Research Article

DNA computing algorithm for realization of DNA Boolean logic based on micro-cantilever deflection

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ABSTRACT

The ability of DNA to perform computation operation has opened a new area of research, popularly known as DNA Computing. This paper contributes by proposing an algorithm, using DNA computing strategy to simulate any kind of logic gate involving a relatively fewer number of biochemical reactions. The specificity of DNA hybridization reaction is used as a key tool in implementing logic operations at the molecular level. For detection of the success of hybridization reactions, the algorithm has been integrated with micro-cantilever. The binding-induced deflection is used to read the output. Re-usability and generalizability are the main strength of the theoretical model presented in this paper.

Keywords: DNA, DNA Computing, Molecular Computing, Hybridization, Logic Gate, Micro-cantilever.

INTRODUCTION

Adleman [1] made the first observation that DNA could be used to solve combinatorial problems, and thus opens a new area of research. Since then, due to growing attention from researchers, the idea of DNA computing has been extends to multiple disciplines, starting from solving NP problems to molecular simulation of logic gates and Boolean circuits. Ogihara and Ray[2] came up with the first DNA Boolean circuit by using constructive approach. The shortcoming of the approach was the excessive use of error prone process like PCR and too much dependence on enzymatic reactions. Secondly the model evaluates Boolean circuit made of only and OR gate and thus lacks the generalizability. M.Amos and Dunne[3] proposed a molecular NAND gate. Run time complexity of Amos's model was proportional to the depth of the circuit, whereas Ogihara's model was proportional to the size of the circuit. Erik [4] simulated another Boolean circuit by using finite splicing system. Mulawka et al.[5] also reported a DNA-NAND gate model using restriction enzyme. Ahrabian and Nawzari [6] demonstrated a DNA fan-in Boolean circuit. Contemporary to those, Liu et al.[7] published a theoretic NAND Gate simulation model, but since the time complexity is linear to the size of the circuit, this model would not function in case of circuits with large number of levels. Mahnaz Kadkhoda [8] claimed to develop a NAND gate in 2006. Ehud Shapiro and Binyamin Gil [9] constructed a DNA logic gate with biomedical application in the field of cancer detection and release drugs. Recently Christy M.Gearheart et al.[10] presents a new homogeneous logic gate. Drawbacks of the previous models were that they lack generalization and involved several error prone biochemical reactions. Generalization is said to be achieved only if a single model could be used to simulate any kind of logic gate. Zoraida et al. [11] added a novel algorithm which claims to fulfill the criteria like generalization, reusability and easy implementation, but it uses blocker sequence which requires great precision while designing. The model has another major demerit of using free-floating DNA strands. It's difficult to keep track of the gate strand in the solution and suffers from the chance of getting lost, as a result it is not reusable. Recently the numbers of work have been reported by Morteza et al. [12], Genot et al.[13] and Wei Li et al. [14,15] relating to DNA circuit simulation. To address the challenges of the above developments, a theoretical model is proposed to simulate Boolean gate with less biochemical reactions without using any blocker sequence. Our method is reusable and excludes error prone process like PCR.

Algorithm to Design Operator Strand

A Boolean variable may be in either OFF state (=0) or in ON state (=1). For 'n' numbers of input variables, maximum possible output states can be expressed in 2^n. If I_1 and I_2 are two Boolean variables, for simplicity of understanding notations are assigned to each variable depending on its values. $I_{1,0}$ and $I_{2,0}$ is assigned when I_1=0 and I_2=0 similarly $I_{1,1}$ and $I_{2,1}$ represents I_1=1and I_2=1 respectively .Complements of the variable are denoted as $\bar{I}_{1,0}$, $\bar{I}_{1,1}$, $\bar{I}_{2,0}$ and $\bar{I}_{2,1}$. Each of the variable notation, i.e. $I_{1,0}$, $I_{2,0}$, $I_{1,1}$ and $I_{2,1}$ has been allotted a unique single stranded $3' \rightarrow 5'$ oligonucleotide sequence and its complementary variables $\bar{I}_{1,0}$, $\bar{I}_{1,1}$, $\bar{I}_{2,0}$ and $\bar{I}_{2,1}$ are assigned with complements of pre-assigned sequences with $5' \rightarrow 3'$ orientation as shown in Table 1. Table 2 shows a general form of a truth table with I_1, I_2 as input variables and Z_i as output.

