

1

Introduction

1.1 History Background and Brief Overview

In the latter part of the 20th century, nanotechnology experienced a remarkable evolution, transitioning from an abstract concept to a tangible science that revolutionized numerous scientific fields. This period was characterized by the advent of several groundbreaking nanotechnological tools and techniques, which allowed researchers to explore and manipulate matter at the atomic and molecular scales. During this period, nanotechnologies such as atomic force microscopy (AFM), magnetic tweezers (MTs), acoustic tweezers (ATs), optical tweezers (OTs), and others emerged successively, each with its own strengths.

The AFM, also known as the scanning force microscopy (SFM), represents a groundbreaking advancement in nanotechnology and materials science (Figure 1.1). As a high-resolution scanning probe microscope, AFM operates at the nanoscale, offering imaging capabilities that surpass the optical diffraction limit by an astonishing factor of 1000. This remarkable achievement has revolutionized our ability to observe and manipulate matter at the atomic level.

The development of AFM can be traced back to its precursor, the scanning tunneling microscopy (STM), which was conceived by Calvin Quate, Gerd Binnig, and Christoph Gerber at the IBM Zurich Research Laboratory in 1986. Building upon the principles of STM, AFM has evolved into one of the most indispensable tools in nanotechnology, serving critical functions in material manipulation, imaging, and precise measurement at the nanoscale.

The operational principle of AFM centers around a sophisticated detection system consisting of a microcantilever and an extremely sharp probe attached to its end. This configuration allows for the precise “sensing” of surface characteristics through the interaction between the probe and the sample surface. The system’s precision is further enhanced by piezoelectric elements, which enable extremely accurate and controlled movements of either the sample or the scanner, typically in the range of nanometers.

One of the most significant advancements in AFM technology is the development of conductive cantilevers and specialized conductive AFM attachments. These innovations allow researchers to measure the current bias of samples, opening up new possibilities in electrical characterization at the nanoscale. More sophisticated AFM

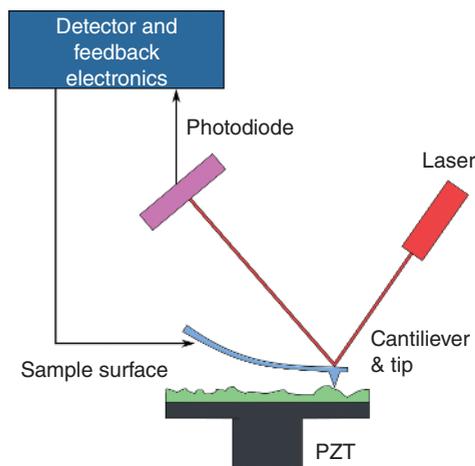


Figure 1.1 An AFM generates images by scanning a small cantilever over the surface of a sample.

Source: OverlordQ's / Wikimedia Commons / CC BY-SA 4.0 / https://en.wikipedia.org/wiki/Atomic_force_microscopy#/media/File:Atomic_force_microscope_block_diagram.svg / last accessed by October 03, 2025.

instruments have pushed these capabilities even further, enabling the measurement of current on the probe itself. This capability allows for the assessment of a sample's conductivity and the observation of electron movement on the sample's underside. However, it is important to note that these advanced measurements present considerable technical challenges, and only a select number of research laboratories worldwide have successfully reported consistent and reliable data from such experiments.

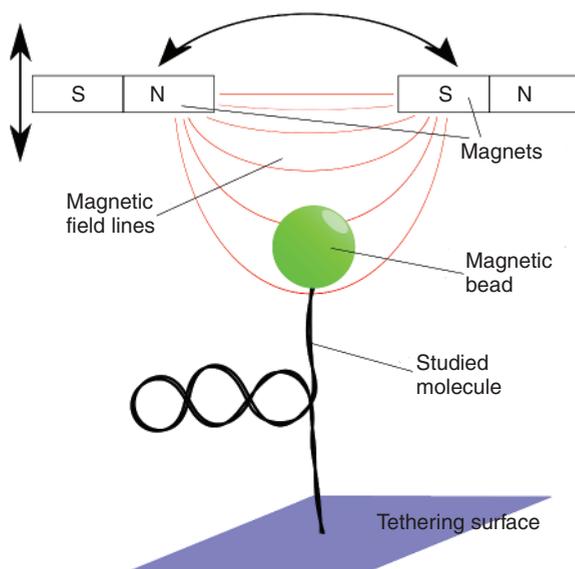
The true power of AFM lies in its ability to sense and amplify the interaction forces between the sharp probe on the cantilever and the atoms of the sample being examined. This capability is what enables AFM to achieve its extraordinary atomic-level resolution. By precisely measuring these atomic-scale interactions, AFM provides researchers with unprecedented insights into the structure and properties of materials at the most fundamental level.

