# The Revolution in Techniques Used in Observation and Imagery

Fozilova Mohichehra Dilmurod qizi<sup>1</sup>, Bakirov Eldorbek Valijon oʻgʻli<sup>2</sup>, Abdubannobov Moʻydin Iqboljon oʻgʻli<sup>3</sup>

Annotation: The Revolution in Techniques Used in Observation and ImageryThis chapter explores the revolutionary advancements in the techniques used for observation and imaging in the context of nanoscience and technology. It highlights the shift from traditional methods of visualizing and analyzing materials and biological systems to cutting-edge techniques that allow scientists to study matter at the atomic and molecular levels with unprecedented precision. In essence, this chapter highlights how innovations in observation and imaging techniques are enabling scientists to explore the world at the smallest scales, unlocking new opportunities for both fundamental research and practical applications in nanotechnology. The revolution in these techniques has opened doors to deeper understanding and manipulation of materials, biological systems, and even fundamental physical properties at the nanoscale.

**key words:** Observation techniques, Imaging techniques, Nanoscience Nanotechnology, Atomic scale, Electron microscopy, Transmission Electron Microscopy (TEM), Scanning Electron Microscopy (SEM).

#### Introduction

The Revolution in Techniques Used in Observation and Imagery In recent decades, advances in observation and imaging techniques have sparked a revolution in the field of nanoscience and nanotechnology. These innovations have provided scientists with unprecedented access to the atomic and molecular worlds, unlocking new frontiers for both fundamental research and practical applications. At the core of this revolution is the ability to observe, manipulate, and understand materials at the atomic scale, a level of precision once thought impossible.

The traditional methods of visualizing objects—such as optical microscopy—were long limited by the diffraction limit of light. As scientists sought to study smaller and smaller structures, it became evident that more advanced methods were needed. The development of electron microscopy marked a critical turning point. By utilizing electron beams instead of light, techniques like Transmission Electron Microscopy (TEM) and Scanning Electron Microscopy (SEM) have enabled researchers to visualize objects down to the scale of individual atoms and molecules.

TEM offers the ability to observe the internal structure of samples by transmitting electrons through them, while SEM uses scanning electron beams to produce detailed surface images. These tools have become essential in materials science, enabling the study of nanomaterials with remarkable resolution and clarity. Scanning Probe Microscopy (including Atomic Force Microscopy (AFM) and Scanning Tunneling Microscopy (STM)) has further expanded the range of possibilities, allowing scientists to manipulate individual atoms and molecules directly, opening doors to nanofabrication and precise nanomanipulation.

The ability to visualize and study materials at this scale is not only a triumph for scientific exploration but also has profound implications for real-world applications in areas such as electronics, medicine, and energy. From understanding nanoscale phenomena to designing nanostructures with specific properties, these imaging technologies are shaping the future of technology and scientific discovery.

<sup>&</sup>lt;sup>1,2,3</sup> FBTUIT assistant

As we continue to push the boundaries of observation, these revolutionary techniques allow for deeper understanding, new technological innovations, and an ever-expanding knowledge of the nanoscale world, providing new insights into the behavior of materials and biological systems. In this chapter, we explore the significance of these advancements and their transformative impact on nanotechnology research and beyond.

## **Literature Review**

The Revolution in Techniques Used in Observation and Imagery. The evolution of observation and imaging techniques has been one of the key drivers of progress in nanoscience and nanotechnology. In order to explore matter at the atomic and molecular scales, researchers have developed a range of techniques that allow for unprecedented resolution and precision. This section reviews the key literature that has contributed to the development of modern imaging technologies, particularly those that have enabled the study of materials and biological systems at the nanoscale.

- 1. The Limitations of Traditional Observation Techniques.Early efforts to observe microscopic structures relied heavily on optical microscopy, which, due to the diffraction limit of light, could not resolve objects smaller than approximately 200 nm. The Rayleigh criterion, established by Lord Rayleigh in 1879, sets a fundamental limit on the resolution of optical microscopes, meaning that the details of structures at the nanoscale (typically under 100 nm) were inaccessible using visible light. This limitation spurred the search for alternative techniques capable of imaging smaller objects with greater precision.
- 2. Advances in Electron Microscopy.A major breakthrough came with the development of electron microscopy, which employs electron beams instead of light to visualize objects. The de Broglie wavelength of electrons is much smaller than that of visible light, allowing for much higher resolution. The first Transmission Electron Microscope (TEM) was developed in the 1930s by Max Knoll and Ernst Ruska, which allowed for the direct observation of internal structures of specimens at the atomic scale. In the 1960s, Scanning Electron Microscopy (SEM) was introduced, offering improved resolution for surface imaging.

Recent reviews (e.g., Egerton, 2011) highlight that TEM allows for the study of fine details within materials, such as atomic arrangements and crystal defects, while SEM has become essential for analyzing surface morphology and topography of nanomaterials. These techniques have played a crucial role in the understanding of nanomaterials, semiconductor fabrication, and the development of nanostructured materials.

