Title	Nanomedicine: A new frontier in cancer therapeutics
Author(s)	Lee, PY; Wong, KKY
Citation	Current Drug Delivery, 2011, v. 8 n. 3, p. 245-253
Issued Date	2011
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Nanomedicine: A New Frontier in Cancer Therapeutics

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Abstract: Nanotechnology is a cutting edge and rapidly evolving technology in medicine. The potential of nanomedicine in cancer therapy is infinitely promising due to the fact that novel developments are constantly being explored. This is particularly the case in the use of nanoparticles in both tumor diagnosis, as well as treatment. This article will attempt to describe some recent advances using nanoparticle drug delivery system in cancer therapy. The evolution history, the challenges and the role of nanoparticles in cancer drug delivery will briefly be discussed together with additional opportunities in cancer therapy. An overall understanding of these issues will help with further advancement of designing better drug delivery system that can be applied clinically.

Keywords: Nanotechnology, drug delivery, tumor therapy.

INTRODUCTION

With more than one million new cases of cancer diagnosed in United states 2009 [1] and the fact that clinical outcome of current cancer therapeutics using either radiotherapy or chemotherapy is far from optimal, the development of an effective cancer treatment is crucial. Furthermore, cancer patients require frequent hospital visits for chemotherapy care and often suffer from undesirable side effects. Efforts have thus been focused on the development of novel drug delivery systems for cancer therapy.

Nanotechnology is impacting positively in pharmaceutics and healthcare. Products in cosmetic and medicine consisting of nanoscale material have already been available in the market. Furthermore National Cancer Institute (NCI) has created an alliance with nanotechnology area in the hope to develop new breakthroughs in therapeutic and diagnostic modalities for cancer treatment. Numerous new generations of nanoparticles for therapeutic and diagnostic application have progressed to clinical trial stage in alliance with NCI. The funding allocated to nanotechnology has been increased to 1 billion in 2006 and predicted to reach 3.7 billion by 2010 [2]. This advance is impossible to emerge without substantial basic knowledge of nanotechnology. The impact of nanotechnology is showing immense promise in improving the tumor therapeutic and diagnostic modalities. Due to the recent success, nanotechnology has received even more public awareness based on its future potential to revolutionize development of clinical useful drug delivery system.

EVOLUTION OF NANOMEDICINE

"Nanomedicine" is the application of nanotechnology into medicine. The evolution from the concept of "nano" to nanomedicine has taken a long journey. The concept of nanoscale was first coined by Richard Feynman [3]. His talk in 1959 at a meeting of the American Physic Society titled "There's plenty of room at the bottom" raised attention about the importance of controlling matter down to bottom atomic or molecular precision. His vision to suggest that manipulation scaling down to atomic nanolevel (starting from the bottom) inspired the eventual development for technology advancement. Although the concept of "nano" originated four decades back, the association with drug delivery and its applications in medicine has not been met with enormous enthusiasm until recently. Indeed, despite the fact that the basic discovery of many traditional drug delivery systems such as liposomes, polymeric micelles, nanoparticles, and dendrimers was made in the 1960's and 1970's, these particles were only confirmed to be in the nanoparticle size range recently. Dating back to more than 150 years ago, the first nanosized colloidal gold particles in fact, were prepared by Michael Faraday. This discovery at the early time served as a precursor for the development and advancement of drug delivery system. In 1994, the first report regarding surface modified liposome nanoparticles for sustained circulation was described [4]. Nanomaterials are often defined as materials or structures in the nanometer size range less than 100nm according to National Nanotechnology Institute [5]. Some have defined nanomateial as materials with boarder range of nanosize scale in 1-1000nm [6].

In 1990s, the discoveries of nanosized devices such as nanowire, nanosensor and quantum dot bioconjugate were successively described [7-9]. Very recently, fabrication of drug delivery system in nanosized by ink-jet printing has also been reported [10]. Indeed, there seems a never ending reporting of new development of "nano" devices everyday. What initiated this recent excitement over the magic of nanotechnology? The attraction of nanoparticles appears to stem from their unique physical, chemical and maybe even biological characteristics, including nanosize, large surface/volume ratio and capability in encapsulation for a wide variety of drugs. For example, the small size can be useful in overcoming biological and biomedical barrier, easily enter the cells and organelles and can interact with intraceullar proteins as most cells are only 10-20μm in size. In this re-

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1567-2018/11 $58.00+.00

spect, the framework of nano periodic table was announced only very recently in 2009 in the NSF nanoscale science and engineering meeting [11].

