Resonator nanophotonic standing-wave array trap for single-molecule manipulation and measurement

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Nanophotonic tweezers represent emerging platforms with significant potential for parallel manipulation and measurements of single biological molecules on-chip. However, trapping force generation represents a substantial obstacle for their broader utility. Here, we present a resonator nanophotonic standing-wave array trap (resonator-nSWAT) that demonstrates significant force enhancement. This platform integrates a critically-coupled resonator design to the nSWAT and incorporates a novel trap reset scheme. The nSWAT can now perform standard single-molecule experiments, including stretching DNA molecules to measure their force-extension relations, unzipping DNA molecules, and disrupting and mapping protein-DNA interactions. These experiments have realized trapping forces on the order of 20 pN while demonstrating base-pair resolution with measurements performed on multiple molecules in parallel. Thus, the resonator-nSWAT platform now meets the benchmarks of a tabletop precision optical trapping instrument in terms of force generation and resolution. This represents the first demonstration of a nanophotonic platform for such single-molecule experiments.
Optical trapping is a cornerstone biophysical technique to investigate biological systems and has allowed unprecedented insight into molecular interactions, including measuring the forces, torques, and step sizes of motor proteins, examining the kinetics of protein unfolding, probing the biophysical properties of DNA and RNA, and disrupting proteins bound to DNA. This technology has been hampered by the requirements of specialized laboratory space and complex tabletop equipment, as well as the inherent low throughput of manipulating one molecule at a time.

The second-generation nSWAT was instead based on Si$_3$N$_4$ sample heating that is not desirable for biological experiments. The nSWAT creates an array of traps by recycling optical traps, the nSWAT creates an array of traps by recycling the input light path (Supplementary Movie 2). This return light may cause back-reflection to the laser source and induce waveguide burning near the input coupling region, limiting these devices to low-force applications.

In this work, we present the next-generation nSWAT, which can generate sufficient force for precision single-molecule measurements. This design incorporates a critically-coupled design which significantly minimizes the return light, while permitting local light enhancement at the trapping region. Furthermore, we have incorporated a novel microheater modulation scheme to enable sustained high force generation over a long manipulation distance. These two features have permitted significant trapping force enhancement. These enhancements have enabled standard optical trapping applications, including stretching DNA molecules to measure their force-extension relations, unzipping DNA molecules, and disrupting and mapping protein–DNA interactions.

**Results**

**Resonator-nSWAT design and fabrication.** The current nSWAT devices were designed and fabricated by integrating Si$_3$N$_4$ nanophotonic resonators, 3-dimensional (3D) microelectronic structures, and a microfluidic channel all on-chip, in a sophisticated, multi-layered configuration (Fig. 1b, c; "Methods"; Supplementary Figs. 1–3; Supplementary Methods 1–4). An external 1064 nm laser is coupled fiber-optically to each device through a tapered nanophotonic waveguide. The light is then split via a 50/50 beam splitter into two paths, which are directed to two separate nSWATs that work in conjunction in an experiment. Stable coupling is maintained via a suspended sample holder design (Supplementary Fig. 4; Supplementary Method 5). The single-molecule sample is sent into the nSWAT flow chamber ("Methods") through a gravity flow cell system (Supplementary Method 6). In each nSWAT, the light passes through another 50/50 beam splitter that creates counter-propagating waves to form a standing wave in a loop which also serves as the bus waveguide for coupling to an adjacent resonator. Thus, a standing wave is also formed in the resonator, where the trapping region resides, and whose resonance is tuned via on-chip ‘resonator heaters’ located over the resonator. To translocate the array trap, ‘translocation microheaters’ located over the bus waveguide modulate the phase of the light to control the position of the standing wave in the resonator (Supplementary Movie 3). Thus, the two nSWATs are independently controlled and tuned.

**Critical coupling and resonance tuning.** To limit the return light, each resonator is designed to operate near its critical-coupling condition (Fig. 2a; Supplementary Fig. 5). When the resonator is tuned to resonance, nearly all light in the bus waveguide is coupled into the resonator, and the return light is greatly minimized. Because the trapping region resides at the resonator, scattering occurs at the boundaries of the etched fluid pool and at the trapped beads, inevitably reducing the quality factor (Q) of the resonator. Fortuitously, the resulting resonance broadening reduces the sensitivity of the critical-coupling condition to the separation distance between the bus waveguide and the resonator so that critical coupling can be realized within the tolerance of the fabrication uncertainties. In addition, the resonance condition is also less susceptible to changes in the number of beads trapped on the waveguide.

