



Supporting Information

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A Unidirectional DNA Walker Moving Autonomously Along a Track,

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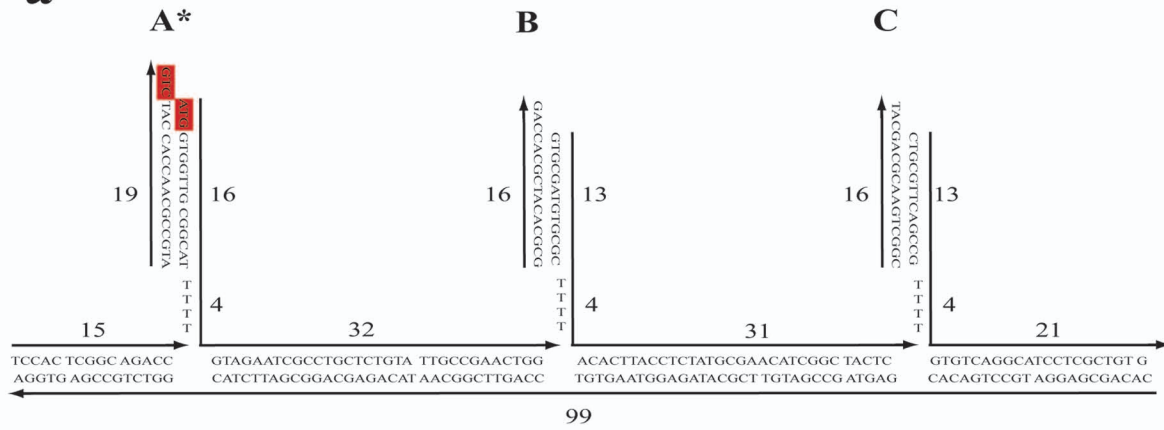
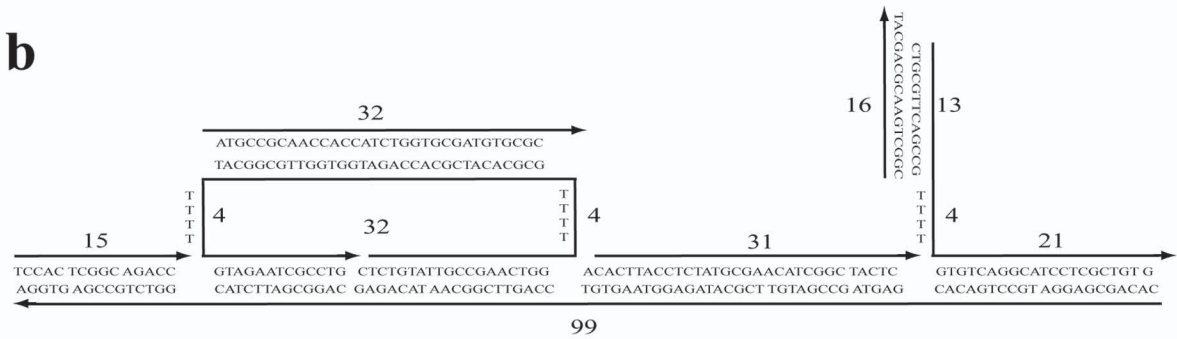
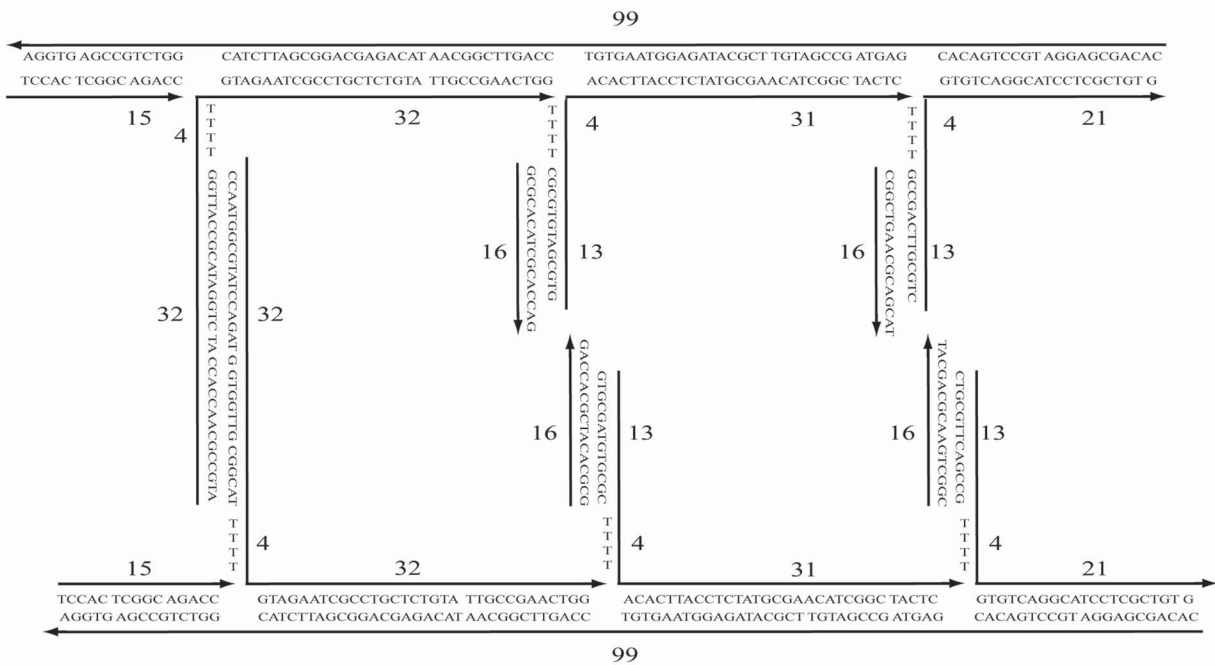
Supplemental Figure S1. DNA strand structure and sequences. **a)** Base sequences of the oligonucleotides that make up the molecular device. **b)** and **c)** Base sequences of the oligonucleotides used to construct the monomer and dimer control molecules described in the caption to Supplemental Figure S3.

