3DNA: A Tool for DNA Sculpting

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Abstract—DNA self-assembly is a robust and programmable
approach for building structures at nanoscale. Researchers
around the world have proposed and implemented different
techniques to build two dimensional and three dimensional nano
structures. One such technique involves the implementation of
DNA Bricks [1], proposed by Ke et al., 2012 to create complex
three-dimensional (3D) structures. Modeling these DNA nano
structures can prove to be a cumbersome and tedious task.
Exploiting the programmability of base-pairing to produce
self-assembling custom shapes, we present a software suite
3DNA, which can be used for modeling, editing and visualizing
such complex structures. 3DNA is an open source software which
works on the simple and modular self assembly of DNA Bricks,
offering a more intuitive better approach for constructing 3D
shapes. Apart from modeling and envisaging shapes through a
simple graphical user interface, 3DNA also supports an integrated
random sequence generator that generates DNA sequences cor-
responding to the designed model. The software is available at
www.guptalab.org/3dna

Keywords—DNA origami, DNA pen, DNA self assembly, nanotech-
ology, bottom-up fabrication, DNA computing, molecular
canvas, DNA bricks, software, open source.

I. INTRODUCTION

Construction of nano devices and nano-structures using
the approach on self assembly is one of the most engrossing
and upcoming field of research in DNA nanotechnology [2].
Along with static structures, various dynamic models like
molecular switches, DNA walkers, DNA robots, molecular
circuits [3], [4], [5] are being developed. These structures are
built by designing the DNA sequences in a specific manner
that enforces the DNA to bind with its complementary base
pairs. In the forefront, researchers have made attempts to build
structures arbitrarily, but the field received a boost with the
introduction of the idea of DNA origami by Paul Rothemund,
in which a scaffold DNA sequence (which is often a viral
genomic DNA), can be folded into a desired fashion by using
synthetic staples strands [6], [7], [8], [9], [10], [11], [12].
There are list of different 2D and 3D nano structures [13],
[14], [8], [15], [16] built by using various DNA self assembly
approaches. In subsequent years, Peng Yin et al., gave rise
to an approach of modular self-assembly which employs
DNA tiles. These tiles are single stranded tiles(SSTs) and they
ultimately assemble into finite 2D shapes [17]. This technique
has paved the way for an efficient, simple and systematic
approach to self assembly. Following the method of DNA tiles,
Ke et al. extended the idea to DNA bricks [1], which allows
the construction of 3D shapes.

To aid the various applications of building these 3D DNA
structures, 3DNA implements the concept of modular assembly
of DNA bricks to construct 3D shapes. 3DNA can be employed
to minimize the time-consuming and error-prone task of
designing DNA sequences to model these formations. The soft-
ware provides a 3D molecular canvas interface where the user
can model/design complex DNA structures. It also includes
a sequence generator which computes the DNA sequences cor-
responding to the structure.

The sequences generated by the software self-assemble in
one step annealing reactions into prescribed 3D shapes. The
software interface includes a 3D molecular canvas of
varying dimensions composed of several molecular pixels,
which ultimately represent DNA bricks. By deselecting pixels
from the molecular canvas we have been able to create different
shapes of varying dimensions. Using 3DNA we have designed
complex shapes with intricate interior cavities. The use of
3DNA significantly reduces the effort required to design 3D
DNA structures.

This paper is organized as follows. Section 2 describes an
outline of GUI and section 3 provide a detailed description of
the software functionality. Algorithms for strand construction
and workflow are defined in section 4 followed by detailed
analysis of a sample case study in section 5 and finally con-
clusion in section 6. Section 7 provides a link for downloading
the software and related materials.

II. GRAPHICAL USER INTERFACE

The graphical user interface (GUI) for 3DNA has been
developed to enable the user to edit/model and visualize
complex 3D shapes on the molecular canvas (as shown in Fig.
1). By rendering the 3D canvas, the user can edit molecular
pixels and envisage the shape drawn at nano scale.

A. Molecular Canvas

The customizable Java3D based molecular canvas can be
viewed as a block(cube) of molecular pixels, each representing
an 8–nt duplex and has a dimension of approximately 2.5 by
2.5 by 2.7nm. The convention for setting the dimensions for
the 3D molecular canvas are: height and width in terms of
DNA helices along the x & y axis respectively, depth in terms
of DNA base-pairs (multiples of 8) along the z-axis. This
3D model contains the positional information of each 8–bp
duplex in every pixel, which can be removed independently.

