Digest Journal of Nanomaterials and Biostructures Vol. 3, No.3, September 2008, p. 135 - 140

NANOMATERIALS AS EMERGING TOOL IN CANCER DIAGNOSIS AND TREATMENT

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Cancer is the widespread disease all over the world and it is leading cause of death. The war against cancer using nanotechnology revolutionized the world with its unique approaches to early detection diagnosis and therapy that could not be anticipated with conventional technology. New tools at nano-meter size which is much smaller than a human cell will enable researchers and clinicians to detect cancer earlier and treat it with much greater accuracy and lesser side effects. Data for this review article were identified through MEDLINE and PubMed searches for published reports using the terms cancer, nanotechnology, genetics, epidemiology, predictive markers, medicine and therapy. Only articles published in English were included. Several recent critical advances and high potentive techniques with use of nano-materials for identifying, early diagnosis, and therapy related to cancer, have dramatically accelerated the pace of research which can help in preventing cancer and curing it.

(Received June 20, 2008; accepted June 24, 2008)

Keywords: Cancer therapeutics, Nanoparticles, Quantum dots, Biomarkers, Nanomedicine.

1. Introduction

Cancer is major problem all over the world, and approximately 10.9 million people worldwide are diagnosed with cancer. Approximately 45% of these cases have been identifided in Asia, because of the high density of population [1]. Here, each year 6.7 million people die from this disease that is around 12% of deaths worldwide [1, 2]. We have facing such a serious situation; there has been no substantial progress in the past 50 years against cancer. In this new era of world a new technology is emerging that have a diagnostic and therapeutic potential against this disease. Particularly in this review, we focus on the nanoscience and their role in cancer diagnosis and therapy. It is expected that nanotechnology will be developed at several levels: materials, devices and systems. At present, the nanomaterials level is the most advanced in scientific knowledge as well as in commercial applications. A decade ago, nanoparticles were studied because of their size-dependent, physical and chemical properties [3]. Traditionally, the most common cancer treatments were limited to chemotherapy, radiation, and surgery. Limitations in cancer treatment are a result of current challenges seen in established cancer therapies, including lack of early disease detection, nonspecific systemic distribution, inadequate drug concentrations reaching the tumor, and inability to monitor therapeutic responses. Poor drug delivery and residence at the target site leads to significant complications, such as multi-drug resistance [4].

Nanotechnology has the potential to offer solutions to these current obstacles in cancer therapies, because of its unique size (1-100 nm) and large surface to volume ratios [5]. Nanotechnologies may have properties of self-assembly, stability, specificity, drug encapsulation and biocompatibility as a result of their material composition [6]. Nano-particles are being actively developed for tumour imaging in vivo, bio-molecular profiling of cancer biomarkers, and targeted

drug delivery. These nanotechnology based techniques can be applied widely in the management of different malignant diseases. Several nano-technological approaches have been used to improve delivery of chemotherapeutic agents to cancer cells with the goal of minimizing toxic effects on healthy tissues while maintaining anti-tumor efficacy [7].

2. Cancer disease

Sixteen million new cases are estimated every year by 2020. Mortality rate of cancer in the world are predicted to accelerate with an estimated 9.0 million death from cancer in 2015 and 11.4 million death in 2030 [8].

Biomarkers are characterized by potent tools for monitoring the course of cancer and estimating the efficacy and safety of novel therapeutic agents, as an important biological indicator of cancer status and progression for the physiological state of the cell at a specific time. They can have tremendous therapeutic impact in clinical oncology, especially if the biomarker is detected before clinical symptoms or enable real-time monitoring of drug response. There is a critical need for expedited development of biomarkers and their use to improve diagnosis and treatment for cancer. Malignant transformation involves alterations in protein expression with subsequent clonal proliferation of the altered cells. These alterations can be monitored at the protein level, both qualitatively and quantitatively. Protein signatures in cancer provide valuable information that may be an aid to more effective diagnosis, prognosis, and response to therapy. The recent progress of proteomics has opened new avenues for cancer-related biomarker discovery. Advances in proteomics are contributing to the understanding of patho-physiology of neoplasia, cancer diagnosis, and anticancer drug discovery. Continued refinement of techniques and methods to determine the abundance and status of proteins holds great promise for the future study of cancer and the development of cancer therapies [8, 9].