### Methods

The optical microscope in visible light The optical microscope was the first instrument that enabled man to observe objects normally invisible to the naked eye. As the microscope is subject to the laws of optics, its resolution is limited to several tenths of a micron. In order to study samples from living organisms, the samples must be prepared with coloration techniques. A new generation of microscope which uses laser light appeared in the 1980s. It has enabled scientists to create three-dimensional images at different levels of depth of the matter being studied by using focalization and laser beam scanning. This type of microscope is known as a confocal microscope 1 and is particularly adapted for use in the natural world. One very interesting use of these microscopes corresponds to their ability to work with fluorescent markers. The laser beam excites a fluorescent substance which has been added to the sample, for which we know the affinity for certain molecular sites. Thanks to these markers we can, for example, selectively view certain reactions. The fluorescent signals are detected by electronic. sensors and these signals are then amplified. The image is then processed by computer.

X-ray machines X-rays are photons with a wavelength that is much shorter than the wavelength of ultraviolet light. X-rays are produced from an accelerated shock of electrons against a metallic target. One of the first applications of machines using X-rays was in the macroscopic domain. The X-rays benefit from the fact that this radiation has a strong penetrating power in materials with the rate of absorption depending on the density of the material. Radiation transmitted through a body coated with

a phosphorescent or photosensitive substance is commonly known as radio waves. A sophisticated version of this type of machine is the X-ray scanner. The transmitter turns around the object at the same time as the receptor does, measuring the intensity of the X-rays transmitted. The data is processed by a computer which reconstructs crosssections of the object, in other words 3-D imagery. The resolution is determined by the quality of the X-ray beam used. This type of machine is used in many applications, especially in medical imagery. Another type of machine, which uses the interactive properties of X-rays with crystalline structures, is used in X-ray spectroscopy. These machines enable scientists to investigate objects in the nanoworld. Their operation rests on the following principle: a crystal is made up of identical patterns of atoms following a particular lattice whose chain is the same size as the wavelength of the X-ray. The X-rays are realigned by selective reflection in predetermined directions and then form diffraction figures. The information contained in the diffraction figures clearly deals with the structure of the lattice and, more specifically, the rather complex three-dimensional structures of atomic patterns. This analysis is possible, firstly, due to the quality of today's machines and, secondly, because of the sophisticated calculation techniques used.

Observing with electrons Electron microscopy uses the wave properties of electrons. However, as particles they need a vacuum in order to travel. Microscopes are in the form of a metal vacuum enclosure in which the following can be found: – The electron gun, such as in cathode ray tubes used in television sets. – The different elements of electronic optics, such as electromagnetic lenses (equivalent to traditional optic lenses) which control the trajectories of the electrons as well as the support of the object to be studied. There are two types of electron microscope.

The scanning electron microscope (SEM) The surface of the sample under study is scanned with an electron beam. The size of the scanned surface depends on the level of enlargement desired. The interaction between the electrons and the sample gives rise to different signals (the emission of electrons and photons) which when gathered and analyzed bring together the image of the surface of the observed sample without using any mathematical process, contrary to the process of the TEM. The resolution of this type of instrument, limited by the machine's technology, enables scientists to view objects at an atomic scale (1/10 of a nanometer). A significant restriction of this microscope, as is the case for the TEM, is that it needs a vacuum. The samples need to be prepared in a specific manner, in other words they need to be plated, cooled, and cut into thin sections, all of which are clearly impossible when observing living organisms. A new generation of SEM has overcome this restriction; they are known as environmental scanning electron microscopes. These SEMs enable scientists to observe objects in their natural state. The difference between the environmental scanning electron microscope and the conventional ones, which need a high vacuum on all levels of the columns that make up the microscope, is that the sample remains at a determined pressure thanks to a differential diaphragm pump system used in the observation room.

Touching the atoms The atomic microscope is mainly used in research laboratories. It works on a simple principle, but with very sophisticated technology. Scientists create an image of vertical displacement from a point on the surface of a sample. This point is made up of some atoms (eg thinned down tungsten microcrystal atoms) and the precision of displacement is to the nearest 1/10 of a nanometer. In the first version invented by IBM researchers3 in 1981, the control signal is the current, albeit extremely weak, existing between the point of the microscope and the surface of the sample without any contact between the microscope and the sample. However, they are at a distance where the electrons can pass through by using the tunnel effect. In this case, we are referring to the scanningtunneling microscope. When there is contact between the point of the microscope and the surface of the sample, the microscope is called an atomic force microscope. This is the nano equivalent of our old gramophones. This type of microscope enables scientists to analyze surfaces with insulating properties, which is impossible with the scanningtunneling microscope. An optical version has existed for a short time now, and it is based on the presence of an optical wave that does not move. This evanescent wave is present on the illuminated surface of a sample which can only be detected on a nanoscopic level. With these instruments, scientists can see the atoms of a surface, but they can also use these instruments to move the atoms, form

### **Results and Discussion**

The Revolution in Techniques Used in Observation and Imagery. The advent of new and advanced observation and imaging techniques has drastically altered the way scientists study materials, biological systems, and nanostructures. These technological innovations have provided critical insights into the nanoscale world, opening new possibilities for research and application in fields such as nanotechnology, materials science, medicine, and biology. The results of these advancements have not only expanded our understanding of fundamental physical and biological processes but have also led to the development of practical, real-world applications. Below, we discuss the key results from the evolution of these techniques and their broader implications.

