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Assessing the potential of surface-immobilized molecular logic machines for integration with solid state technology

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ABSTRACT

Molecular computation with DNA has great potential for low power, highly parallel information processing in a biological or biochemical context. However, significant challenges remain for the field of DNA computation. New technology is needed to allow multiplexed label-free readout and to enable regulation of molecular state without addition of new DNA strands. These capabilities could be provided by hybrid bioelectronic systems in which biomolecular computing is integrated with conventional electronics through immobilization of DNA machines on the surface of electronic circuitry. Here we present a quantitative experimental analysis of a surface-immobilized OR gate made from DNA and driven by strand displacement. The purpose of our work is to examine the performance of a simple representative surface-immobilized DNA logic machine, to provide valuable information for future work on hybrid bioelectronic systems involving DNA devices. We used a quartz crystal microbalance to examine a DNA monolayer containing approximately 5×10^{11} gates cm^{-2}, with an inter-gate separation of approximately 14 nm, and we found that the ensemble of gates took approximately 6 min to switch. The gates could be switched repeatedly, but the switching efficiency was significantly degraded on the second and subsequent cycles when the binding site for the input was near to the surface. Otherwise, the switching efficiency could be 80% or better, and the power dissipated by the ensemble of gates during switching was approximately 0.1 nW cm^{-2}, which is orders of magnitude less than the power dissipated during switching of an equivalent array of transistors. We propose an architecture for hybrid DNA-electronic systems in which information can be stored and processed, either in series or in parallel, by a combination of molecular machines and conventional electronics. In this architecture, information can flow freely and in both directions between the solution-phase and the underlying electronics via surface-immobilized DNA machines that provide the interface between the molecular and electronic domains.

1. Introduction

The use of biological molecules to perform computation was pioneered by Adleman (Adleman, 1994), who demonstrated that it was possible to solve an instance of the Hamiltonian path problem with DNA strands. DNA is particularly attractive for molecular computation because the interactions between oligonucleotides are highly predictable and programmable (through the base sequence), and the raw materials are relatively cheap and easy to acquire. Various types of logic gates have been assembled using DNA (Seelig et al., 2006; Stojanovic et al., 2002), and they have also been implemented in biocompatible nanorobots that could in principle be used for smart drug delivery (Douglas et al., 2012). It has also been shown that simple gates can be combined to form adders (Lederman et al., 2006) and subtractors (Lin et al., 2015). Recently, DNA circuits have been proven to be capable of computing a square root (Qian and Winfree, 2011) and a DNA-based neural network has been demonstrated (Qian et al., 2011). Mixtures of DNA and DNA-manipulating enzymes have been used for computation, and enabled the implementation of a finite automaton (Benenson et al., 2001). Later, Costa Santini et al. (2013) constructed a DNA finite state machine in which transitions from one state to another were triggered by a clock signal.

Most of these previous achievements involved solution-phase reactions, and it is the use of freely diffusing molecules that underpins many of the challenges that currently limit DNA computation. At present, most techniques used to read out the results of computations involve the use of probes or reporter complexes which carry fluorescent labels. This severely limits the potential for multiplexing and complicates the measurement process. Furthermore, it is difficult to control the state of the ensemble of biological machines without adding new molecular species. We suggest that

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it would be possible to address these challenges through development of hybrid DNA-electronic technology, in which computational machines made from DNA are immobilized directly onto underlying electronic circuitry.

In our proposed hybrid approach, information would be able to pass freely in both directions across the molecular–electronic interface. The underlying electronic components could interrogate the state of the surface-immobilized DNA machines, apply logic processing to the data received and act accordingly to regulate the molecular state. This would enable multiplexed readout of the DNA machines, eliminating the limitations imposed by the use of fluorescent labels, and could provide a new approach for the construction of cascades of multiple DNA machines, where the underlying electronics would be used to form connections between individual devices.

Importantly, biomolecular processes in solution offer huge potential for parallelization of computation, and this could be harnessed in a hybrid system, while it would also be possible to interface the technology with biological matter for applications in biology and medicine. Conventional semiconductor devices could be used to perform high-speed operations, complementing the slow bio-compatible molecular elements.

Surface-immobilization of DNA machines is integral to our proposal for hybrid computation, but to date, surface-based DNA computation has received comparatively little attention. However, with a combination of DNA molecules, an endonuclease, and a ligase, it is possible to implement a finite automaton on a surface, as described by Soreni et al. (2005), while Frezza et al. (2007) proved that DNA strand displacement can be used to operate and cascade logic gates immobilized on the surface of a gold nanoparticle.

