Nanotechnology: Nanomedicine

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ABBREVIATIONS: HIV= Human Immunodeficiency Virus; MIT= Massachusetts Institute of Technology; MRI = magnetic resonance imaging; COPD= chronic obstructive pulmonary disease; FDA= Federal Drug Administration, surface enhanced Raman scattering (SERS), Quantum dots (QDots®).

INDEX TERMS: Jain quantum dots, vibrational spectroscopy, nanofluidic devices, magnetic nanoparticles, surface-enhanced Raman scattering, quantum dots (QDots®)

LEARNING OBJECTIVES:

1. Discuss applications of nanotechnology in the clinical laboratory.
2. Describe how infectious diseases may be rapidly detected.
3. List nanotechnology methods to detect cancer.
4. Give an example of an organism that can be detected using nanotechnology.
5. Discuss recent developments in applications of nanotechnology to treatment options.

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Nanomedicine

Nanomedicine is a branch of nanotechnology that is used for *in vivo* and *in vitro* diagnostic testing and treatment of diseases (Figure 1). Nanotechnology has the potential to repair or even replace tissues and structural units such as the human nephron.[1] The proposed 2010 budget for the National Nanotechnology Initiative is $1.6 billion. From this fund, the proposed allotment for the National Institute for Health (NIH) is $326 million.[2] The NIH has established eight Nanomedicine Development Centers across the country to research different applications of nanomedicine.[3] For example, the center at Baylor College of Medicine in Houston, is the Center for Protein Folding Machinery, while the Center of Cell Control (CCC) is at the University of California, Los Angeles.[3] These eight centers work in partnership to understand nanoscale properties of cells and tissues over a wide range of diseases and conditions, while at the same time each center focuses on a specific type of disease. The Center for Protein Folding Machinery, focuses on protein misfolding diseases such as Huntington's disease while the CCC is concentrating its efforts on developing drug cocktails for HIV.[4,5]

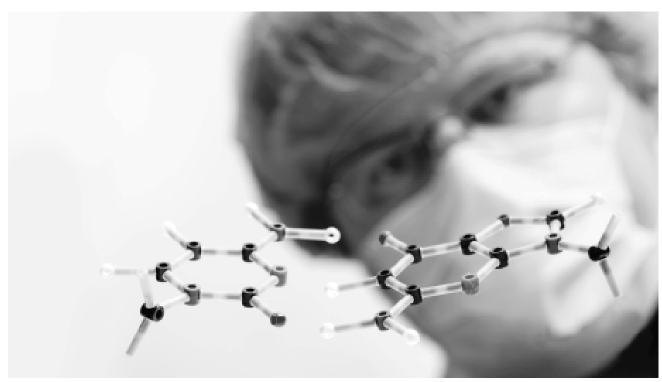

Figure 1. Nanomedicine

In recent months, Dr. Chad Mirkin of the *Northwestern University Institute for Nanotechnology* was awarded the $500,000 Lemelson-MIT Prize as the world's most-cited nanomedicine researcher. The major objective of much of his work has been that of very early detection of disease-related molecules. His Verigene ID System® (Nanosphere, Inc.) can simultaneously test patients for a multitude of markers in less than an hour.[6] Mirkin's bar-code test for Alzheimer's disease can detect minute traces of protein associated with the disease.[6] In association with Aurasense®, he is working with nanostructures made of gold (nanotherapy agents) that are reported to be nontoxic and highly active in promoting gene regulation in cardiology and oncology patients.