The applications of AFM span across numerous scientific disciplines, including materials science, nanotechnology, biology, and surface chemistry. Its ability to operate in various environments (vacuum, liquid, or air) and modes (contact, noncontact, or tapping) makes it an exceptionally versatile tool for nanoscale research and development. As the AFM technology continues to evolve, it promises to unlock even more possibilities in our understanding and manipulation of matter at the atomic scale, driving innovation in fields ranging from semiconductor technology to biomedical research [1].

Magnetic tweezers (MT) have become a significant and adaptable tool in biophysics and soft matter studies, facilitating the precise control and analysis of single biomolecules or polymers (Figure 1.2). These advanced instruments are engineered to apply specific forces and torques at the molecular level, offering scientists an unparalleled understanding of the mechanical attributes and behaviors of biological macromolecules.

The principal function of MTs is their capacity to assess foundational mechanical properties, such as the tensile strength of molecules or the forces involved in molecular interactions. This ability has rendered them, especially useful in single-molecule experiments, where they are predominantly used to examine the

Figure 1.2 Typical configuration for magnetic tweezers. *Source:* Daniel Förster / Wikimedia Commons / CC BY-SA 4.0 / https://en.wikipedia.org/wiki/Magnetic_tweezers#/media/File:Magnetic_tweezers.svg / last accessed by October 03, 2025.



mechanical characteristics of biological macromolecules like DNA and proteins. In addition to these uses, MTs have proved beneficial across a variety of research domains, including the study of the rheology of soft matter systems and the exploration of force-dependent processes in living cells. The usual range of forces for these experiments extends from piconewtons to nanonewtons, making them perfectly suited for examining the delicate mechanical interactions that underpin biological functions.

One of the key advantages of MTs is their relatively simple and robust architecture, which has contributed to their widespread adoption as a biophysical tool. The experimental setup typically involves tethering the molecule of interest to a magnetic microparticle, which serves as the handle for manipulation. The MT system is equipped with precisely controlled magnets that generate the necessary magnetic fields to manipulate these particles in three-dimensional space. The position and movement of the magnetic particles are then monitored with high precision using advanced video microscopy techniques, allowing for the real-time tracking and analysis of molecular behavior.

The versatility of MTs extends their ability to perform both force spectroscopy and torque measurements, making them particularly valuable for studying the mechanical properties of DNA and proteins. For instance, they have been instrumental in elucidating the mechanisms of DNA supercoiling, protein folding, and the behavior of molecular motors. In soft matter research, MTs have been employed to investigate the viscoelastic properties of complex fluids and the mechanical response of biological membranes.

Recent advancements in the MT technology have further enhanced their capabilities, including improvements in force resolution, the development of multiplexed systems for parallel measurements, and the integration with other single-molecule techniques. These developments have opened up new possibilities for studying complex biological processes and material properties at the molecular level [2].

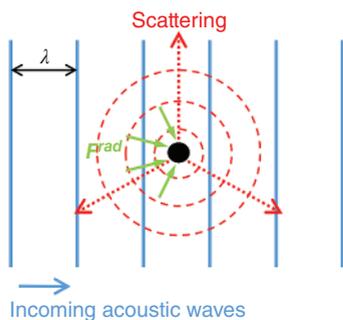


Figure 1.3 Acoustic radiation force on a small particle. *Source:* Rsla1 / Wikimedia Commons / CC BY-SA 4.0 / https://en.wikipedia.org/wiki/Acoustic_tweezers#/media/File:Figure1_zhangming.png / last accessed by October 03, 2025.

Acoustic tweezers (AT) represent a cutting-edge technology that harnesses the power of sound waves to precisely manipulate the position and movement of microscopic and sub-millimeter objects (Figure 1.3). This innovative approach to particle manipulation has opened up new possibilities in various scientific fields, particularly in biomedical research and nanotechnology. At its core, ATs carefully utilize controlled sound waves to create acoustic pressure fields that can influence the position of small objects. While the term “AT” strictly refers to a single-beam configuration, the broader concept encompasses two primary configurations: single-beam and standing-wave setups. The technology operates by manipulating the position of acoustic pressure nodes, which serve as stable points that attract and hold objects within a standing acoustic field. For effective manipulation, the target object must be significantly smaller than the wavelength of the sound used, typically ranging from microscopic particles to individual cells.