Table 1: Sequence assignment to variables

Symbol	3' → 5'	Symbol	Complementary 5' → 3'
$I_{1,1}$	TCTGAC	$\bar{I}_{1,1}$	AGACTG
$I_{1,0}$	GCTATT	$\bar{I}_{1,0}$	CGATAA
$I_{2,1}$	GTAACA	$\bar{I}_{2,1}$	CATTGT
$I_{2,0}$	GACTTA	$\bar{I}_{2,0}$	CTGAAT

Table 2: Truth table with two inputs

I_1	I_2	Z_i
0	0	Z_1
0	1	Z_2
1	0	Z_3
1	1	Z_4

In the proposed algorithm all the rows must be scanned one after another and accordingly associated variable notations are inserted into an array. The insertion process depends on the output value, i.e. when Z_i =1, $I_{1,value}$ and $I_{2,value}$ are stored in the array in linear fashion one after another, but when Z_i =0, complements of the variable associated to I_1 and I_2 are inserted i.e. $\bar{I}_{1,value}$, $\bar{I}_{2,value}$ are inserted into the array. Scanning and insertion of input variable into the array is continued until the end of the table is reached. The insertion technique of elements into the array can be summarized as:

$$\text{Strand\_array}[\,] = \begin{cases} \text{Strand\_array}[\text{index}++] \leftarrow I_j & \text{, if } Z_i = 1 \\ \text{Strand\_array}[\text{index}++] \leftarrow \bar{I}_j & \text{, if } Z_i = 0 \end{cases}$$

Where 'j' is the counter to count the number of input variables associated with the truth table. "Strand_array" is the name of the array where variable notations are inserted as elements.

After successful execution of the algorithm, the elements in the array must be replaced by the corresponding DNA strands allotted earlier. The strand obtains at the end is the desired DNA gate stand with orientation 3' → 5'(shown in Table 3.). The above method can be expressed in terms of an algorithm with name "Operator_design". Parameters to be provided are (a) desired truth table (b) a counter 'n' used for counting the number of variables associated with the truth table. (c) a counter 'i' used for counting the rows to be scanned, i.e. i=1,2……2^n. This algorithm can be implemented to simulate logic functionality of any truth table. The pseudo code of the proposed algorithm is shown below.

Operator_design (truth_table, i, n)

{

int i←1, index←0;

WHILE i<=2^n// read all the rows of table

IF (Z_i = =0)//Scanning the output for each row

{

WHILE j<=n //scan all the variables or columns

IF (Ij= =0)

Strand_array[++index] ← $\bar{I}_{j,0}$;

ELSE

Strand_array[++index] ← $\bar{I}_{j,1}$;

ADD 1 to j;

END WHILE

}

IF (Z_i ==1)

{

WHILE j<=n// scan all the variables or columns

IF (Ij==0)

Strand_array[++index] ← $I_{j,0}$;

ELSE

Strand_array[++index] ← $I_{j,1}$;

ADD 1 to j;

END WHILE

}

ADD 1 to i;

END WHILE

}// end algorithm

Strand_array obtained for some two input logic gates after applying our algorithm is shown in Table 3. The 5' end of gate strand is immobilized on the surface of the cantilever. The input strand act as ligand and the gate strand acts as a receptor. The binding between the two single stranded DNA (ssDNA) would cause binding-induced deflection. This deformation due to surface chemical reaction is transduced into measurable quantitative signal.

Input design

For the successful execution of the algorithm, special care must be taken while providing the inputs. Digital inputs must be encoded in the form of DNA sequence without any distortion of information. Its design principle is relatively easy compared to "operator_design" algorithm. The Input sequence is the concatenation of the complements of the desired input variables, Table .4 shows the designated inputs of any gate having I_1 and I_2 as inputs.