For each resonator, we measured its Q via the scattered light of the intensity samplers of the resonator and the return light via scattered light at the ‘return’ light sensor (Supplementary Fig. 6; Supplementary Method 2; Supplementary Method 4). As shown in Fig. 2a, as the resonator is tuned towards resonance, the light intensity in the resonator peaks, while the return laser light...
Intensity decreases to a minimum. We found that the phase response curve of a resonator is symmetric at lower input laser powers but becomes asymmetric with an increase in the laser power. This asymmetry is indicative of nonlinearity due to a light-induced thermo-optic effect. For our devices, we found that $Q$ is $\sim2 \times 10^5$ at 60 mW of input laser power and decreases to $\sim0.5 \times 10^5$ at the 1.3 W of input laser power typically used in subsequent DNA stretching and unzipping experiments (Supplementary Fig. 7).

Microheater power overshoot to achieve high force. To precisely translocate an array of beads on an nSWAT over a long distance, we ramp the power in a pair of translocation microheaters (Fig. 2b). To translocate over a distance greater than one spatial period of the trapping potential, we rapidly reset the power to each microheater after the trap array has been moved by precisely one period of the trapping potential before re-ramping its power. This process is repeated until the translocation reaches the desired distance. While this method permits long-distance translocation without overheating the microheaters, it may also restrict the force generation capacity if the reset is not sufficiently rapid. During the brief reset, a bead initially trapped may drift in the reverse direction under the influence of the reverse moving trapping potential. This drift becomes more evident if the bead is also experiencing an external force opposing the translocation (e.g., from an attached DNA molecule). Based on analysis of the non-linear dynamics of the bead motion in an array trap (Supplementary Fig. 8), the slower the reset speed and the higher the external force, the greater the drift distance. If the bead drifts beyond half of the spatial period of the trapping potential, a stable trapping condition can no longer be sustained, and during the reset, the bead will slip along the waveguide.

Thus, rapid reset is crucial for obtaining a high trapping force in a translocating nSWAT. Our analysis shows that if the bead is under an external force of 30 pN, the microheater reset needs to be 2 $\mu$s in order to obtain 90% of the maximum trapping force. This reset speed is significantly faster than that previously demonstrated (~30 $\mu$s)\textsuperscript{32,36}. Although the reset speed can be improved by placing the microheater closer to the waveguide, this solution leads to greater light loss due to increased scattering by the microheater. Instead, we have circumvented this problem by applying a brief overshoot (~2 $\mu$s) to one of the translocation microheaters at the beginning of the reset to over-drive this microheater. As shown in Fig. 2c and Supplementary Fig. 9, this method reduces the effective microheater reset time to 1.0 $\mu$s (Supplementary Method 7). Therefore, the microheater reset speed is no longer a limiting factor for high force generation.
Single-molecule applications. We then evaluated the performance of this device with three standard single-molecule optical trapping experiments. First, we stretched DNA to measure the force-extension relation of DNA. Here, we first sorted an array of DNA tethers—single molecules of DNA with a bead attached at each end—from a mixture of other bead species, using a method similar to what has been described ("Methods"; Supplementary Fig. 10a). Subsequently, we stretched this array of DNA molecules to ~15 pN by translocating one nSWAT relative to another nSWAT held stationary. During the stretch, both z and x positions of all beads in the array were monitored continuously using an image-based tracking technique ("Methods"), providing a direct measurement of the extension (end-to-end distance) of each DNA molecule in the array (Fig. 3a). For each DNA tether, \( F_z \) (or \( F_x \)), the force along \( ^z \) (or \( ^x \)), was also determined using the \( z \) (or \( x \)) displacement of the bead held in the stationary nSWAT and the calibrated trap stiffness (Fig. 3b; "Methods"; Supplementary Figs. 11 and 12; Supplementary Method 8). The net force \( F \) on the DNA was then determined from these force components. Thus, the force-extension relation, characteristic of DNA elastic properties, was simultaneously measured for each DNA molecule in the array.

Figure 3c shows an experiment where the force-extension relations from seven DNA tethers were obtained in parallel. Six of the seven traces show an agreement with the relation expected for a single dsDNA molecule under the experimental condition used ("Methods"). One trace deviated from expectation and is consistent with a tether formed by two dsDNA molecules instead of one, a configuration that occasionally occurs due to the stochastic nature of the tether formation process. This experiment is akin to DNA stretching using conventional optical tweezers, except that manipulation is performed on multiple DNA molecules simultaneously, illustrating the capacity of nSWAT for parallel precision force measurements on-chip.