Another strategy presented for surface-based DNA computing is as follows. DNA molecules encoding 'words' are immobilized on a surface, and each word contains both a representation of variable values and a label for directing word selection through controlled hybridization. Application of specific oligonucleotides and DNA-processing enzymes to the surface enables a computation to be performed, where some strands are eliminated and others are retained. This approach was used by Liu et al. (2000) to solve a four-variable four-clause 3-SAT (satisfiability) problem, and Wang et al. (2000) extended the technique to accommodate DNA strands containing multiple words. Subsequently, Su and Smith (2004) showed that such a model could be used to construct a universal computer based on NOR and OR gates.

In this paper we present a quantitative assessment of the performance of a surface-immobilized OR gate made from DNA and driven by DNA strand displacement. It is not directly linked to electronic circuitry, but by studying the behaviour and properties of the gate when it is immobilized on the surface of a gold electrode we can obtain valuable insights into the advantages and constraints of hybrid computation. We have designed it to serve as a useful case study to inform future development of systems of the kind described above, and to be a representative example of a surface-immobilized molecular logic machine.

The implementation of our logic gate is very similar to that of Frezza et al. (2007), but our design is simpler and our choice of experimental technique enables us to use a reporter-free method to read the output and to observe the kinetics of switching. We also examine the behaviour of the gate when it is restored to its original condition and switched repeatedly. Based on our results, we present a quantitative assessment of the performance and attributes of the device, in terms of speed, error frequency, feature size, potential for parallel processing, and power dissipation, in comparison with conventional silicon technology. We proceed to suggest a possible architecture for hybrid systems.

2. Experimental setup and methods

Our DNA OR gate is shown in Fig. 1. To study the operation of our surface-immobilized DNA logic gate, we used the technique of Quartz Crystal Microbalance with Dissipation monitoring (QCM-D), which involves the measurement of the resonant frequencies of oscillation and associated Q-values for an acoustic wave generated by a driven piezoelectric sensor crystal, as described in Dixon (2008). The crystal has a gold electrode on either side, across which the driving voltage is applied. One of the electrodes is exposed to a solution, such that molecules from the solution can bind to the surface or molecules from the surface can dissociate into solution. In this paper we consider changes in resonant frequency, which are closely correlated with changes in surface-immobilized mass. In general, an increase in frequency implies that mass has been lost from the surface, while a decrease in frequency implies that mass has been added. Measurements can be made at multiple resonant frequencies (overtones), and here we focus on the 13th overtone, which has the shortest penetration depth – equal to about 70 nm in pure water - and is thus less sensitive to changes in the bulk solution. The apparatus also allows us to measure the dissipation of energy by the acoustic wave as it propagates from the sensor through the molecular layer and into solution, where the dissipation is defined as the inverse of the quality factor.

The experimental procedure we used is as follows. All experiments were performed in 1× TE buffer in the presence of 1 M NaCl and DNA sequences are provided in Table 1.

The logic gate consists of a partially double-stranded DNA molecule (the 'capture complex') and a 'set' strand G that binds to

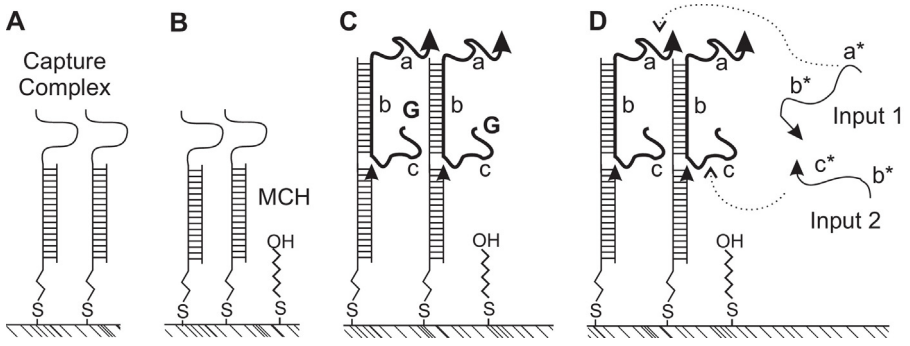

Fig. 1. Testing a surface-immobilized OR gate made from DNA. (A) The pre-formed capture complex is immobilized on the surface to form a monolayer of DNA gates. (B) The backfilling agent mercaptohexanol (MCH) is used to fill in spaces between the DNA molecules in the monolayer. (C) Strand G (bold line) is hybridized with the capture complex (the gate is 'set'). Strand G consists of three domains, indicated by 'a', 'b' and 'c'. (D) The DNA-functionalized surface is exposed to input DNA strands. The domains labelled a* and c* on the input strands bind to the recognition domains of G, as indicated by the dotted arrows. When G has been displaced by either input, the system reverts to the state shown in (B) and must be reset by re-hybridization with G before it can respond to a new input. The displacement of G corresponds to the OR operation.