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DNA-Based Photonic Logic Gates: AND, NAND, and INHIBIT

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Molecular computation is a term that includes a number of distinct bottom-up approaches toward the design of molecular scale electronics, chemical, and biological computers.[1−8] By analogy to conventional microprocessors which use elementary logic gates[9] to form electronic circuits capable of performing Boolean logic, design of addressable molecular logic gates has been a major goal in the field of molecular computation. One approach has been to use organic and/or inorganic materials in the fabrication of patterned nanoscale electronic logic gates and circuits.[2,3] An alternative approach has employed molecular or supramolecular systems to create logic gates that respond to chemical and photonic signals.[5] The advantage afforded by electronic systems in the assembly of circuits lies in its speed and the common input/output signal used, the electron, which permits gates to easily be connected (wired) together.[2−4] By analogy, if a supramolecular system can be developed using a single class of input/output molecules, then chemical circuits should also be within reach. Herein we report progress toward this goal by using the recognition properties of DNA to create photonic logic gates capable of AND, NAND, and INHIBIT logic operation.

AND logic is represented by the situation where the output of an AND gate is true only if both inputs are true.[9] Chemical AND gates have been developed by using two chemical inputs to influence the fluorescent emission of another molecule in solution, which represents the output of the gate.[5b] To create a DNA-based AND gate we coupled two molecular recognition events in series to cause a change in the photonic output of the gate in solution.

The DNA-based AND gate is a 16-mer oligonucleotide modified at the 3′-terminus with a carboxy fluorescein moiety. The two inputs are a complementary 16-mer oligonucleotide, ANDin1 (AND gate input 1), and the DNA minor groove binder Hoechst 33342,[10−11] AND gate input 2). The Hoechst 33342 has an absorption maximum at 350 nm and a broad emission maximum at 450 nm, which overlaps with the 490 nm absorption maximum of the 3′ fluorescein attached to the AND gate. The output of the gate is measured upon excitation at 350 nm by fluorescence output at 520 nm, the emission wavelength of fluorescein. If energy transfer[12] between ANDin2 and the 3′ fluorescein of the AND gate takes place, the system will have a strong emission at 520 nm, otherwise there should be little or no fluorescence at this wavelength.

Experiments were performed by testing the AND gate oligonucleotide with the four possible input combinations (0,0; 1,0; 0,1; 1,1) for a binary gate (see Figure 1). The measured fluorescence output of the AND gate at 520 nm was 0 except for the 1,1 combination, which had a strong fluorescent signal. Mechanistically this output is the result of two sequential molecular recognition events. The first is duplex formation between the AND gate and ANDin1. This interaction creates a minor groove binding site for the Hoechst 33342, ANDin2, placing it near the 3′ fluoroscein of the AND gate. The proximity of the two dyes in the 1,1 complex results in FRET[12] from Hoechst 33342 dye to the 3′ fluoroscein

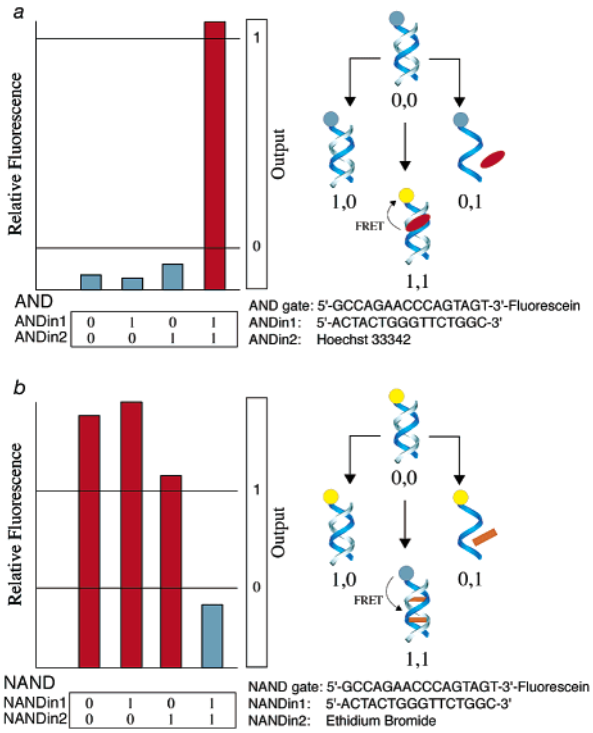

Figure 1. Relative fluorescence emission of the AND and NAND gates and schematic representation of the molecular basis of the logic operations. (a) The molecular components the DNA-based AND gate include the AND gate: a 16-mer oligonucleotide with a 3′ flourescein (blue oligo); the first input ANDin1: a 16-mer complementary oligonucleotide (gray oligo); and the second input ANDin2: Hoechst 33342 a DNA minor groove binder (red oval). The scheme illustrates that only in the presence of both inputs (1,1) the duplex formation between the AND gate and ANDin1 facilitates the binding of ANDin2 nearby the 3′ fluorescein moiety of the AND gate, thereby enabling photonic output (1) due to FRET between ANDin2 and the AND gate. (b) The NAND gate inverts the AND gate by using ethidium bromide instead as the second input NANDin2 (orange rectangle). The spectral overlap between the ethidium bromide absorption band and the fluorescein emission spectra causes quenching of the fluorescein output (0) only when both inputs are present (1,1), thus creating the NAND logic operation.