Next, we unzipped DNA by mechanically separating a double-stranded DNA (dsDNA) molecule into two single-stranded DNA (ssDNA) molecules (Fig. 4a; Supplementary Fig. 10b). Here, the two strands of dsDNA were suspended between two beads via two dsDNA adapter arms, and the distal end of the dsDNA was capped by a DNA hairpin. As an array of such tethers was
simultaneously stretched, the force and extension relation of each DNA molecule in the array was measured concurrently (Fig. 4b). At the beginning of each trace, the force in the DNA was insufficient to unzip the dsDNA, so the force-extension curve followed that of dsDNA from the two arms. Once the arms were extended, and the force rose to ~12 pN, unzipping the dsDNA began with unzipping force fluctuating in a manner fully predictable according to the DNA sequence. When the unzipping fork encountered the end-capped hairpin, the hairpin was fully unfolded, and further stretching extended both the ssDNA and the dsDNA arms, with the endpoint of the hairpin mimicking a strongly bound protein that resists unzipping20. For each trace, the measured force-extension curve (Fig. 1b) was correlated with the predicted force and converted to force versus the number of base pairs (bp) unzipped (Fig. 4c), using the elastic properties of dsDNA and ssDNA (Supplementary Method 9). The location of the endpoint of the hairpin was identified by a force rise above the baseline at the end of the trace. We found that the hairpin was located with base-pair accuracy and precision (bottom plot of Fig. 4c), representing the first demonstration of any nanophotonic platforms to achieve a level of performance comparable to that of a conventional precision optical trapping instrument20.

Finally, we applied the DNA unzipping technique for parallel mapping of protein–DNA interactions on nSWAT (Fig. 5a). Just as with a DNA hairpin, a bound protein on the DNA is also able to resist unzipping, producing a similar force rise when encountered by the unzipping fork14,18, a technique also termed ‘unzipping mapper’21. We first unzipped an array of dsDNA molecules, each containing a binding site for the restriction enzyme, ZraI, which targets a unique 6-bp DNA sequence. For the dsDNA unzipping segment shown in Fig. 5b, there is a ZraI binding site centered at 859 bp from the start of the unzipping. To enable ZraI binding without its DNA enzymatic cutting activity, the experiment was carried out in the absence of magnesium which is required for cutting. Under the experimental conditions used, we found that a fraction of the traces showed a force signature consistent with a bound ZraI: a force rise was detected at a ZraI binding site, followed by a return of the force to the unzipping baseline after the unzipping fork passed the ZraI site. Thus, the nSWAT generated sufficiently strong forces to disrupt a bound ZraI, with a mean disruption force of 15.7 ± 1.5 pN (mean ± SD).

We then used the unzipping mapper to locate a bound dCas9, which is the non-catalytic mutant of Cas9 and has been broadly used as a strong roadblock for DNA processing enzymes42. When dCas9 is complexed with a single guide RNA (sgRNA), it targets a 20-bp DNA sequence complementary to the sgRNA43. We designed the sgRNA to target a sequence centered at 1145 bp from the start of the unzipping. We unzipped an array of DNA molecules, each containing a bound dCas9, with the unzipping...

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**Fig. 3 Measuring force-extension curves of DNA on nSWAT.**

*a* Force diagram of a DNA tether being stretched. A DNA molecule, tethered between two 380-nm polystyrene beads, was stretched by translocating one nSWAT (top) relative to a stationary nSWAT (bottom). The positions of both beads were monitored via image tracking. The total force along the DNA was obtained from force components along x and z, using the displacements of the bead in the stationary trap, Δx and Δz (“Methods”; Supplementary Figs. 11 and 12; Supplementary Method 8). This method of force measurements allows a more accurate determination of force by using both measured Δx and Δz.

*b* Example of force measurements of a DNA tether being stretched. Data were taken at 1000 Hz (gray), and smoothed to 20 Hz (black).