All together, the ultimate goal of using nanotechnology is to develop an effective drug delivery device with programmed functions. The history of nanomedicine evolution reveals the promise of nanotechnology in the development of drug delivery systems to overcome the existing challenges in cancer therapy.

CHALLENGES IN CANCER DRUG THERAPY

The clinical outcome of treatment with many anticancer drugs has not been met with universal success as hoped. The major hurdle of current anticancer drugs is to kill tumor cells specifically without significant side effects. The theoretical basis on which existing anticancer drugs exert their effects, relies on the higher mitotic rate in the tumor cells than that of normal cells. This often results in high systemic toxicity and the therapeutic window is narrow. Repeated dosage is thus limited. The concept of specific targeting has emerged and is crucial to help reduce uptake by normal tissue and increase the payload of the drug inside the tumor. Furthermore, as more than 40% of anticancer drug is poorly soluble in aqueous environment, the ultimate bioavailability and therapeutic efficiency can be significantly hampered. Conventionally, solvents and emulsifiers have been used to dissolve poorly water soluble anticancer drugs. They are suggested to be potentially carcinogenic [12] and can be toxic to liver and nervous system [13].

a) Tumor Barrier

Understanding the cancer structure is paramount to overcoming the challenges in the design of the drug delivery system. Cancer has a unique architecture characterized by vascular pores, heterogeneous vasculature and increased blood supply. Tumor tissues are characterized with disorganized and leaky tumor vasculature with heterogeneous gaps and pores (ranging from 200-800nm). This allows macromolecules up to 400-500nm leaving the vascular bed and accumulating inside the interstitial space in tumor. Together with poor lymphatic drainage, this contributes to the significant retention of marcomolecules, which is known as the enhanced permeation and retention (EPR) effect [14]. Besides the defective vascular architecture, the high vascular density keeps blood supply for tumor growth. This supports enormous amount of tumor tissue growth, move than in normal state. Thus, a major principle of cancer chemotherapy is to block, or to reduce tumor angiogenesis. Another action would be to induce apoptosis of growing tumor cells. Meanwhile, delivery of the therapeutic agents systemically is often compromised by the body's normal physiology, including hepatic and renal clearance, uptake by RES (reticuloendothelial system) organs, degradation by enzymes as well as by endosome/lysosome, resulting in less dose level to the tumor cells.

The physiological barrier depends partially upon the size of nanoparticle carriers, in which particle size can determine the clearance kinetics of these nanoparticles. It has largely been reported that the ideal size of nanoparticle for cancer treatment lies between 70-200nm [15, 16]. The size of nanoparticle smaller than 10 nm is rapidly eliminated by kidney clearance. On the other hand, those bigger than 200 nm are primarily taken up by the RES system, such as Kupffer cells in the liver and spleen [17]. Further entrapment of the nanoparticles escaping from circulation is effected by fenestra and sinusoids in liver and spleen [18-20] if the particle size is bigger than the pore size of fenestra or sinusoids (up to 150nm) [21, 22]. Particle size can also determine the efficiency of tumor uptake. In addition to the lower size limit, due to the unique feature of the leaky vasculature in tumors, known as EPR (enhanced permeability and retention) effect, 400-500 nm is upper size limit for the extravasation of nanoparticle carrier into tumor tissue. The size growth of nanoparticle can occur by aggregation. Small particles have larger surface area and higher particle number than bigger particles at the same mass concentration. Thus, extremely small nanoparticles (diameter $\leq$ 3nm) could not be easily stabilized and aggregated rapidly [23]. This could be detrimental in blood circulation and may result in blockage of the capillaries. Considering the ability to pass through the capillaries, the nanoparticle size without aggregation is more possible to prevent thromboembolism compared to microparticle size and thus can be beneficial for systemic delivery. Surface characteristics can control the extent of particle aggregation at interstitial sites. For example, hydrophilic nanoparticles are more likely to interact poorly with ground substances at interstitial sites.

b) Blood Brain Barrier (BBB)

The blood brain barrier is formed by tight intracellular gap junctions of endothelial cells and high density of cells filtering against the entry of molecules into the cerebrospinal fluid. Only 5% of current drugs can pass through the BBB and affect the CNS. It is challenging even for any drug in the nanosized range to permeate through the BBB. Reports have demonstrated that both size and hydrophobicity represent the major components in determining the crossing of the BBB and thus are both crucial in the design of "nano" drugs for CNS delivery [24, 25]. For example, small molecule histamine (~100 Da, ~0.01nm) is not able to pass through the BBB due to highly dense hydrophilic hydrogen-bonded end groups [26]. Apparently, drugs that are lipid soluble with molecular size range between 400-600Da can pass through the BBB. However, the tight gap junction at the BBB can prevent the crossing of even very hydrophobic molecules [27, 28].