1. Enhanced Resolution and Precision in Imaging

One of the most significant results of recent advancements in observation techniques is the dramatic increase in resolution. Traditional optical microscopy, limited by the diffraction of light, could not visualize objects smaller than 200 nm. However, the development of electron microscopy (EM), especially Transmission Electron Microscopy (TEM) and Scanning Electron Microscopy (SEM), has enabled scientists to image objects down to the atomic scale.

- Transmission Electron Microscopy (TEM) has become a cornerstone in materials science for its ability to provide high-resolution images of the internal structure of materials. The spatial resolution of modern TEM has improved to the sub-nanometer range, allowing for the study of atomic arrangements, crystal defects, and even chemical bonding within materials.
- Scanning Electron Microscopy (SEM), while primarily used for surface imaging, has also achieved nanometer-scale resolution, making it indispensable in characterizing the surface topography of nanomaterials such as carbon nanotubes, nanowires, and graphene. According to Li et al. (2019), SEM has become one of the most widely used techniques for analyzing nanostructures, particularly for materials with complex surface morphologies.

The ability to examine materials and biological samples with this unprecedented resolution has made it possible to visualize phenomena previously hidden from view, such as the arrangement of atoms in nanomaterials and the structural details of proteins and viruses.

2. Atomic-Scale Imaging and Manipulation

The development of Scanning Probe Microscopy (SPM), including Atomic Force Microscopy (AFM) and Scanning Tunneling Microscopy (STM), has provided even more profound insights into the nanoscale world by allowing scientists to directly interact with individual atoms and molecules.

- Atomic Force Microscopy (AFM) is particularly useful for imaging soft, delicate, or biological samples that cannot withstand the high-energy electron beams used in traditional electron microscopy. AFM provides not only high-resolution images of surface topography but also mechanical properties such as stiffness, elasticity, and adhesion at the nanoscale. According to Kisielowski et al. (2020), AFM has become a powerful tool for studying the properties of biomolecules, polymers, and thin-film materials.
- Scanning Tunneling Microscopy (STM), on the other hand, operates by measuring the tunneling current between a sharp metal tip and the surface of a material, allowing researchers to directly image individual atoms and study their electronic properties. STM has been instrumental in nanofabrication, enabling the manipulation and assembly of single molecules and atoms for the construction of complex nanostructures. Binnig and Rohrer (1981), in their groundbreaking work on STM, demonstrated the first imaging of individual atoms on a surface, a significant leap forward in nanotechnology.

These atomic-level techniques have revolutionized the way we understand and manipulate matter at the nanoscale. The ability to visualize and even move atoms has implications for the development of advanced materials, quantum computing, and molecular electronics.

3. Multimodal and Hybrid Imaging Techniques

Another result of the technological revolution is the integration of multiple imaging techniques into multimodal imaging systems. By combining different methods such as electron microscopy, atomic force microscopy, and X-ray diffraction, scientists can obtain more comprehensive information about the material being studied.

- Multimodal imaging enables researchers to correlate different types of data, such as structural and mechanical properties, in a single experiment. For example, a scanning electron microscope (SEM) might be used to obtain surface morphology, while atomic force microscopy (AFM) can provide insights into the mechanical properties, and X-ray diffraction (XRD) can be used to understand crystallography. This integration allows for a more holistic view of a material's characteristics.
- ➤ In the realm of biological imaging, combining fluorescence microscopy with electron microscopy has opened new doors to the study of protein-protein interactions and cellular processes. Techniques such as super-resolution microscopy (STORM and PALM) have enabled the imaging of cellular structures at nanometer resolution, facilitating a deeper understanding of biological phenomena like membrane dynamics, gene expression, and intracellular trafficking.

This integration of different techniques allows for a deeper understanding of complex materials and biological systems, making it possible to study multiple properties of the same sample simultaneously.

The Continuing Revolution in Nanoscale Observation. The revolution in imaging and observation techniques has significantly transformed our ability to explore and manipulate the nanoscale world. From electron microscopy and scanning probe microscopy to multimodal and hybrid imaging systems, these innovations have enabled scientists to study materials, biological systems, and molecular interactions with unprecedented precision. As these techniques continue to evolve, they promise to unlock even greater insights, driving forward advancements in nanotechnology, medicine, and materials science. However, challenges such as sample preparation, imaging speed, and the need for real-time observation remain, requiring continued innovation and collaboration across disciplines.

The integration of these advanced techniques into nanoscience is not only expanding our fundamental understanding of the natural world but is also paving the way for the development of new technologies that could revolutionize various industries, from medicine to electronics and energy. The next wave of innovations will likely involve the fusion of imaging with other computational techniques, such as machine learning and artificial intelligence, to further accelerate discoveries and applications at the nanoscale.

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