*c* Simultaneous force-extension measurements of dsDNA molecules. The left panel shows an optical image of seven dsDNA tethers that were simultaneously stretched between a stationary nSWAT and translocating nSWAT. The red lines were manually added to indicate the locations of the dsDNA molecules which are not visible on this image. The resulting measured force-extension curves are shown in the right panel, along with the theoretical prediction (black curves; “Methods”). Each curve is in agreement with that predicted for a single dsDNA molecule, except for tether 4, which is consistent with a double dsDNA tether. Source data for this Figure are provided as a Source data file.
fork approaching the bound dCas9 from the RNA/DNA hybrid side. For each trace, we detected a force rise within the sgRNA target region, consistent with Cas9 interaction sites with DNA from previous studies. Although nSWAT did not disrupt the bound Cas9, nSWAT could still accurately map the Cas9 location because the force was significantly greater than the baseline force. The precision of the detected ZraI binding site was 0.6 bp, similar to that of the hairpin, while the precision of the detected Cas9 binding sites was 1.7 bp, somewhat larger than that of the hairpin, likely due to different conformations that these proteins may adapt upon binding to DNA.

Discussion

Taken together, these applications demonstrate that the resonator-nSWAT platform can perform as a precision optical trapping instrument. Our Si$_3$N$_4$ resonator-nSWAT platform provides significant trapping force enhancement, generating forces on the order of 20 pN, while allowing for base-pair resolution measurements of multiple molecules at the same time. This has enabled applications of nSWAT in several significant single-molecule optical trapping assays: measuring DNA elasticity, unzipping DNA molecules, and mapping protein–DNA interactions. Thus, this work presents a substantial advancement of the nanophotonic tweezers for future applications of a broad range of biological studies to be performed with nSWAT.

Methods

Fabrication protocol for Si$_3$N$_4$ resonator-nSWAT device. The Si$_3$N$_4$ resonator-nSWAT utilized deep ultraviolet (DUV) lithography for the waveguide patterning and had an N$_2$ annealing process for minimizing the optical loss after the waveguides were plasma etched with SF$_6$/CH$_2$F$_2$/N$_2$. Ni microheaters were evaporated on top of the waveguides to modulate the phase of standing-wave array traps. Moreover, several key design changes and improvements to the published Si$_3$N$_4$ protocol were made in the development of resonator-nSWAT (Fig. 1; Supplementary Fig. 1).

(1) The addition of the resonator design requires additional microheaters to tune the resonance condition of the resonators.

(2) To accommodate these additional microheater electronics on the chip, a 3D electronic architecture was implemented with aluminum vias to connect the microheaters to the electronics contact pads.

(3) The layout of the waveguides on the resonator-nSWAT was designed to minimize waveguide bending loss. The number of bends was minimized, and the radius of bend curvature was kept at > 80 μm to reduce the bending loss (Fig. 1c).

(4) For each resonator-nSWAT device, two local light intensity samplers are coupled to each bus waveguide, and two local light intensity samplers are coupled to each resonator loop. Each intensity sampler is a waveguide that is...
weakly coupled (1:99) to the main waveguide. At the terminus of each sampler waveguide is a Si3N4 scattering grating. Using these samplers, the local light intensities traveling in both the clockwise and counterclockwise directions in each nSWAT were detected by an IR camera at a frame rate of 30 Hz. This intensity may also be used in an intensity clamp by a control software to feedback on the applied voltages to the resonator microheaters.

5. To define the perimeter of a flow channel, each chip was patterned with a 15-μm tall and 200-μm wide SU8 barrier wall that encloses the trapping regions of 8 devices. To regulate the flow of UV glue while sealing the flow channel, one 15-μm tall and 100-μm wide SU8 boundary and two 7-μm tall and 100-μm wide SU8 boundaries are patterned outside the periphery of the flow channel, with 100-μm separations in between (Supplementary Fig. 1).

Detailed information of the device parameters and design calculations are presented in Supplementary Methods 1-4.

Flow cell assembly. In contrast to the coverslip-paraffin-chip flow cell assembly for previous nSWAT designs[32], the flow cell assembly protocol for the resonator-nSWAT devices has been significantly improved for better flow cell quality, robustness, and reproducibility. A shallow and well-defined flow cell is critical to bring the biological sample to the resonator-nSWAT devices while also allowing for a high NA, water immersion objective (Nikon MRD07602) to image the trapped beads. To form a flow cell sample chamber on top of the fabricated resonator-nSWAT devices, a #1.5 coverglass is glued with a UV curing optical adhesive (Norland). First, a coverglass is cut to the appropriate size to cover the diced resonator-nSWAT chip. Entry and exit ports are drilled into the coverslip with a sandblaster and Tygon tubes are glued onto the top of the coverglass with UV glue (Norland NOA68). Next, the coverglass is placed onto the resonator-nSWAT chip and held in place by applying a vacuum to the Tygon tubes. The SU8 pattern (Supplementary Fig. 1) provides a seal and defines the flow cell boundary. Small drops of moderate viscosity UV curing glue (Norland NOA86 & NOA87) to allow the glue to be pulled up to the SU8 flow cell boundary without being pulled past the boundary and into the flow cell. When the UV glue has reached the SU8 flow cell boundary, it is cured by UV light.