3. Current tumor markers

Early diagnosis of cancer is difficult because of the lack of specific symptoms in early disease and the limited understanding of etiology and oncogenesis. For example, blood tumor markers for breast cancer such as cancer antigen (CA) 15-3 are useless for early detection because of low sensitivity [10]. Therefore measurement of carcino embryonic antigen (CEA) and HER-2 in abnormal nipple discharge has been approved for diagnosis of breast cancer in some countries [11]. More than 98% of cervical cancer is related to human papilloma virus (HPV) infection. The identification and functional verification of host proteins associated with HPV E6 and E7 oncoproteins may provide useful information for the understanding of cervical carcinogenesis and the development of cervical cancer-specific markers [12]. For hepatocellular carcinoma (HCC), the common method of screening high risk patients by alpha-fetoprotein (AFP) and ultrasonography has been shown to result in earlier detection and consequently more easily treatable tumors and longer survival. Of the other tumor markers, the newer high sensitive desgamma- carboxy-prothrombin has been found to be useful.

In addition, the AFP fractions L3, P4/5, and the +II band are highly specific for HCC. Among routinely assayed tumor markers in the laboratory, CA-125 is more sensitive for HCC than AFP but far less specific [13]. Currently available screening tests for ovarian cancer include CA-125, transvaginal ultrasound, or a combination of both. CA-125 has provided a useful serum tumor marker for monitoring response to chemotherapy. A rapid fall in CA-125 during chemotherapy predicts a favorable prognosis and can be used to redistribute patients on multi-armed randomized clinical trials. Prostate-specific antigen (PSA) is the most important tumor marker in all solid tumors, indispensable in the management of prostate cancer [14]. However, most currently available screening tests for cancers lack high sensitivity and specificity to be useful in screening the general population, so the differentiation between some benign and malignant tumors is still a

clinical challenge. The advent of Nanotechnology will provide hope in the screening, early diagnosis, and therapy.

4. Nanotechnology and its diagnostic role in cancer

Nanotechnology today deals mainly with two rather different but complementary types of material: nano-sized structures (or nano-particles) and nano-porous materials [15]. Although the number of different types of nano-particles is increasing rapidly, most can be classified into two major types: particles that contain organic molecules as a major building material and those that use inorganic elements, usually metals, as a core [7].

Liposomes, dendrimers, carbon nano-tubes, emulsions, and other polymers are a large and well-established group of organic particles. Use of these organic nano-particles has already produced exciting results [16-22]. Liposomes are being used as vehicles for drug delivery in different human tumors, including breast cancer [16, 17]. Dendrimers, used in MRI as contrast agents, have aided visualization of various pathological processes [18, 19]. Conjugated with pharmacological agents and targeting molecules, organic nano-vectors are potent vehicles for drug delivery and selective imaging of different human cancers [18-22]. Most inorganic nano-particles share the same basic structure—a central core that defines the fluorescence, optical, magnetic, and electronic properties of the particle, with a protective organic coating on the surface. This outside layer protects the core from degradation in a physiologically aggressive environment and can form electrostatic or covalent bonds, or both, with positively charged agents and biomolecules that have basic functional groups such as amines and thiols. Several research groups have successfully linked fluorescent nano-particles to peptides, proteins, and oligonucleotides [23, 24]. Quantum dots are fluorescent nano-particles with sizes of 2–10 nm that contain a core of hundreds to thousands of atoms of group II and VI elements (eg, cadmium, technetium, zinc, and selenide) or group III (eg, tantalum) and V elements (eg, indium) [25, 26]. They can be linked to biomolecules to form sensitive long-lived probes that target and identify specific cellular compounds. As fluorescent probes, QDs have several advantages over conventional organic dyes: their emission spectra are narrow and symmetrical on the basis of their size and material composition, and they exhibit excellent photostability [15]. In addition, they display broad absorption spectra, which make it possible to excite many QDs to different colours with a single excitation light source [23]. This is certainly an advantage in studying multiple biological targets simultaneously in the cell. The high photostability of QDs also allows real-time monitoring or tracking of intracellular processes in vivo over extended periods. Bioconjugated QDs have also been used for DNA hybridization and high throughput genotyping of single-nucleotide polymorphisms (SNPs), the most common type of genetic variation between individuals. SNPs are very good markers for disease causing genes, and they hold further potential for personalized medicine as markers for differential drug responses [27]. As the labeling of individual molecules or cell structures in living cells or tissues is becoming an increasingly important tool in diagnostics, QDs, because of their many advantages over organic dyes, have a large potential for new and improved diagnostic tests in medicine. Supermagnetic nano-particles contain a metal core (eg, iron, cobalt, or nickel) that is magnetically active, and are used as contrast enhancement agents to improve the sensitivity of MRI. Magnetic particles, when coated with an organic outer layer, can also be conjugated to biomolecules and used as site-specific drug-delivery agents for cancer treatment. Iron-oxide-based magnetic materials have been used widely in clinical practice as magnetic resonance agents and in studies of gene expression, angiogenesis imaging, and cellular trafficking [28, 29]. Metal nano-particles in combination with fluorescent active molecules can be used for combined optical and magnetic imaging [30].