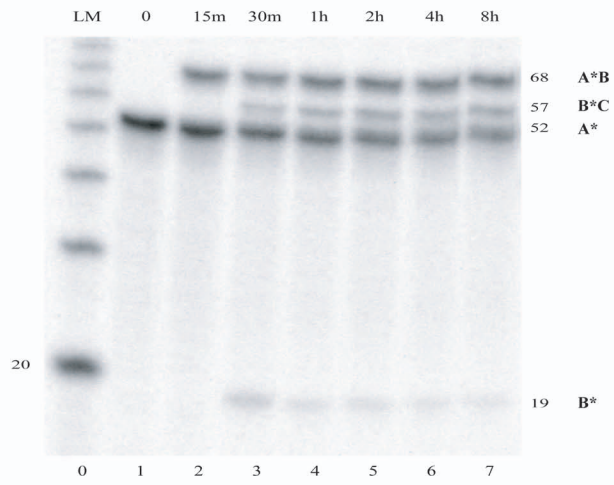
Supplemental Figure S2. Time course experiment. Supplemental Figure S2 is an autoradiograph of a 20% denaturing polyacrylamide gel showing the time course of the device's motion under conditions corresponding to Figure 2b Lane 3. Lane 0: 10 bp ladder marker. Lane 1: device with no enzymes (control). Lanes 2-7 contain samples incubated with T4 ligase and PflM I at 37 °C for 15 minutes, 30 minutes, 1 hour, 2 hours, 4 hours, and 8 hours respectively. The monotonic increase in the concentration of the product B*C, and the decrease in the concentration of the intermediate B* after the first 30 minutes, are consistent with the designed unidirectional motion of the walker.

Supplemental Figure S3. Test for inter-molecular reactions. Complexes produced during the operation of the device were analyzed using a native gel to test for the formation of dimers caused by cross-linkage of two devices. **a** and **c** depict the molecular designs of 'monomer' and 'dimer' control complexes. The designs of the controls are shown in Figure S1b and Figure S1c respectively. The control complexes do not have exactly the

same sequences or structures as the corresponding states of the device; they have approximately the same structures and are designed to migrate at approximately the same rates without forming higher multimers. The monomer control corresponds approximately to the state of a single device at the end of process I or III in Figure 1c. The dimer control represents an intermolecular complex formed by ligation of anchorages on different motors. **b).** Autoradiograph of the 8% native polyacrylamide gel used to test for inter-molecular reactions. The assembled device system was incubated at 37 °C in hybridization buffer supplemented with ATP and BSA and in the presence of various combinations of enzymes. Lane 1: labeled monomer control. Lane 2: device with no enzymes (control). Lane 3: device with T4 ligase. Lane 4: device with T4 ligase, endonucleases PflM I and BstAP I. Lane 5: labeled dimer control. No dimer band was detected in Lanes 2-4, indicating the absence of inter-molecular interactions during the operation of the device.

We note that there is a slight displacement between bands in Lanes 1 and 2, and a matching broadening of bands in Lanes 3 and 4. This is consistent with the hypothesis that a device with no linkages between its anchorages (present in Lane 2 and as part of the population in Lanes 3 and 4) migrates slightly more slowly than a device with two anchorages ligated together (control Lane 1 and part of the population in Lanes 3 and 4).

a**b****c**



a
Monomer control