Lipid coating procedure. The waveguide surface is passivated to prevent nonspecific binding of beads and proteins by coating the flow cell chamber surfaces with a lipid bilayer following a protocol similar to that previously described[32] with the following changes. A fresh ampoule of 1,2-dioleoyl-sn-glycero-3-phosphocholine (DOPC) in chloroform (Avanti 850375C) is dried by a steady stream of nitrogen gas. The lipids are re-suspended in a high salt buffer (10 mM Tris-HCl pH 8.0, 100 mM NaCl) to a final concentration of 10 mg/mL. The re-suspended lipids

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**Fig. 5 Mapping protein-DNA interactions on nSWAT.**

**a** Schematic of the DNA unzipping mapper configuration. This experimental configuration is similar to that used in Fig. 4a, except that unzipping was carried out in the presence of a protein that can bind to a specific location on the DNA to be unzipped. **b** Simultaneous mapping of a bound restriction enzyme ZraI on five DNA molecules. For each trace with a bound ZraI, a force rise above the baseline indicates that the unzipping fork encountered a bound ZraI protein and provides a measure of the location of the bound ZraI. The two vertical dashed lines bracket the 6 bp recognition site of ZraI. The bottom panel shows a histogram of the detected location of a bound ZraI from these traces: 856.0 ± 0.6 bp (mean ± SD). Source data for this Figure are provided as a Source data file.

**c** Simultaneous mapping of a bound dCas9 on four DNA molecules. The two vertical dashed lines bracket the 20-bp DNA sequence that the sgRNA recognizes when complexed with dCas9. The bottom panel shows a histogram of the detected location of a bound dCas9 from these traces: 1145.0 ± 1.7 bp (mean ± SD). Source data for this Figure are provided as a Source data file.
are then sonicated for an hour to produce small vesicles. The lipids are diluted to 5 mg/mL by the addition of the high salt buffer. The lipid solution is then introduced to the flow cell and incubated overnight for thorough coverage of the waveguides. The high salt buffer subsequently flushes out the lipid solution to remove excess lipid inside the flow chamber. After that, a low salt buffer (15 mM Tris-HCl pH 7.8, 0.2 mg/mL BSA, 0.5 mM EDTA, 0.77 mM NaCl) is then flowed in to allow lipid annealing for 3 h before subsequent trapping experiments.

Preparation of DNA

Stretching template. The 13,709 bp dsDNA stretching template is made via PCR from λ DNA using LongAmp DNA polymerase, a 5’-biotin-labeled forward primer 5’-GCT GAT GTC TGA ACC CGC AGT TGC, and a 5’-digoxigenin-labeled reverse primer 5’-GTC TATT TGG CTT GCC TTG TTT GAG TCT GCT ACT AC for arm 1; upper2: 5’-GTC TATT TGG CTT GCC TTG TTT GAG TCT GCT ACT AC for arm 1; lower2: 5’-GTC TATT TGG CTT GCC TTG TTT GAG TCT GCT ACT AC for arm 2). Each ligated arm is gel purified using Zymo Large Fragment DNA Recovery Kit (D4045).

Data acquisition and bead tracking. The nSWAT data collection software is implemented in LabVIEW 2017. Trapped beads are imaged by a high NA water objective and are also tracked to the cell flow. A cross-correlation-based method to maintain a near-resonance condition. To this high laser power, we implemented a dynamic resonator microheater tuning method to increase the laser power. The resonator microheaters were used to control the on and off states of each nSWAT. Since the resonance peak of the resonance changes with the number of beads trapped on an nSWAT, we implemented an adaptive microheater current optimization method to maximize the trapping light intensity in the resonator. Both trapping and sorting were operated at a low laser power input to the device (∼150 mW).

In this method, the tether orientation was not controlled so that a streptavidin-coated bead in a DNA tether could be trapped on either the top or bottom nSWAT. This orientation degeneracy should not affect the measured force and extension of a DNA molecule.