Another technology (Nscriptor™), licensed by Nano-Iink®, is focused upon the interaction between individual cells and viruses at the cell membrane-docking interface. This will provide important information about how viruses infect cells and about the physicochemical differences between normal and malignantly transformed cells.[6]

Drug Delivery

Dr. Edwin Donath of the *Institute of Medical Physics and Biophysics* (Leipzig, Germany) takes a different approach. Instead of "building nanodevices from the ground up", he proposes that viruses be rendered harmless and engineered as artificial nanoparticles. His objective is that of modifying and "arranging chemical functionalities with nanoscale precision" to accomplish drug delivery and genetic engineering.[7] In the event of the latter, the genome of the virus would be able to deliver essential genes to correct deficiencies and harmlessly continue replication.[7]

Nanodrug delivery may benefit patients with cancer as well as patients with benign diseases (Figure 2). Researchers in India conducted a study on healthy volunteers to determine if inhaled nano-sized particles of salbutamol, which is commonly used to treat asthma and chronic obstructive pulmonary disorders (COPD), would penetrate the lungs better than the currently inhaled micro-sized particles.[8] They found that fewer nano-sized particles were absorbed by the gastrointestinal tract and that there is improved lung penetration over traditionally used micro-sized particles. More studies are in progress, but this work shows promise for improved treatment for millions of people who suffer from asthma or COPD.

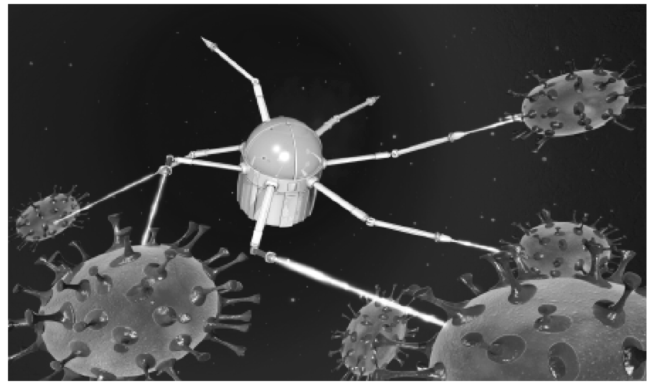

Figure 2. Nanodrug delivery.

Clinical trials investigating the use of nanotechnology methods to deliver less toxic chemotherapeutic drugs are currently in progress. Presently, chemotherapeutic drugs kill not only cancer cells, but also damage healthy cells and can be very toxic. Nanodrug delivery "can increase the effect of the drug while reducing side effects"[1] by actively targeting the tumor cells.

In vivo Applications

For those interested in hematology, a new synthetic peptide developed at MIT in conjunction with a nanocarrier that is applied to wounds stops bleeding very quickly. It has a lengthy shelf-life, is non-toxic and is easily degraded by the body.[9]

University of Michigan researchers (Dr. Nicholas Kotov, *et al.)* have "created red and white blood cells from artificial bone marrow." This has the potential for providing a continuous blood supply for transfusion. The three-dimensional biomedical scaffold supporting the "marrow" was not implanted into the human body. The system reportedly functions *in vitro* and is termed the "first artificial bone marrow".[10]

Non-invasive lung cancer detection using carbon nanotubes is being developed by Israeli researchers (Dr. Hossam Haick, *et al., Israeli Institute of Technology*). A variety of organic materials were coated onto nanotube array devices and, according to the report, markers specific for lung cancer were detected in the breath of patients.[11]

Cancer that has metastasized to the lymph nodes has been detected using magnetic nanoparticles with magnetic resonance imaging (MRI).[1,12] In a study published in the *New England Journal of Medicine* in 2003, eighty patients with prostate cancer were injected with superparamagnetic nanoparticles with an affinity for lymphocytes. An MRI was performed before and after the injection and the results were correlated with histopathological reports. The findings showed that the MRI with nanoparticles had a higher sensitivity than MRI used alone.[12]

The combined use of nanotubes and stem cells has been reported by researchers at the *University of California, San Diego* to accelerate bone growth. Broken knees and legs often require titanium implants and bed rest for about three months. Patient's own bone marrow mesenchymal stem cells have been implanted on thin titanium nanotubes, which facilitate rapid healing that may eventually be reduced to one month.[13]

Clinical Laboratory Diagnostic Assays

Nanotechnology integrates techniques, from different disciplines, that are already familiar to most clinical laboratorians. Assays using nanoparticles have recently moved into the clinical diagnostics laboratory. The most common nanoparticles used for diagnostic assays are gold nanoparticles, quantum dots (QDs) and magnetic nanoparticles.[14]

A nanotechnology biosensor for heat-shock proteins produced by *Listeria monocytogenes,* a bacterium notorious for causing food poisoning, has been developed by Dr. Arun Bhunia, *et al.* Although there are rapid, effective tests for identifying several strains of *Listeria,* all of them are dependent upon reaction with the appropriate antibody. The nanotechnology-based biosensor has a "microbe capture" efficiency as high as 83 times that of the antibody-based test.[15] Imagine a microbiology laboratory "freed" from overnight culture and tedious read-outs, and patients enjoying virtually instant and more accurate diagnosis.