One of the most significant advantages of ATs is their noninvasive nature and safety for biological applications. Unlike other manipulation techniques that might involve potentially harmful forces or radiation, acoustic waves have been proven to be gentle and safe for biological objects. This characteristic makes ATs particularly valuable in biomedical research, where maintaining cell viability and integrity is crucial. Recent advancements in the AT technology have expanded its applications across various domains, including flow cytometry, where they have been integrated into systems to enable more precise cell sorting and analysis without the need for physical contact or labels. They have also proven effective in separating different cell types based on their physical properties, offering a label-free alternative to traditional separation methods.

Additionally, ATs can create stable traps for individual cells or groups of cells, allowing for long-term observation and manipulation. Researchers can now manipulate individual cells with unprecedented precision, enabling the study of cell mechanics, response to stimuli, and intercellular interactions. Beyond biological applications, ATs have shown promise in the precise arrangement and manipulation of nanomaterials, opening up possibilities in nanotechnology and materials science. The versatility of ATs stems from their ability to create complex acoustic fields that can be dynamically controlled. This capability allows for the manipulation of multiple objects simultaneously, as well as the creation of intricate patterns or arrangements of particles.

Moreover, the contactless nature of acoustic manipulation minimizes the risk of contamination or damage to delicate samples, making it an attractive option for various applications in cleanroom environments or sterile laboratory settings. As research in this field continues to advance, ATs are expected to play an increasingly important role in areas such as tissue engineering, drug delivery, and the development of advanced materials. The ability of the technology to manipulate objects at different scales, from individual cells to nanomaterials, positions it as a powerful tool in the rapidly evolving fields of biotechnology and nanotechnology [3].

Among them, OTs represent one of the techniques in optical micromanipulation. This field utilizes light to trap and control tiny objects (Figure 1.4). The principles of optical manipulation are based on interactions between light and matter, resulting in the creation of various forces for manipulation, such as optical gradient force, optical scattering force, optical torque, optical pulling force, and optical binding force. Research in optical manipulation technology has significantly advanced our understanding of the microscopic world across various scientific disciplines over the past five decades. The concept of optical micro-manipulation, later known as OTs, was introduced by Arthur Ashkin in the 1980s [4]. OT devices use focused laser beams to exert forces (typically in the range of piconewtons) on microscopic transparent objects. The area where the object is held is referred to as an optical trap, and the technique is known as optical trapping. This technology is utilized for moving cells or virus particles, shaping cells, and cooling atoms. Ashkin first explored the use of light radiation pressure to manipulate physical objects in the late 1960s and began working on the optical manipulation of microparticles [5]. In 1986, Ashkin pioneered the use of a single laser beam for trapping and manipulating

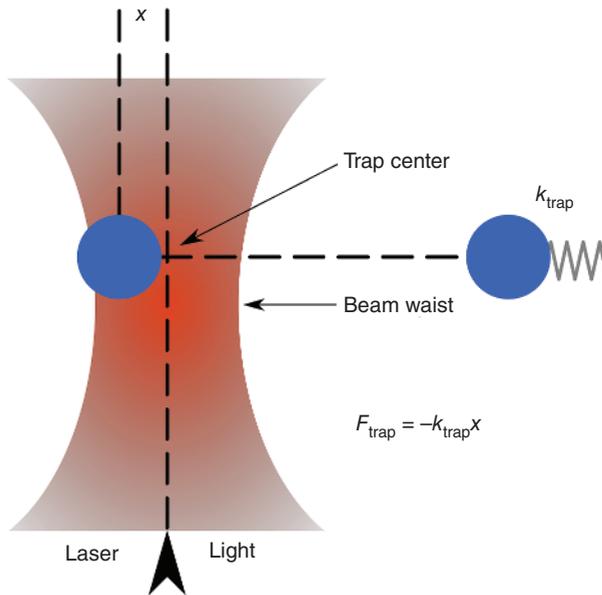


Figure 1.4 Dielectric objects are attracted to the center of the beam. *Source:* Cr4sZz / Wikimedia Commons / CC BY-SA 4.0 / https://en.wikipedia.org/wiki/Optical_tweezers#/media/File:Optical_trap_principle_formula_edit.svg / last accessed by October 03, 2025.