Stretching and unzipping of DNA. DNA stretching and unzipping experiments were operated at a high laser power input to the device (∼1.5 W). Such a high laser power induces a strong thermo-optic effect within the resonator and shifts its resonance peak of the resonance curve changes with the number of beads trapped on each nSWAT. For each bead, the resonant nSWAT measurement, this concentrated ZraI was added to the unzipping DNA stock at 1:200 dilution, and this mixture was incubated at room temperature for 20 °C for future use. Before an nSWAT measurement, this concentrated ZraI was added to the unzipping DNA stock at 1:200 dilution, and this mixture was incubated at room temperature for 30 min.

dCas9. The sgRNA target sequence (GCGCGUAUCAUCCCUUACCG) was cloned into a PUC19 vector containing the sgRNA scaffold followed by an HDV ribozyme. A PCR product containing the sgRNA 17 transcription template was created from this plasmid and was used for in vitro transcription of the sgRNA using T7 RNA polymerase (NEB M0251S). The sgRNA was then purified using denaturing PAGE and stored in TE pH 8.0 (Invitrogen) at −80 °C until use. To test the unzipping sequence, we mixed the sgRNA and dCas9 (NEB M0652S) at 1 nM in the unzipping buffer (50 mM Tris-HCl pH 7.8, 1 mM MgCl2, 0.1 mg/mL BSA, 100 mM NaCl) and then added to the unzipping template at 100 pm. This mixture was incubated at 37 °C for 30 min. After incubation, the DNA template with dCas9 bound was then used to form DNA-tethers following protocols described above.

N shooting and sorting procedure. Trapping and sorting of DNA tethers on an nSWAT were based on the previous method of alternating the fluid flow direction and concurrently modulating the trap power on and off with the following modifications. The resonator microheaters were used to control the on and off states of each nSWAT. Since the resonance peak of the resonance changes with the number of beads trapped on an nSWAT, we implemented an adaptive microheater current optimization method to maximize the trapping light intensity in the resonator. Both trapping and sorting were operated at a low laser power input to the device (∼150 mW).

For Fig. 5, a bound protein was detected at location with a force rise of at least 2.5 pN above the naked DNA unzipping baseline.
available from the corresponding author upon reasonable request. Source data are provided with this paper.

Code availability
All relevant code is available upon reasonable request.