c) Systemic Toxicity

The dosage of systemic delivery of drugs can be restricted by the systemic toxicity. This is particularly true in cancer drug delivery since most of the cancer therapeutics is highly toxic. Depending upon the route of administration and site of application, systemic toxicity varies. One way of reducing this is by local delivery, as a high concentration of drugs can be achieved and stays at the local tissue, with resulting lower systemic toxicity.

In the circulation, systemically administered cancer drugs have higher chance to interact with and be taken up by RES organs, kidney, marrow and central nervous system, resulting in the apoptosis of healthy cells. A nanoparticle carrier

system can minimize systemic uptake is thus crucial for reducing systemic toxicity and more effective cancer therapy.

d) The Mode of Drug Delivery

The route of administration can affect the delivery of nanoparticle and thus the availability of encapsulated drug at the action site. Oral administration of nanoparticle drugs has to first pass through the stomach. Since absorption occurs in stomach and largely in intestine [29], strategies have been employed to resist premature acid degradation in the stomach (pH~2.5) [30]. For local tumor delivery in GI tract, some nanoparticle carriers have been designed to use the unique pH environment in GI tract to control the drug availability. It has been reported that pH sensitive nanoparticles can trigger the release of the encapsulated drug in response to the local pH stimuli, resulting in inhibiting tumor growth locally in the GI tract [31]. On the other hand, some oral drugs have been used to treat tumor in systemic organs. In this case, it may be advantageous to design a carrier system resistant to liver metabolism in addition to the pH sensitive feature, since many oral drugs have to go through first pass metabolism in the liver after absorption. A common mode of delivery for cancer treatment is the intravenous (i.v) route administration which allows the nanoparticle directly going into the bloodstream. Delivery into the tumor destination via the systemic route is considerably more challenging than local administration since drugs will be distributed widely in systemic organs such as heart, brain, liver, kidney and RES organs. In this case, drugs in blood circulation will have more interaction with the systemic organs which may increase the chance for rapid clearance by kidney and RES organs. The extent of tissue distribution can be affected by membrane permeability, the blood supply and the binding of drugs to specific targeted tissue (i.e. tumors). To induce the preferential distribution in tumors via systemic administration, current nanotechnology has been used to design nanoparticle carrier with size and surface chemistry for improving the cell uptake and binding inside tumor [32, 33].

ROLE OF NANOTECHNOLOGY IN DRUG DELIVERY

The importance of the nanoparticles in drug delivery is due to multifactorial features, including the size, structure, encapsulation efficiency, release characteristics and the capability to constitute surface modification. The extremely small size of nanoparticles gives a high surface/volume ratio for better membrane uptake of drug. The nanoparticle carrier can be a good nanotechnology platform to improve the drug efficacy and tumor targeting. An example is the liposomal encapsulation system, which has shown promise in drug delivery by displaying an enhanced binding to the tumor endothelial cells and intracellular uptake through endocytosis [34, 35]. A new generation of liposome encapsulation system is developed by combining physical or chemical modification. The physical modification of liposome carrier platform with local ultrasound enhances the tumor site-specific therapeutic efficiency [36, 37]. However, one shortcoming of this physical approach is that healthy tissue cannot be protected, and bystander damage is also seen [38]. An alternative is chemically modified liposome and has been described to improve the systemic stability and tumor specificity [39]. Furthermore, the use of nanoparticle carrier can develop in drug encapsulation as a drug reservoir system. Utilization of inherent biodegradable nanomaterial such as a composite of liposome and polymer can sustain the drug release in a controlled and regulated manner [40]. Multiple layer of liposome has also been suggested to control the release in a prolonged period of time [41]. This design is favorable to the development of a clinically useful system since the effective dose at therapeutic range can last for an extended period of time.

This is particularly useful if the anticancer drug requires systemic delivery, as it is difficult for sufficient nanoparticle carrier to bypass the physiological, tumor and even in case of brain tumor blood brain barriers and finally reach the tumor destination.