Table 3: "Strand_array" for logic gates

Gate	Derived DNA strand
OR	$\bar{I}_{1,0}$ $\bar{I}_{2,0}$ $I_{1,0}$ $I_{2,1}$ $I_{1,1}$ $I_{2,0}$ $I_{1,1}$ $I_{2,1}$
AND	$\bar{I}_{1,0}$ $\bar{I}_{2,0}$ $\bar{I}_{1,0}$ $\bar{I}_{2,1}$ $\bar{I}_{1,1}$ $\bar{I}_{2,0}$ $I_{1,1}$ $I_{2,1}$
NOT	$I_{1,0}$ $\bar{I}_{1,1}$
NAND	$I_{1,0}$ $I_{2,0}$ $I_{1,0}$ $I_{2,1}$ $I_{1,1}$ $I_{2,0}$ $\bar{I}_{1,1}$ $\bar{I}_{2,1}$
XOR	$\bar{I}_{1,0}$ $\bar{I}_{2,0}$ $I_{1,0}$ $I_{2,1}$ $I_{1,1}$ $I_{2,0}$ $\bar{I}_{1,1}$ $\bar{I}_{2,1}$
XNOR	$I_{1,0}$ $I_{2,0}$ $\bar{I}_{1,0}$ $\bar{I}_{2,1}$ $\bar{I}_{1,1}$ $\bar{I}_{2,0}$ $I_{1,1}$ $I_{2,1}$

Table 4: Input Strands equivalent to digital inputs

Input	Derived Input	DNA input strand
0,0	$\bar{I}_{1,0}$ $\bar{I}_{2,0}$	5' –CGATAACTGAAT- 3'
0,1	$\bar{I}_{1,0}$ $\bar{I}_{2,1}$	5' –CGATAACATTGT- 3'
1,0	$\bar{I}_{1,1}$ $\bar{I}_{2,0}$	5' –AGACTGCATTGT- 3'
1,1	$\bar{I}_{1,1}$ $\bar{I}_{2,1}$	5' –AGACTGCATTGT- 3'

Read-Out Mechanism

Read out step is one of the important phases of any DNA computing model. After the successful execution of computation operation, reliability depends on the accuracy of readout technique. Adelman [1], Ogihara and Ray [2], Amos et al. [3] and Mulawka et al. [5] use gel electrophoresis for reading the output. After the execution of gel electrophoresis, the DNA strands are sorted according to their length, i.e. length is the distinguishing criteria. Presence of strands of a particular length is considered as "true" otherwise as "false". Apart from this, fluorescence labels are also used as a readout technique in works like Zoraida et al. [9], Gear heart et al. [10] Wenbin Liu et al[7]. DNA probes like Molecular Beacons are successfully used as nano molecular sensing tools for targeted sequence detection. Thundat et al. [16] proposed the possibility to use cantilever as nano sensors. He observes the deflection of atomic force microscope (AFM) cantilevers due to change in humidity. Since then microcantilever-based sensors emerges as new label-free detection, alternative to conventional biochemical sensors because of its high sensitivity [18- 21], fast response, and parallelization [22-25].

Typically, microcantilever are fabricated on silicon wafer or silicon-on-insulator(SOI) having dimension ranging from tens to hundred micrometers in length, 20 microns wide, and 1 micron thick. SOI wafer is made of three layers; a top layer of single-crystal silicon or silicon nitrite, middle layer consists of silicon oxide layers and the bottom layer consists of single-crystal silicon. The upper surface is chemically modified that has a high affinity towards the receptor molecules and the lower surface is made inert so that all the reaction is restricted to one surface. The mechanical deformation is transduced to measure quantitative signal. The designed single stranded DNA (ss DNA) gate strands are covalently immobilized on the upper surface of the cantilever beam. When solutions of inputs strands are poured on the beam, hybridization results change in surface stress between the upper and lower surface and hence bending the cantilever.