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References
1. Killian, J. L., Ye, F. & Wang, M. D. Optical tweezers: A force to be reckoned with. Cell 175, 1445–1448 (2018).
2. Bastamante, C. J., Chemla, Y. R., Liu, S. & Wang, M. D. Optical tweezers in single-molecule biophysics. Nat. Rev. Methods Prim. 1, 25 (2021).
3. Svoboda, K., Schmidt, C. F., Schnapp, B. J. & Block, S. M. Direct observation of kinesin stepping by optical trapping interferometry. Nature 365, 721–727 (1993).
4. Wang, M. D. et al. Force and velocity measured for single molecules of RNA polymerase. Science 282, 902–907 (1998).
5. Ma, J., Bai, L. & Wang, M. D. Transcription under torsion. Science 340, 1580–1583 (2013).
6. Ma, J. et al. Transcription factor regulation of RNA polymerase s torque generation capacity. Proc. Natl Acad. Sci. USA 116, 2583–2588 (2019).
7. Kellermayer, M. S., Smith, S. B., Grazier, H. L. & Bastamante, C. Folding-unfolding transitions in single titin molecules characterized with laser tweezers. Science 276, 1112–1116 (1997).
8. Smith, S. B., Cui, Y. & Bastamante, C. Overstretching B-DNA: the elastic response of individual double-stranded and single-stranded DNA molecules. Science 271, 795–799 (1996).
9. Wang, M. D., Yin, H., Landick, R., Gelles, J. & Block, S. M. Stretching DNA with optical tweezers. Biophys. J. 72, 1335–1346 (1997).
10. Le, T. T. et al. Synergistic coordination of chromatin torsional mechanics and topoisomerase activity. Cell 179, 619–631 e615 (2019).
11. Gui, X., Hong, Y., Ye, F., Inman, J. T. & Wang, M. D. Torsional stiffness of extended and pleonastic DNA. Phys. Rev. Lett. 127, 028101 (2021).
12. Liphardt, J., Onoa, B., Smith, S. B. & Bustamante, C. Reversible unfolding transitions in single titin molecules characterized with laser tweezers. Science 276, 1112–1116 (1997).
13. Smith, S. B., Cui, Y. & Bastamante, C. Overstretching B-DNA: the elastic response of individual double-stranded and single-stranded DNA molecules. Science 271, 795–799 (1996).
14. Wang, M. D., Yin, H., Landick, R., Gelles, J. & Block, S. M. Stretching DNA with optical tweezers. Biophys. J. 72, 1335–1346 (1997).
15. Le, T. T. et al. Synergistic coordination of chromatin torsional mechanics and topoisomerase activity. Cell 179, 619–631 e615 (2019).
16. Gui, X., Hong, Y., Ye, F., Inman, J. T. & Wang, M. D. Torsional stiffness of extended and pleonastic DNA. Phys. Rev. Lett. 127, 028101 (2021).
17. Liphardt, J., Onoa, B., Smith, S. B. & Bustamante, C. Reversible unfolding of single RNA molecules by mechanical force. Science 292, 733–737 (2001).
18. Brower-Toland, B. D. et al. Mechanical disruption of individual nucleosomes reveals a reversible multistage release of DNA. Proc. Natl Acad. Sci. USA 99, 1960–1965 (2002).
19. Koch, S. J. & Wang, M. D. Dynamic force spectroscopy of protein-DNA interactions by unzipping DNA. Phys. Rev. Lett. 91, 028103 (2003).
20. Brennan, L. D., Forties, R. A., Patel, S. S. & Wang, M. D. DNA looping mediates nucleosome transfer. Nat. Commun. 7, 13337 (2016).
21. Baker, J. E., Badman, R. P. & Wang, M. D. Nanophotonic trapping: precise manipulation and measurement of biomolecular arrays. Wiley Interdiscip. Rev. Nanomed. Nanobiotechnol. 10, e1477 (2018).
22. Badman, R. P., Ye, F. & Wang, M. D. Towards biological applications of nanophotonic tweezers. Curr. Opin. Chem. Biol. 53, 158–166 (2019).
23. Koch, S. J., Shudrovskiy, A., Jantzen, B. C. & Wang, M. D. Probing protein-DNA interactions by unzipping a single DNA double helix. Biophys. J. 83, 1098–1105 (2002).
24. Shudrovsky, A., Smith, C. L., Liu, J. T., Peterson, C. L. & Wang, M. D. Probing SW1/SNF remodeling of the nucleosome by unzipping single DNA molecules. Nat. Struct. Mol. Biol. 13, 549–554 (2006).
25. Hall, M. A. et al. High-resolution dynamic mapping of histone-DNA interactions in a nucleosome. Nat. Struct. Mol. Biol. 16, 124–129 (2009).
26. Le, T. T. et al. Mfd dynamically regulates transcription via a release and catch-up mechanism. Cell 172, 344–357 e315 (2018).
27. Grujic, K. & Heljes, O. G. Dielectric microsphere manipulation and chain assembly by counter-propagating waves in a channel waveguide. Opt. Express 15, 6470–6477 (2007).
28. Yang, A. H. et al. Optical manipulation of nanoparticles and biomolecules in sub-wavelength slot waveguides. Nature 457, 71–75 (2009).
29. Kühn, S. & Onoa, B. Loss-based optical trap for on-chip particle analysis. Lab Chip 9, 2212–2216 (2009).
30. Juan, M. L., Gordon, R., Pang, Y., Efekharai, F. & Quidian, R. Self-induced back-action optical trapping of dielectric nanoparticles. Nat. Phys. 5, 915–919 (2009).
31. MandaI, S., Serey, X. & Erickson, D. Nanomanipulation using silicon photonic crystal resonators. Nano Lett. 10, 99–104 (2010).
32. Pang, Y. & Gordon, R. Optical trapping of a single protein. Nano Lett. 12, 402–406 (2012).
Author contributions
F.Y. designed and fabricated the devices. F.Y. and J.T.I. designed and implemented the suspended sample stage and upgraded the nanophotonics measurement setup. F.Y. designed and prepared the DNA unzipping template. P.M.H. designed and prepared the sgRNA for dCas9 binding. F.Y. with some help from Y.H. carried out measurements. M.D.W. and F.Y. drafted the manuscript, and all authors edited the manuscript. M.D.W. provided overall guidance on experimental designs and measurements.

Competing interests
The authors declare no competing interests.

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