microparticles, leading to the invention of OTs [4]. OTs have been widely applied in physical, chemical, and biological sciences, revolutionizing research in the microscopic realm. In 2018, Arthur Ashkin was awarded the Nobel Prize in Physics for his work on OTs, highlighting the importance and impact of this technology [6]. Despite their utility, OT faces challenges. They are limited to manipulating small, transparent objects (ranging from nanometers to a few tens of microns) due to the small forces they exert. Additionally, OTs require high numerical aperture lenses and complex beam-shaping optics, and they can cause photothermal damage to biological samples. These challenges have restricted the widespread use of OTs to niche and high-end applications in research settings. Motivated by OTs and the need to address these challenges, significant research efforts have been directed toward developing alternative optical micromanipulation technologies.

A novel optical manipulation technique called opto-thermoelectric tweezers (OTETs) has been developed, harnessing thermoelectric forces directed by light. OTETs operate at significantly lower power levels compared to the traditional OTs, as they rely on temperature gradients rather than absolute temperature changes. The adaptability of OTETs is significantly boosted by the universal thermophoretic effect in solutions, which allows for the handling of a diverse range of materials, including polymers, metals, semiconductors, and dielectric nanostructures of varying dimensions and configurations. In addition, OTETs provide the ability to maneuver metal nanoparticles over a wide range of adjustable operational wavelengths, delivering versatile platforms for real-time optical measurements while minimizing the overlap between manipulation and measurement beams. By designing optothermal substrates with nanoscale heat sources, OTETs achieve spatial resolution that surpasses the diffraction limit in nanoparticle manipulation [7].

The theoretical idea of merging optical trapping with plasmonics was first suggested in 1997. As outlined in scholarly journals, a substantial radiation force can be exerted on a dielectric nanoparticle using the intense electromagnetic (EM) field present near the surface of a metal tip. The theoretical framework posits that nanoparticles can be effectively trapped optically when situated close to such a surface. Following this theoretical insight, numerous research teams have carried out experimental studies on plasmonic optical trapping. This method offers several benefits over traditional optical trapping. One primary advantage is that the EM enhancement effect of plasmons reduces the laser light intensity necessary for stable optical trapping compared to standard OT. Moreover, the movement of the trapped particle is constrained within the plasmonic nanospace, which is much smaller than the diffraction limit of light. Additionally, plasmonic optical trapping holds promise for integration with conventional plasmonic applications, including enhancing photochemical reactions and creating highly sensitive chemical and biological sensors [8]. These integrations could pave the way for innovative plasmonic applications.

Xie et al. developed an innovative approach termed “optoacoustic tweezers” for the dynamic aggregation and arrangement of particles and cells within a microfluidic environment. This method, which is both programmable and suitable for biological systems, leverages opto-thermally induced and acoustically activated

surface bubbles. Optoacoustic tweezers allow for the clustering of particles or cells at designated spots inside a microfluidic chamber without the necessity of permanent structures, making it an excellent option for crafting adaptable and complex cellular patterns. The technique has demonstrated superb biocompatibility and can be seamlessly integrated with other microfluidic components [9]. Fiber optic tweezers (FOTs) have emerged as a particularly valuable tool due to their straightforward setup and seamless integration with lab-on-a-chip systems [10]. The inherent flexibility of optical fibers allows for the precise delivery of laser light to targeted areas, making FOTs an ideal choice for applications requiring compact and portable manipulation systems. Building upon the foundation of the traditional OT, scanning optical tweezers (SOT) were developed by incorporating advanced laser scanning technologies [11]. This innovation enables the dynamic trapping and precise movement of microparticles through the use of scanning mirrors or acousto-optical deflectors (AODs) [12]. The AOD-based systems, in particular, offer rapid beam steering capabilities, allowing for the real-time manipulation of multiple particles with high spatial resolution. This technology has proven particularly useful in biological applications where the dynamic control of cellular components is required. The field witnessed another significant leap with the development of holographic optical tweezers (HOTs), which employ spatial light modulators to create complex interference patterns [13]. By applying the holographic technology, HOT can split a single laser beam into multiple independent beams, enabling the simultaneous manipulation of numerous targets in three-dimensional space. This capability has opened new possibilities in colloidal physics and biological research, where multiple interactions need to be controlled simultaneously.

