New 3D Magnetic Tweezers

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Professor Gwo-Bin Vincent Lee, from National Cheng Kung University, Taiwan, and his colleagues have manufactured three-dimensional, micromachined magnetic tweezers to manipulate DNA molecules. Their method was published in the February 7, 2006 issue of *Nanotechnology*.

"This study could provide a provide a powerful tool for exploring the bio-physical properties of biomolecules, bio-polymers and cells," Lee said.

Lee's team made magnetic tweezers from six, hexagonal micro-electromagnets. The scientists wrapped three-dimensional coils, with a width of 80 um, spacing of 100 um, and thickness of 25 um, 30 times around a permalloy core. They chose permalloy because it magnetizes and demagnetizes with low-magnetic field strength.

The type of DNA was likewise important to the study's success. The team used λ-phage DNA, which had two complementary 12-base, single-stranded 5' overhangs. "These overhangs allow phage DNA to easily be derivatized with various functional groups by base-pairing with a complementary sequence," Lee explained. Each base pair of DNA was 0.34nm, without any external force.

The scientists colored the DNA with a green dye, keeping the base pair to dye molecule ratio at 5 to 1, in order to have a high signal-to-noise ratio. "Since a DNA molecule only has a 2nm thickness, we can't observe it with a normal optical microscope," Lee said. The dye allowed the team to view DNA under a fluorescent microscope.

An important element to Lee's study was a microfluidic channel integrated with the magnetic tweezers. This channel had a width of 5mm, height of 60um, and length of 2cm. "We sealed the microfluidic channel with a glass cover slip (100um thick), using double-sided sticky tape (60um thick)."

"The microfluidic channel allowed us to observe a single DNA molecule in real-time," Lee stressed. "We introduced the DNA by pressuring it with a syringe pump into the channel."

Another element to the study was what to use on DNA extremities. "A DNA specific-end anchoring must meet several rigorous requirements, including specific binding, binding strength, localized binding, and complexity level of the procedure," Lee said.

Both thiol group-gold and streptavidin-biotin met all these requirements. "They provide a highly efficient, strong, and specific anchoring," Lee said.

The scientists first bound both ends of DNA to biotin molecules. Next, the team bound one end of DNA to a streptavidin-coated, thiol-modified magnetic bead. "If we label one DNA-end with a thiol group, it must be bound to a gold, magnetic particle. But the gold, magnetic particle was not commercially available," Lee explained.

Lee's team immobilized the modified magnetic bead on a gold pattern using a current of 500mA and a ring trapper. "The ring trapper collects a tethered-magnetic bead DNA molecule to a specific area."

The scientists bound the other end of DNA to a streptavidin, unmodified magnetic bead. "This extremity is manipulated under a magnetic field generated by micro-electromagnets," Lee said.

Lee and his team put the bead on the center of the magnetic tweezers so that the DNA had equal magnetic force throughout its contour length. The team conducted experiments with current from the microelectromagnets. They found that, at 453 pN, DNA is highly elastic.

"DNA is a negatively-charged polyelectrolyte-- at high ionic strength, the majority of negative charges neutralize along the DNA backbone, causing DNA to be more susceptible to elastic elongation," Lee said.

When the scientists applied a current of 400mA to the single DNA molecule, it stretched to a length of 14.74um. In contrast, the team attached two magnetic beads to two DNA molecules, at the same time, and applied 400mA of current. The two DNA molecules stretched to 6.5um.

Why there a difference in stretching between the single and double DNA molecules? “The extension of two parallel DNA molecules is much lower than that of a single DNA molecule, which causes a higher spring constant,” Lee explained. The scientists theorized that the extension may be lower because of curling, different attachment points, or nicking.

The scientists also experimented using a 2.8um magnetic bead and a 1.0um magnetic bead, with the same magnetic field gradient. “Because the bigger magnetic bead has a higher magnetic moment, it can induce a larger magnetic force,” Lee said.

The scientists have proven the efficiency of the magnetic tweezers. “This method doesn’t lose sensitivity and functionality, unlike the use of large-scale and optical tweezers. Moreover, the small size of the magnetic tweezers allows them to be mass-produced at low cost,” Lee said.