The success of many targeted treatments depends on the expression of specific proteins or genes present in cancer cells. For example, in breast cancers, the level of hormone-receptor expression correlates directly with the benefit of endocrine treatments, and the presence of HER2 protein over-expression or gene amplification, or both, is a prerequisite for benefit from the monoclonal antibody, trastuzumab [31-34]. Immuno-histochemistry is the standard method of

determining the expression of hormone receptors or HER2. Although immuno-histochemical methods combined with automated image analysis can quantify precisely the expression of these biomarkers in clinical breast-cancer specimens, these systems are not widely available [35].

5. Emerging nano-materials therapeutics in cancer

Conventional anticancer treatments are nonspecific to target killing of tumor cells, may induce severe systemic toxicity, and produce drug resistant phenotypic growth. An exciting potential use of nanotechnology in cancer treatments is the exploration of tumor-specific thermal scalpels to heat and burn tumors [36].

Traditional surgical instruments such as scissors, clamps, and so on may be replaced by nano-techniques. Nano-surgery at the level of individual living cells or organelles has already been performed [37, 38]. Promising nano-surgery techniques include the use of atomic force microscopy (AFM) with a nano-needle and femtosecond laser surgery.

A Japanese research group [37, 39] has performed analyses and surgery on living cells at nano-scale resolution using AFM and a modified AFM probe. AFM is a type of microscopy in which a probe is scanned across the sample to obtain information about its surface. The information gathered from the interaction of probe with the surface can be as simple as physical topography or as diverse as the physical, magnetic, or chemical properties of the material. The general AFM probe is designed as a 3 μm pyramid with ~30nm end radius on the end of a cantilever which bends as the topography or other properties of the sample change. The bending of the lever is detected by a laser beam detection system and the information is transmitted to a computer, which generates a map of the topography or other properties of interest.

The properties of the cell surface were investigated by contacting and indenting the cell surface with an AFM probe in the shape of an ultra-thin nano-needle. This new technique has several advantages over the traditional microinjection of proteins, peptides, and genetic material into living cells using micro-capillaries. Damage stemming from the use of micro-capillaries due to the shape of the capillaries and the inaccuracy of the displacement is problematic in relation to the manipulation of many cell types. The advantages of the AFM system are the accuracy of the needle and that the ultra-thin needle does not cause fatal damage to living cells [40].

Femtosecond near-infrared (NIR) laser pulses can be used to perform surgery of nanometer sized structures inside living cells and tissues without creating damage. The intra tissue nano-processing is achieved by the generation of high light intensity (1012W cm−2) by diffraction-limited focusing of the radiation of an NIR (λ = 740 and 800 nm) femtosecond laser on a subfemtolitre volume [38, 41]. The energy delivered by the laser pulses breaks down chemical bonds at the targeted site, vaporizing the tissue without causing side effects such as heating of surrounding tissue. The concept “femtosecond laser” refers to the duration of the laser pulses, which is in the scale of femtoseconds. The energy of the short pulses of femtosecond lasers is so high that instead of destroying the tissue by heat generation (like standard lasers) the photons vaporize the tissue, and the result is a clean hole without necrosis of adjacent tissue [42].