In the early 2000s, researchers introduced OETs, which combine optical stimulation with electric fields through the photoconductive effect of semiconductor materials [14]. This hybrid approach allows for the manipulation of particles using a relatively low optical power, making it particularly suitable for sensitive biological samples. OETs have demonstrated remarkable versatility in handling various types of particles, including cells and nanoparticles.

To overcome the fundamental diffraction limit of light, researchers have explored near-field optical technologies for particle manipulation. These include innovative approaches such as slot waveguides [15], which confine light to subwavelength dimensions by utilizing the strong evanescent field in a narrow air gap between two high-index dielectric materials. This unique configuration allows for intense light confinement, making slot waveguides particularly effective for enhancing optical forces and enabling the precise manipulation of nanoparticles and biomolecules in highly localized regions. Their compatibility with integrated photonic circuits further enhances their utility in lab-on-a-chip systems. Another groundbreaking approach involves photonic crystal resonators [16], which leverage periodic dielectric structures to create highly controlled optical fields. By engineering the bandgap properties of these structures, photonic crystal resonators can trap and manipulate particles with exceptional precision. The ability to tailor the resonance frequencies and field distributions of these devices makes them versatile tools for applications ranging from single-molecule studies to the assembly of photonic devices.

Additionally, photonic nanojets [17] – high-intensity, subwavelength light beams generated by illuminating dielectric microspheres – have emerged as a powerful tool for nanoscale manipulation. These nanojets exhibit a unique combination of high spatial resolution and extended depth of focus, enabling the precise trapping and manipulation of nanoparticles in three-dimensional space. Their simplicity and effectiveness have made them particularly attractive for applications in biosensing, nanofabrication, and single-cell analysis. Furthermore, nanofibers [18] have been employed to achieve highly localized optical fields through their ability to guide light with minimal loss and strong evanescent coupling. By tapering optical fibers to subwavelength diameters, researchers can create intense optical fields at the fiber surface, enabling the trapping and manipulation of nanoparticles, atoms, and even single molecules. Nanofibers are particularly valuable in quantum optics and biological applications, where their flexibility and compatibility with existing optical systems offer significant advantages. These near-field techniques have expanded the capabilities of optical manipulation into the realm of nanotechnology, opening new possibilities for single-molecule studies and nanofabrication applications.

Among these advancements, OETs, pioneered by Ming C. Wu and his team in 2005, have emerged as a promising technology, gaining considerable attention due to their exceptional performance across various domains [14]. This potent opto-electro-fluidic approach uses light-induced electrokinetic forces to manipulate and actuate objects at the nanoscale and microscale. By integrating the strengths of photonics and electronics, OETs offer an accessible solution for versatile and concurrent micromanipulation tasks. In contrast to OT, which requires coherent laser sources, OET systems typically employ a regular LED light source along with a digital micromirror device (DMD). This DMD modulates LED light to create desired light patterns, whether static or dynamic. These patterns are then projected onto the photoconductive substrate of an OET device through an optical microscope. Without light exposure, the photoconductive substrate acts as a high-impedance resistor. However, upon illumination, the photoelectric effect drastically reduces its impedance, effectively turning it into a conductor. As a result, “light-induced virtual electrodes” are established on the substrate, creating differentiated illuminated and dark regions. These virtual electrodes generate an uneven electric field within the liquid medium, interacting with samples to apply electrokinetic forces that manage their positions. Notably, the spatial distribution of this nonuniform electric field can be dynamically adjusted through the projected light patterns, enabling the flexible and swift manipulation of multiple targets at once. Compared to OTs, OETs allow simultaneous manipulation of up to 10,000 traps, demonstrating their capacity for massively parallel operations. Moreover, OETs can exert significantly stronger manipulation forces at a given light intensity. This technology proves suitable where OT might falter, such as in the simultaneous handling of many targets or moving objects across a wide size spectrum, ranging from tens of nanometers to hundreds of micrometers. Additionally, the operation of OETs does not necessitate a coherent light source and functions effectively at low light power densities (below 1 W/cm^2), making it optimal for working with heat-sensitive biological specimens, unlike other heat-assisted optical micromanipulation methods.