Thundat et al. [16, 17] detected mercury adsorption on cantilever up to pictogram precision. A significant amount of work has been published during the last 20 years where micro-cantilever-based sensors are used in the detection of chemical, DNA hybridization, explosives, bio-molecules, antigen-antibody binding and markers for cancer [18, 19, 23, 24, 26- 34,36-38]. Fritz et al. performed a series of experiments to indicating that micro-cantilever-based sensors have intrinsic sensitivity to discriminate even single nucleotide polymorphism. Hansen et al. [19,23] experimentally showed that the number and position of mismatch pairs affect the deflection of the cantilever and hence could be used to detect mismatches. In Experiment conducted by Stachowiak et al.[30] it is indicated that the effects of the immobilization density, chain length and hybridization efficiencies are coupled, and the cantilever deflection may primarily depend on factors like length and sequence [19], salt concentration [20], temperature [25], grafting density [30], hybridization density [30], moisture concentration [29] and time [18-22,25,30,40,41]. Fritz et al. [18] hypothesized that the cantilever deflection is the result of electrostatic repulsion arises due to the negative charges on the DNA strands and relaxation of steric hindrance. Yue Zhao et al. [42] proposed a model for predicting hybridization induced bending of micro-cantilever based on the minimization of the energy functional that accounts for cantilever bending energy and DNA inter-chain interactions. N.H. Zhang et al. [43] concluded from experiments and theories related to interaction of single stranded DNA immobilized on micro-cantilevers that at high grafting density cantilever deflection is actually controlled by hydration force and not on the effect of electrostatic and conformational entropy.

In the proposed model micro cantilever is used for sensing the hybridization reaction as it has high sensitivity and ability to read the biochemical reaction on the surface.

Formal Validation of the Proposed Model

NAND GATE

The proposed model could be implemented to simulate any logic gate, but in this paper theoretic simulation of the NAND gate is shown. The NAND gate evaluates '1' as output if at least one of the inputs is false. The aim of our algorithm is to design a DNA operator strand which can mimic the functionality of the desired logic gate on providing inputs. The Simulation process mainly takes place in three phases: First, the operator strands are immobilized on a micro cantilever surface. Secondly, input strands are added, bathing the cantilever surface. In third phase finally, output is read out by detecting the presence

of hybridized strand. The operation of DNA-NAND gate with two inputs is shown in Fig 1-5. Gate strand obtained by the method is fixed on the surface of cantilever at 5' end [7]. Input of each DNA-based logic gate is provided once at a time. The input strand has 5' → 3' orientation. Since the operator strand (gate strand) and input strand are complementary they tend to hybridize. Table.4 shows derived input strand.

Case a

When I_1 =0 and I_2 =0: Inputs are provided in the form of 5' $\bar{I}_{1,0}$ $\bar{I}_{2,0}$ 3' strand which is DNA equivalent to binary 0,0. The gate strand and input strand successfully hybridize which results in deflection of the cantilever surface as shown in Fig .1.

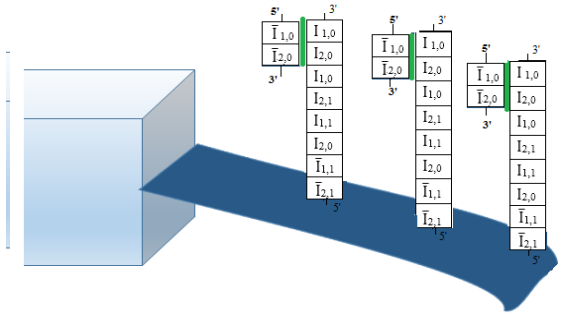

Figure 1: Input (0, 0) is poured.

Case b: When I_1 =0 and I_2 =1: As shown in Table 4 binary inputs 0,1 is provided in the form of 5' $\bar{I}_{1,0}$ $\bar{I}_{2,1}$ 3' strand which results in successful hybridized with the 3' $I_{1,0}I_{2,1}$ 5' section of the gate strand as shown in Fig. 2.

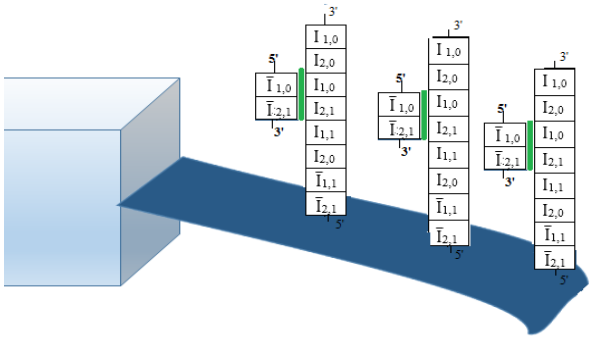

Figure 2: Input (0, 1) is poured.