In 2007, the FDA approved a genetic test to detect patients with increased risk for serious bleeding when taking Warfarin.[16] Patients who have the genotype that makes them more sensitive to Warfarin would obviously be prescribed an alternate medication. This test is made by Nanosphere to be performed on its Verigene® System that uses gold nanoparticle technology to detect protein and nucleic acid targets. Assays such as these that fall under the category of personalized medicine, will increase dramatically over the next few years, because they not only improve patient outcomes, possibly even saving lives, but they are ultimately more cost effective. In October 2009, Nanosphere received FDA approval for the Verigene® Respiratory Virus Nucleic Acid Test$_{SP}$ (VRNAT) that detects influenza A, influenza B and RSV to be used on the Verigene® system using proprietary gold nanoparticle probes. This method is a combination of molecular techniques and nanotechnology.[17] Nanosphere has recently submitted a 510(k) application to the FDA for a cardiac troponin 1 test and has several more tests in development.[18]

Surface-enhanced Raman scattering (SERS) is a method that combines microscopy and spectroscopy. SERS was initially developed in the 1970's.[19] It is a highly sensitive technique for detecting low levels of viruses and bacteria by comparing the spectra of a target molecule to the known Raman spectrum of a particular organism.[19] SERS has not been widely used due partially to the difficulty of making metal-coated substrates that have a long shelf life and produce consistent results.[19] With new nanofabrication techniques which employ silver nanorod substrates, Tripp, Dluhy and Zhao (2008) have been able to not only differentiate between various strains of respiratory syncytial virus (RSV), but have also been able to detect as few as 1–10 plaque-forming units (PFU) of the virus.[19]

Quantum dots are tiny crystals about a nanometer in size that contain a hundred to thousands of atoms.[20] Invitrogen™ is currently producing custom made Qdot® nanocrystals for specific laboratory applications such as flow cytometry. These nanocrystals have the usual properties of fluorescence (absorbing light of one wavelength and emitting light of a longer wavelength) but they are coated with a semiconductor material to improve their optical properties.[22] The Qdot® nanocrystals range in color from red, orange, yellow, green, and blue. The size of the nanocrystal determines the color it emits. The Qdot® has much longer photostability than typical fluorescent dyes.[22]

The development of nanoliter droplet real-time polymerase reaction (PCR) with laser-assisted heating has been reported by Dr. Hanyoup Kim, *et al.* of SRI International. This assay performance was completed in 40 cycles (370 sec.), and is amenable to series of assays "loaded" simultaneously. The authors reported that the assay "was quantitative and its amplification efficiency was comparable to that of a commercial instrument."[22] This methodology has the potential for examining minuscule specimens in a vastly shortened "run" time.

Another group of researchers, Son and colleagues, have developed a nanoparticle-DNA based assay for detection of polycystic kidney disease (PKD)[1,23]. PKD is an autosomal-dominant disorder that is caused by a single nucleotide polymorphism (SNP)[23]. The nanotechnology protocol for detecting the single base pair change does not require a PCR step. Hopefully this protocol will be further developed to be used with other SNPs.

Conclusion

"In the next wave of clinical laboratory nanotechnology, assays will be developed that completely replace current methods of laboratory diagnostics".[24] Most Medical Laboratory Scientists (MLS) enjoy the challenge of learning new technologies. Just as molecular diagnostic techniques brought a new wave of excitement to the clinical laboratory, nanotechnology will do the same. These new techniques emphasize the need for more highly trained medical technologists, cytotechnologists and histotechnologists.

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