1.2 Importance and Applications in Various Fields

OETs stand out among optical micromanipulation techniques for their ability to deliver stronger manipulation forces at comparable light intensities. This unique advantage makes OETs particularly well-suited for the parallel manipulation of multiple objects, including micromachines with large, complex structures or composed of various components. The manipulation force in OETs is primarily governed by the interaction between the electric field and the objects, allowing the precise control over micromachines made from a wide range of materials, including insulators, semiconductors, and conductors. This versatility positions OETs as a powerful tool in advancing research on micromachinery, driving the development of sophisticated, optically controlled micromachines across diverse applications.

OETs have emerged as a versatile and powerful tool for the manipulation and assembly of various nanomaterials in non-biological applications. Employing the principles of light-induced dielectrophoresis (DEP), OETs allow precise control over nanomaterials, offering unique advantages for research and industry. Among the wide array of materials manipulated using OETs are semiconductor and metallic nanowires, carbon nanotubes, and graphene nanoplatelets. These materials play significant roles in the development of advanced electronic and photonic devices due to their exceptional electrical and thermal properties.

Furthermore, conductive nanoparticles and metal ions can also be intricately organized and positioned using OET. The manipulation capabilities of OET effectively cover nanoparticles ranging from just a few tens to several hundreds of nanometers, highlighting its versatility at the nanoscale. This ability facilitates the construction of complex nanoscale architectures and paves the way for innovations in nanotechnology and materials science.

In addition to nanomaterials, OETs are adept at handling microscale objects. These include not only dielectric and metallic micro-particles but also versatile media such as oil and water droplets, gas bubbles, and even specific electronic and photonic components. Such diverse applications demonstrate the proficiency of OETs in managing objects several microns to hundreds of microns in size, making it indispensable for the microsystem technology and lab-on-a-chip applications.

To ensure the effectiveness and efficiency of OET systems, polystyrene spherical microbeads are frequently utilized as standard test samples. The predictable shapes and favorable dielectric properties of these microbeads render them ideal for characterizing OET devices. By leveraging these microbeads, researchers can optimize the system performance and conduct in-depth studies of electrokinetic phenomena under varying experimental conditions. As a result, these microbeads are prevalent not only in device calibration but also in advancing the understanding of the underlying physics that governs OET operations.

In the biomedical arena, OETs have become an invaluable tool, offering novel and effective solutions for manipulating biological materials. Their application spans several cutting-edge areas, such as cell sorting, cell analysis, cell electroporation, and bio-fabrication, each of which plays a critical role in advancing biomedical research and therapeutic development.

A significant advantage of OETs over traditional OTs lies in the reduced reliance of the methodology on high-power light beams. Traditional OT systems often require intense laser light to trap and manipulate cells, which can inadvertently induce heating and cause potential physiological damage to the delicate biological samples. This issue poses a challenge, as it may compromise cell viability and affect the outcome of experiments, particularly in sensitive contexts.

The OET technology, on the other hand, minimizes these risks by employing the principles of light-induced DEP at much lower light intensities. Consequently, it provides a substantially gentler alternative for interacting with biological samples, avoiding the detrimental effects associated with optical heating. This characteristic makes OETs particularly advantageous for manipulating living cells, ensuring their integrity while enabling precise control over their positioning and sorting.

In cell sorting applications, OETs allow for the efficient separation of different cell types based on their unique dielectric properties. This capability is essential for isolating specific cell populations from heterogeneous samples in both research and clinical settings. The technology's suitability for cell analysis also facilitates the study of cellular behaviors and interactions under various conditions, contributing valuable insights into cellular biology and pathology.

Beyond manipulation and analysis, OETs offer significant potential in cell electroporation, where precise electrical fields are applied to temporarily permeabilize cell membranes. This process enables the introduction of foreign molecules, such as drugs or genetic material, into cells, proving instrumental for genetic engineering and drug delivery applications.

Furthermore, in the field of bio-fabrication, the precise control of OETs facilitates the construction and manipulation of cell-based biostructures and micromachines. This capability can lead to the development of complex tissue models and biomimetic devices, which have promising applications in tissue engineering and regenerative medicine.

Overall, OETs demonstrate substantial potential across a wide range of scientific and technological applications. Its versatility and efficacy in manipulating diverse materials highlight its transformative role in both non-biological and biomedical fields. As researchers continue to explore and refine the OET technology, its applications are likely to expand, paving the way for groundbreaking discoveries and innovations in multiple disciplines.

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