Case c: When I_1 =1 and I_2 =0: In a Boolean Circuit NAND gate produce output as '1' when inputs are in the form 1,0. In our model this case can be evaluated on providing input in the form of 5' $\bar{I}_{1,1}$ $\bar{I}_{2,0}$ 3' strands and allowed to anneal as shown in Fig.3.

Case d

When I_1 =1 and I_2 =1.

Input (1, 1) of the DNA- NAND gate is provided as 5' $\bar{I}_{1,1}$ $\bar{I}_{2,1}$ 3' strands. No hybridization has occurred since the operator strand (gate strand) doesn't contain any complementary segment to let the input strand hybridize, i.e. both contain 3' $\bar{I}_{1,1}$ $\bar{I}_{2,1}$ 5' fragment (Fig.4).

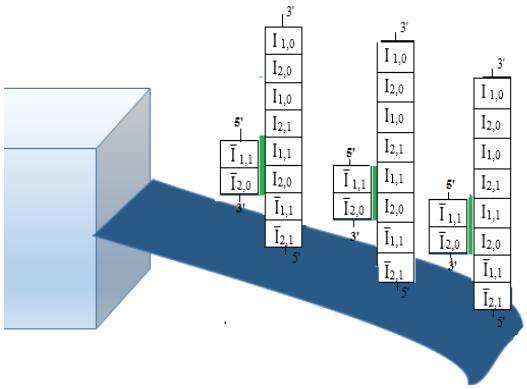

Figure 3: Input (1, 0) is poured.

It is already stated that unsuccessful hybridization is read as '0' as there is no deflection. Fig 5. Shows the hybridization reaction of the DNA-NAND gate on the basis of four input cases.

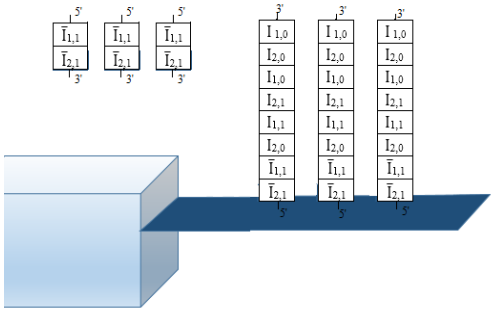

Figure 4: Input (1,1) is poured

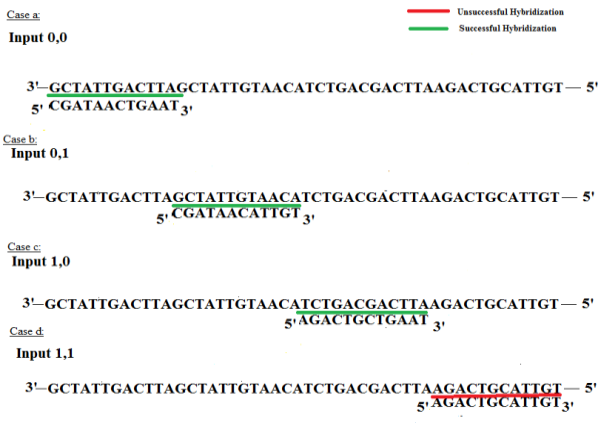

Figure 5: Reaction of NAND gate.

CONCLUSION

In this paper an algorithm is proposed to design a DNA based logic gate. Problems which can be expressed in terms of truth table can be easily simulated by this algorithm. The theoretical model involves only hybridization process and avoids use of error prone process like PCR. The hybridization-induced bending of the micro-cantilever is used to read the output. The paper focuses on issues like re-usability and reliability. The

immobilization of DNA strands on the cantilever surface enhances the reusability property of the model. The length of the gate strand is proportional to the number of input variables and thus poses a constraint in the maximum number of inputs allowed. Though this model seems to be theoretically sound, but we acknowledge that the physical realization in the laboratory involves a practical difficulty. Future works must be carried out emphasizing the implementation aspects.

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