An ideal nanoparticle drug carrier should be able to direct the drug payload into tumor and the entire drug should release locally within the tumor environment. This would be beneficial to reduce systemic toxicity and enhance the efficiency of cancer therapeutics. But the current development and design is not perfect yet. To design an ideal drug delivery system for cancer therapy, there are four criteria: (1) The nanopaticle carrier should be biodegradable and biocompatible in considering the safety concerns (2) The nanoparticle carrier should be stable in the circulation. (3) The accumulation of nanoparticle carrier should be preferential and efficient in the tumor (4) The drug should release efficiently and remain within the tumor tissue structure.

The following will describe several significant merits of nanotechnology development in drug delivery:

a) Basic Nano Systems

Lipid/Polymer Nanoparticles

Lipid or polymer nanoparticles provide significant contribution in the strategy to improve therapeutic properties of drugs [42]. Considerable efforts have been attempted to complex cationic lipids and/or polymers with plasmid, DNA or siRNA for antitumor therapy application [43]. Furthermore, numerous drug delivery systems such as liposomes, nanospheres and polymeric nanocarrier, if adsorbed or covalently binded with protein antigens, can be a strong adjuvant to phagocytic cells [44-46]. Positively charged lipid based carrier are known to trigger strong immune response when injected into the body. Positively charged liposomes are more able to induce the antigen specific Th1 type immune response [47, 48] when compared with neutral or negatively charged liposome. This explains the recent trend in the use of liposome as an immunoadjuvant against cancer. Nonetheless, this can be problematic when it is injected systemically due to the possible uptake of highly toxic anti-cancer drug by T cells and resulting in the killing of these T cells.

Rapid clearance of drugs from the circulatory system by phagocytic cells is common. Major phagocytic cells like macrophages and dendritic cells are well known in playing key roles in initiating and generating the immune response [49]. Opsonization is one predominant mechanism (a surface deposition of nonspecific opsonic factors in blood such as fibronectin, immunoglobins and complement proteins) to aid and trigger the recognition by macrophages and dendritic

cells via complement activation when the drug is injected systemically [49, 50]. By activating complement system, the positively charged nanoparticle carrier facilitates clearance from blood by the Kupffer cells of RES organs [51]. Besides surface charge, particle size is another predominant factor to determine the extent of immune response, in which the extent of opsonization decreases when the size of liposome decreases [52]. Nonetheless, positively charged lipid based nanoparticle can still be a promising option in cancer therapy since it facilitates the binding and uptake by the tumor cells, resulting in increased therapeutic efficiency and anti-tumor activity [53, 54]. Positive charge is often shielded in case of designing a long circulating carrier that is potentially useful for systemic administration. As blood circulates through RES organs more than the tumor, there is a higher possibility for the nanoparticle to interact with RES organs. If the positive charge on nanoparticle surface is not protected, RES organs will take up significant amount of nanoparticles. Thus, surface engineering of liposomes has been attempted to shield the charged surface using pegylation (conjugation of Poly(ethylene) glycol, abbreviated as PEG). This approach can enable reduced opsonization and thus the RES uptake.

In the case where a large portion of new drug candidate is poorly water soluble, leading to reduced development potential because of low drug bioavailability, one option to enhance the drug bioavailability is nanosuspension. This is the colloidal dispersion of drugs. Nanosuspension has been reported to be useful for drugs that are not soluble in both aqueous and oil phase. For drugs that are poorly soluble in water but highly soluble in oil, oil in water microemulsion is an alternative. Lipidic nanoparticle carrier composed of emulsifying wax with surfactant stabilizer Brij has been reported to encapsulate successfully a wide range of lipophillic drugs [55-57]. It has demonstrated high encapsulation efficiency (>95%) of lipophillic drugs. This means that high loading of lipophillic drug can be achieved by the formulation. It is highly stable and compatible with blood. Our preclinical study in neuroblastoma bearing model showed that the lipidic carrier is able to reduce toxicity and enhance the anti-tumor activity [58].

Polymeric Micelles

Polymeric micelles are formed in aqueous solution as core shell structures and often composed of block copolymer with hydrophobic and hydrophilic components. The hydrophobic core serves as a reservoir for hydrophobic drugs and the hydrophilic shell reduces opsonization and subsequent rapid clearance. This allows the water insoluble drugs to be entrapped in the core for solubility. The features of nanosize, easy manipulation of surface chemistry make them suitable as nanoparticle carriers. Change of polymer block composition to form complex with polycationic polymer such as poly(L-lysine) or polyaspartamide derivative, poly{N-[N-(2-aminoethyl)-2-aminoethyl]aspartamide} (PAsp(DET)) has developed as a carrier to deliver a wide variety of siRNA and plasmid DNA for antiangiogenic gene therapy [59, 60]. Efficient *in vivo* and *in vitro* gene delivery in tumors has been observed using the polyplex micelles [61]. Surface modification of the polyplex micelles with RGD ligands has also shown improved intracellular trafficking [62].