B. Canvas Control Panel

The user interface provides the ability to control and fiddle
with the 3D environment by changing the camera view using
the Canvas Control Panel. The panel is equipped with zoom in,
zoom out and other directional buttons for viewing the canvas
from different angles and a better visualization of the structure.
III. FUNCTIONALITY

The following subsections are aimed to give a brief idea about the functionality (Fig. 2) of the software as a whole. The main functional features of the software are its input modules (the 3D canvas) and import, its computational modules as well as its output modules which include saving sequences into files and export.

A. Creating a new canvas

The 3D molecular canvas can be accessed by selecting the New Canvas button from the menu bar. On initiating the molecular canvas, the user will be prompted to enter the dimensions i.e. the height (DNA helices), width (DNA helices) and depth (base pairs and a multiple of 8) of the 3D canvas (Fig. 3). After entering the dimensions a custom Java3D based molecular canvas appears on the screen with the specified dimensions.

B. Creating structures

The smallest unit of the 3D molecular canvas is the molecular pixel, which represents 8−bp. Each of these pixels combine according to the software prescribed algorithm into full (32−nt) or half (16−nt) bricks and can be removed independently. To create structures, the canvas allows the user to freely deselect a molecular pixel by simply clicking on it, thereby depicting sculpting of the DNA block. Fig. 4 displays a sample structure of a 3D hollow cube sculpted by removing the unwanted pixels from an original cube of dimensions 10H × 10H × 80B (more details on the same can be found in section 5).

C. Visualization of the structure

Visualization can be used to gain deeper insights on the structural bindings of the sculpture. It shows how the base pairs interact and associate with each other to envisage the shape at nanoscale.

D. Analysis of sequences

The final set of sequences generated by the software can be analyzed by the Graphical Analysis option on the menu bar. It generates a statistical analysis of the number of pairs of 8−base
domains that contain 8, 7, or 6 identical bases among all the
domains in the final sculpted structure. Fig. 5 demonstrates
this functionality for a $6H \times 6H \times 48B$ cube. It indicates
the number of 8, 7, or 6 identical bases in the entire group
of 432 domains required make up the targeted structure. This
analysis is useful to understand the nature of single stranded
DNA sequences when conducting experiments. One can use
this analysis to measure the stability and tolerance of identical
bases in the domain sequences of the actual self-assembled
structures.

E. OUTPUT

When a user sculpts the targeted 3D structure on the can-
avas, the corresponding DNA sequences are generated. 3DNA
allows the user to save the DNA sequences in a variety of
formats:

- Save as .pdf file: DNA sequences designed by the
  software for creating the prescribed shape on the 3D
  molecular canvas can be saved in a .pdf format by
  accessing the Save as .pdf button. The sequences
  pertaining to the full and half bricks are specified in
  a table along with the corresponding molecular pixels
  (voxels) to which they assigned. Each file saved in this
  format contains a unique bar code for identification.
- Save as .csv file: The final DNA sequences along with
  the voxel coordinates generated by the software are
  saved into a .csv file by accessing the Save as .csv
  button.
- Save as .tex file: To save the final sequences into a
  .tex file, one can simply click on the the
  Save as .tex button. Any existing project
  which has been exported previously can be opened and
  modified/viewed through the Import functionality by
  clicking on the Import button. Any existing project
  with .tex extension can be imported.

F. Cost Estimator

It calculates the cost of experiment(USD) based on the
number of nucleotides.

IV. ALGORITHMS AND WORKFLOW

The following section expounds upon the methods and
algorithms used during strand design and modelling of se-
quences. It also covers the workflow of the software starting
from creating the basic canvas to obtaining the final sequences
and the explanation of the intermediate steps in its process.

1) Domain Sequence Generation: Each of the four 8−nt
domains of the canonical full brick and two 8−nt domains of
half bricks are designed by imposing the following constraints
on completely random assignment of base pairs(A−T, G−C).
(These constraints may or may not apply to the complimentary
strands of domains):

2) Molecular Pixel to Brick Sequence Generation: In order
to understand how each molecular pixel on the canvas is
ultimately mapped to its corresponding brick sequence, it is
important to understand the theory of the DNA single stranded
bricks, their types, orientation and modeling:

- Full brick: A full brick (Fig. 6) is 32−nt and is
  conceptualized as four consecutive 8−nt domains.
  Each DNA brick has a unique nucleotide sequence. An
  identical shape is assumed by all the DNA bricks upon
  the target formation, with the two 16−nt antiparallel
  helices joined by a single phosphate linkage in the
  center.
- Half brick: The half brick(Fig. 7) is a bisection of
  the full brick representing a single helix with two
  domains.
- Orientation and modeling of bricks: As introduced by
  Ke et al. the bricks adopt the LEGO modeling [1] of
  DNA. Each brick can be conceptualized as a LEGO
  cube: the domains 1 and 4 form the protruding ends
  and domains 2 and 3 form the backbone of the LEGO
  cube. The bricks adopt one of four orientations—
  north, west, south or east and therefore must be either
  horizontal or vertical. A full brick attaches to four
  immediate neighbors, which are complementary in
  sequence and perpendicular in orientation with it (Fig.
  8).