Specific targeting of tumor cells is an important goal for the design of nano-therapeutics for the treatment of cancer. Recently, viruses have been explored as nano-containers for specific targeting applications. . Interestingly, there exists a subset of viruses with natural affinity for receptors on tumor cells that could be exploited for nanotechnology applications. For example, the canine parvovirus (CPV) utilizes transferring receptors (TfRs) for binding and cell entry into canine as well as human cells. TfRs are over-expressed by a variety of tumor cells and are widely being investigated for tumor-targeted drug delivery. Towards this goal, CPV virus-like particles (VLPs) produced by expression of the CPV-VP2 capsid protein in a baculovirus expression system were examined for attachment of small molecules and delivery to tumor cells. Dye conjugation also demonstrated that the CPV-VLPs could withstand conditions for chemical modification on lysines. Attachment of fluorescent dyes neither impaired binding to the TfRs nor affected internalization of the 26 nm-sized VLPs into several human tumor cell lines. CPV-VLPs therefore

exhibit highly favorable characteristics for development as a novel nano-material for tumor targeting [43].

Photodynamic cancer therapy is based on the destruction of the cancer cells by laser generated atomic oxygen, which is cytotoxic. A greater quantity of a special dye that is used to generate the atomic oxygen is taken in by the cancer cells when compared with a healthy tissue. Hence, only the cancer cells are destroyed then exposed to a laser radiation. Unfortunately, the remaining dye molecules migrate to the skin and the eyes and make the patient very sensitive to the daylight exposure. This effect can last for up to six weeks. To avoid this side effect, the hydrophobic version of the dye molecule was enclosed inside a porous nano-particle [44].

Liposomes carrying chemotherapeutic small molecule drugs have been approved since the mid-1990s. Liposomes (~100 nm and larger) can give extended circulation times if they are stabilized (DaunoXome, Doxil®) but do not provide intracellular delivery of drug molecules (45). Thus, they are not effective against disease that is resistant to cell surface pumps. Additionally, they provide no control for the time of drug release. Their use is mainly in solubilizing drugs and extending circulation times to favor higher tumor uptake of drugs. Albumin-based nano-particles (Abraxane) were approved by the US Food and Drug Administration in 2005 [46], but are not nano-particle therapeutics, in that they dissolve upon administration into the circulatory system. Nano-crystals of drug molecules (Rapamune, Emend) are also approved for oral administration; however, these nano-particles never reach the bloodstream. These first approved nano-particle formulations prove that nano-particle-based therapeutics can safely be administered to patients and can enhance the safety and efficacy of other drug molecules. However, newer nano-particle systems have great advantages over these early nano-particle products. The types of particles include liposomes, polymer micelles, and polymer-based nano-particles. For each case, e.g., DOX versus SP1049C, NK911 and Doxil®, the nano-particle alters the PK properties of the drug molecule. These nano-particles can provide longer circulation times that allow them to adequately interrogate the body for the presence of tumors if in fact they extravagate into tumors. Small particles like polymeric micelles (<100 nm) have been shown to accumulate more readily in tumors than the larger liposomes [47]. Additionally, movement of a particle throughout a tumor is also size-dependent. It is speculated that nano-particles between 10 and 100 nm in diameter will be optimal for tumor penetration [48].

6. Conclusions

Advances in nanotechnology proved that nanomaterials are highly potent soldiers in the war with cancer. It offers the possibility of new and intriguing opportunities in nano-particle based early detection, diagnosis, and treatment of diseases.

Scientists have already begun to create novel nano-devices to detect and treat cancers, though the ultimate impact remains to be seen due to limited exposure in the broader scientific and medical communities. The use of nano-particles conjugated to antibodies allows the possibility of simultaneously detecting multiple molecular targets in small tumour samples, on which treatment decisions can be made. Protein and gene expression in an individual tumour can be correlated using nano-particle tags. The use of nano-particles in imaging in vivo is rapidly evolving, and could allow simultaneous detection and targeting of cancer-related antigens. Nano-particles offer a new method of tumour targeting, already available in clinical practice, which can concomitantly improve the efficacy and decrease the toxicity of existing or novel anticancer agents.

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