and a strong emission at 520 nm. Neither of the AND inputs alone causes a change in the fluorescence emission of the AND gate, which is evident by the 1,0 and 0,1 combinations. The pattern of chemical inputs to fluorescence outputs successfully recreates the truth table for an AND gate and demonstrates the feasibility of using DNA in logic gate design.

To demonstrate the flexibility of the DNA-based logic gates, we used the AND gate as a platform to design a NAND gate and an INHIBIT gate. In electronics NAND logic is the result of sending an AND gate through an inverter which causes all 0 states to switch to 1 and vice versa. Thus, NAND logic has an output of 1 for all combinations of binary inputs except the 1,1 situation where the

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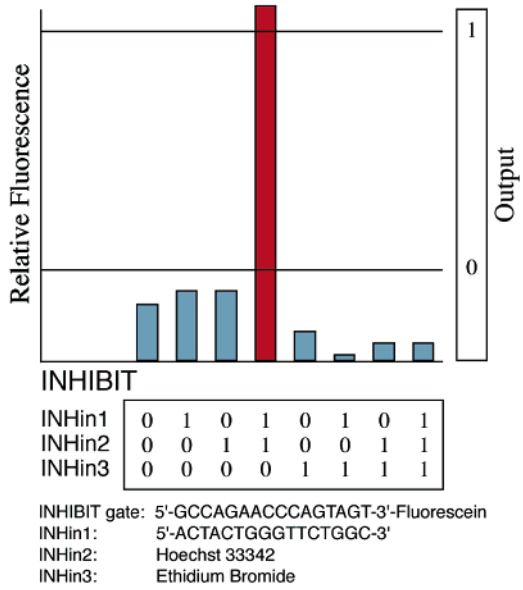

Figure 2. Bar graph representation of the output of the INHIBIT gate at 520 nm upon irradiation at 490 nm. In the absence of INHin3 the INHIBIT gate reduces to an AND gate. The presence of INHin3 quenches the fluorescence at 520 nm to recreate INHIBIT logic.

output now becomes 0. The DNA-based AND gate is converted to a NAND gate by simply using ethidium bromide as NANDin2 (NAND gate input 2) instead of the Hoecsht 33342 and switching the excitation wavelength to 490 nm (excitation wavelength of fluorescein) instead of 350 nm. NANDin1 (NAND gate input 1) and the NAND gate are identical to ANDin1 and the AND gate. In the absence of any inputs the excitation of the NAND gate at 490 nm results in fluorescence emission from the 3′ fluorescein of the NAND gate. Addition of either NAND inputs individually has no effect on the emission and the photonic output of the gate remains high. Conversely, the presence of both NAND inputs results in the formation of a duplex between the NAND gate and NANDin1 allowing the ethidum bromide, NANDin2, to intercalate into the DNA duplex structure placing it near the fluorescein moeity.[13] The spectral overlap between the 3′ fluorescein and the ethidium bromide, NANDin2, leads to quenching of the fluorescein emission for the 1,1 combination.[10,13] The resulting pattern of photonic output versus chemical inputs mimics that of an electronic NAND gate with a low output state occurring only in the presence of both inputs.

By combining the molecular components of AND and NAND gates we were also able to construct a three-input INHIBIT gate.[5d] The inputs for the INHIBIT gate are the complementary 16-mer oligonucleotide (INHin1, INHBIT gate input 1), Hoechst 33342 (INHin2), and ethidium bromide (INHin3) (see Figure 2). The output of the gate is determined by irradiation at 350 nm and measuring the fluorescence at 520 nm. The third input, the inhibit bit, inhibits the output of the gate regardless of what other inputs are present. In the absence of INHin3 the DNA-based INHIBIT gate reduces to the AND gate with a strong fluorescence in the presence of INH inputs 1 and 2. The addition of ethidium bromide as INHin3 effectively quenches all fluorescence to give a truth table of an INHIBIT gate. Chemically, this phenomenon results from the quenching of the fluorescence output that results when the ethidium bromide binds near the fluorescein such as in the 1,1 combination of the NAND gate. Together these results validate the use of DNA as a specific supramolecular platform for fabrication of addressable molecular logic gates.

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Supporting Information Available: Methods and fluorescence spectra (PDF). This material is available free of charge via the Internet at http://pubs.acs.org.

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