A. WORKFLOW

The complete workflow starting from creation of canvas to
saving the subsequent sequences into files is illustrated in Fig.
3DNA is designed to obey the following steps (workflow)
for every target structure:

1) Build 3D canvas of preferred dimensions.

Fig. 6. A depiction of a full brick and its four domains through a helical
single−stranded structure

Fig. 7. A depiction of a half brick and its domains through a helical
single−stranded structure
Fig. 8. Interaction of a full brick with its neighbors, showing the way complementary strands of the domains (marked by the same color) bind to each other.

Fig. 9. A depiction of the workflow in 3DNA.

2) Design sequences for all the full and half bricks comprising the entire canvas.

3) Process the sculpted shape after user has removed the unwanted molecular pixels (voxels) and select the required subset of bricks needed for the structure.

4) Modify the acquired subset of sequences and assign protector bricks (unpaired strands composed of 8 continuous thymidines to prevent unwanted interactions between exposed single-stranded domains) and boundary bricks (48–nt strands formed by merging a 32-nt full brick and a 16-nt half brick).

5) Save the final sequences pertaining to the targeted 3D formation.

The final structures are self-assembled into their target shapes in one-step reactions. (DNA brick self-assembly is the process by which DNA strands behave as LEGO bricks and adopt a defined arrangement without guidance or management from an external source.)

V. SAMPLE STRUCTURES

Using 3DNA, we have been able to sculpt various shapes of different sizes and ratios. For example, an initial cube of dimensions 8H × 8H × 64B, which measures approximately 20nm × 20nm × 21.6nm was sculpted into shapes like a hollow cube (Fig 4) and a gear (Fig 10). The canvas thus consists of 8 × 8 × 8 molecular pixels, accounting for a total of 1024 Domains, which constitute a sum of 288 single strands of DNA. These 288 strands and further classified into full bricks and half bricks which are 224 and 64 in number. The total number of nucleotides (A,T,G,C) required to build this cube is 8192. Details of the sample files generated for the hollow cube and gear are mentioned below in sections A and B respectively.

A. Hollow Cube

The following inferences can be made about the hollow cube (as shown in Fig 4):

- The hollow cube is obtained by deselecting 256 of the 512 molecular pixels from the canvas.
- The sculpture has the same dimensions i.e., approximately 20nm × 20nm × 21.6nm. This is due to the fact that the deselected pixels are all internal pixels.
- The hollow cube contains 512 domains, which constitute a total of 168 strands, out of which 80 are half bricks and the remaining 88 are full bricks.
- This structure requires a total of 4096 nucleotides for its formation.
- Assuming that the cost per base is U.S. Dollar 0.004, the total cost of the experiment would be 16.3Dollars.

B. 3D Gear

The following inferences can be made about the gear (as shown in Fig 10):

- The gear is sculpted form a 10H × 10H × 80B canvas.
- The gear is obtained by deselecting 440 of the 1000 molecular pixels from the canvas.
- The sculpture has the same dimensions as the initial canvas i.e., approximately 25nm × 25nm × 27nm.
- The gear contains 1200 domains, which constitute a total of 380 strands containing both full and half bricks.
- This structure requires a total of 9600 nucleotides for its formation.
- Fig [11] shows a statistical estimation of the similarity of 8, 7 and 6 bases among the domains. It can be used...
Fig. 11. A graphical analysis of a cube of $10 \times 10 \times 80$ molecular canvas

measure the stability and tolerance of identical bases in the domain sequences of the actual self–assembled structures.

- Assuming that the cost per base is U.S. Dollar 0.004 , the total cost of the experiment would be Dollars 38.4.

VI. CONCLUSION

3DNA has been developed for the sole–purpose of generating a user–friendly, interactive environment for users to envisage their DNA structures, and get the actual DNA sequences required to make the physical formations. With the feature of edit dimensions, user can scale the shape in desire dimension and can view it with different orientations. Thus the output sequences can be experimentally used to make the nanoscale architectures with specified brick design. In the future, we expect to enhance the functionality of the software and enable the user to draw more complex structures.

VII. SOFTWARE AVAILABILITY

The software source code, user manual, and supplementary materials can be downloaded from: http://www.guptalab.org/3dna.

VIII. ACKNOWLEDGMENT

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