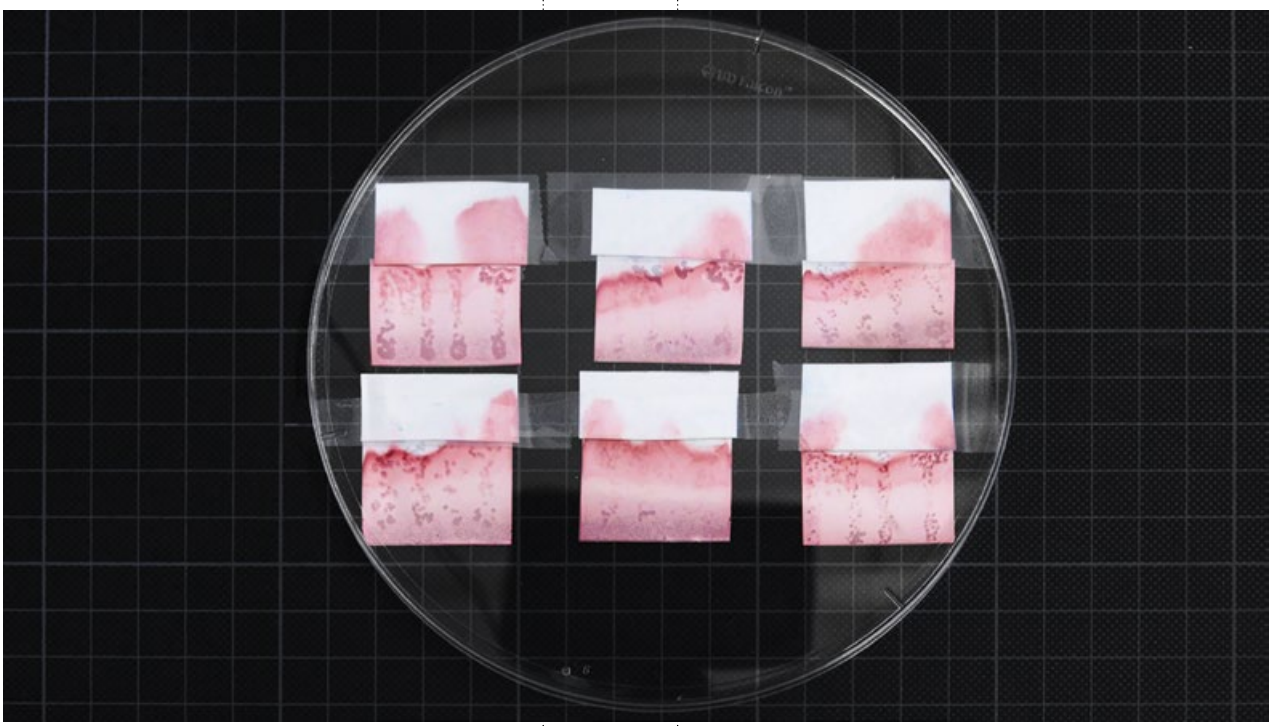
Architectural Applications

On the surface, printed DNA drawings simply demonstrate the programmable binding of proteins and DNA. However, they conceptually go much further and point towards the capability for DNA to act as a type of smart ink for two-dimensional bioprinting and architectural surfaces. Avant-garde architecture has long been interested in transformation, both physically and visually, but the reality of today's architectural details for sensing and transformation rely solely on robotics-like motors, mechanisms, sensor devices, electronics and traditional notions of computing. These details are not scalable due to factors including their prohibitive cost, failure-prone nature, lengthy assembly time and lack of interoperability. Dynamic transformation has thus been relegated to machinery, products and other economies of scale, rather than architectural domains. However, the future of architectural details can now be revolutionised with printed biological material due to its simple, cheap and elegant response to the environment. DNA is everywhere; it is within our bodies, our buildings and surrounding environment. Programming this abundant material may therefore offer a world of transformation, sensing and environmental consciousness that has previously been inaccessible to architects.

In future, DNA-based architectural materials could be designed to react to the touch of a person's hand, a building's air quality or external environment such as pollution, sunlight or even acidity levels, showing a smart response between architectural surfaces and their dynamic surroundings. There is a strong potential for architectural material finishes to transform from one pattern, level of transparency, texture or shape into another. Like electronic displays made with DNA, these new building surfaces could sense, transform, compute or store information directly in the walls themselves. DNA facades, inks, signage and material textures may ultimately respond to the quality of a building's internal or external environment, and programmably adapt to user interaction. In a world where information technology, biological response and environmental consciousness are blending, printed DNA substrates may become the future of architectural details. ◘

Jose Gomez-Marquez, Anna Young, Lina Kara'in and Skylar Tibbits, Six DNA Drawings, MIT Self-Assembly Lab, 2013
top: Six DNA drawings were produced from the Game of Life algorithm, demonstrating unique patterns based on the interaction between CNC-printed thrombin proteins and DNA with gold nanoparticles.

Lina Kara'in and Skylar Tibbits, MIT Game of Life, MIT Self-Assembly Lab, 2013
bottom: Six generations of the Game of Life were produced from an initial starting condition in the form of 'MIT'. The following generations erode away based on three simple rules: overcrowding, under-population and reproduction. The generated patterns were used for protein deposition and DNA display.



Notes

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10. This work was developed for BioMod 2013, a DNA/biomolecular design competition, and would not have been possible without strong collaboration between the MIT Self-Assembly Lab (Skylar Tibbits and Lina Kara'in), the Little Devices Lab, MIT (Jose Gomez-Marquez, Anna Young and Joaquin Navarro), Gehrke Lab, MIT (Lee Gehrke, Helena de Puig and Justina Tam) and Autodesk Inc (Joseph Schaeffer and Carlos Olguin).
11. See LC Bock, LC Griffin, JA Latham, EH Vermaas and JJ Toole, 'Selection of Single Stranded DNA Molecules that Bind and Inhibit Human Thrombin', *Nature*, 355 (6360), 1992, p 564, and Besik I Kankia and Luis A Marky, 'Folding of the Thrombin Aptamer into a G-Quadruplex with Sr2+: Stability, Heat, and Hydration', *Journal of the American Chemical Society*, 123 (44), 2001, p 10799.
12. Astrid Joachimi, Günter Mayer and Jörg S Hartig, 'A New Anticoagulant-Antidote Pair: Control of Thrombin Activity by Aptamers and Porphyrins', *Journal of the American Chemical Society*, 129 (11), 2007, p 3036, and Helena de Puig, Anna Cifuentes Rius, Dorma Flemister, Salmaan H Baxamusa and Kimberley Hamad-Schifferli, 'Selective Light-Triggered Release of DNA from Gold Nanorods Switches Blood Clotting On and Off,' PLOS ONE, 8 (7), 2013, e68511.
13. John Conway, 'The Game of Life', *Scientific American*, 223 (4), 1970, p 4.

Jose Gomez-Marquez, Anna Young, Lina Kara'in and Skylar Tibbits, DNA Handling, MIT Self-Assembly Lab, Massachusetts Institute of Technology (MIT), Cambridge, Massachusetts, 2013
DNA is often stored in small test tubes, must be handled extremely carefully and cannot be seen by the human eye.

The project attempts to take on the challenge of utilising DNA as a new programmable design medium by making it both physical and visual for the built environment. The aim was to allow anyone to design and print with DNA, eliminating the expensive and difficult step of DNA imaging and opening up possibilities of biological printing for architectural surfaces.