Gold Nanoparticles

Gold nanoparticles have commonly been used as nanosized diagnostic and therapeutic agents. Due to the unique chemical structure, the use of colloid gold has shown promise in drug delivery. Gold nanoparticles functionalized with citrate [63, 64] and transferrin [65] played a vital role in facilitating cellular internalization. Recent study of the citrate conjugated gold nanoparticle determined how the size of nanoparticles could influence the ability of cellular internalization. By using induced coupled-plasma atomic emission spectroscopy (ICP-AES), it was found that the particle size of citrate conjugated gold nanoparticles around 50nm are mostly internalized by HeLa cells (in a size range of 14-100 nm) [66]. Our lab routinely determines the tissue distribution in different organs of gold nanoparticle compounds using induced coupled plasma- mass spectroscopy (ICP-MS).

Gold composites have further been explored as "nanobullets" and "nanobombs" for killing cancer. Laser induced explosion of the light absorbing gold nanoparticles at local tumor site was reported to kill cancer cells [67]. The hollow gold nanoparticles can be developed as targeted anti-cancer drug carrier by decorating the surface with targeting molecules. After homing to the tumor site, the cancer cells are "cooked" by laser irradiation [67, 68]. Further, this system can be a dual functional delivery system as a control drug release carrier and a photothermal ablation mediator when doxorubicin was coated on the inner and outer surface of hollow gold nanoparticles. The near-infrared irradiation was used to mediate both the release of doxorubicin and photothermal ablation of cancer cells [69].

Effort has been made to entrap gold nanoparticles into a lipidic carrier to enhance the potential in antitumor application. Gold porphyrin has been documented as a very potent antitumor drug [70]. We complexed a lipidic nanoparticle carrier with gold porphyrin and demonstrated a reduced systemic toxicity and enhanced the tumor uptake [58].

Dendrimers

Dendrimer are highly branched synthetic polymers first invented by Tomalia in early 1980s [71]. It consists of center core, integral region and numerous functional end groups. The growth of polymer occurs in outward direction from central core by stepwise polymerization. It is characterized with numerous cavities in the core structure creating channels and cages. Precise control of size can be achieved by the extent of polymerization. The intracellular uptake of dendrimer by receptor mediated endocytosis can be aided using the conjugation of biotin [72]. Dendrimers contribute to drug delivery either by binding the drug on the periphery as prodrug or entrapping the drug in the center core. Recent progress has been made to dendrimer formulary as a biocompatible drug carrier for cancer targeting therapy. Surface of dendrimer enables the coating with poly(ethylene) glycol (PEG) or targeting ligand for folate receptors and showed potential in improved cellular targeting for cancer therapy [73,74].

Carbon Nanotubes (Naked and Functionalized)

Carbon nanotubes consist of graphite sheets rolled up into tubes and belong to the fullerene family. They can be

single walled or multi-walled nanotubes. The size and diameter varies between the two structures. The single-walled has the diameter of 0.5-3nm and length of 20-1000nm while the multi-walled comprising of multiple layer has the diameter of 1.5-100nm and length of 1-50μm [75] with interlayer spacing of approximately 0.34 nm [76]. They are considered as inert and hydrophobic nanomaterials. Carbon nanotubes exhibit a unique feature of thermal, electric and mechanical properties. They can absorb energy through irradiation, magnetic field or near-infrared and transform the energy into heat. Surface functionalization with acetate nanocomposites can provide ways to resist better thermal degradation and demonstrated better thermal stability [77]. To improve the water solubility, surface functionalization with water soluble ethylene glycol or PEG has been explored [78]. Nonetheless, carbon nanotubes could be associated with internal tissue damage and subsequent development of mesothelioma, which currently remain major concerns of the FDA [79].

Modification for Drug Delivery System-Poly(Ethylene) Glycol (PEG)

PEG first emerged in the early 90s for surface modification of liposomes. Due to the neutral charge and inert feature, PEG reduces opsonization and stabilizes the nanoparticle carrier by adding steric barrier. Surface modification of nanoparticles using PEG (known as pegylation) is often referred as "stealth", as they are more able to escape the surveillance of the RES organs and thus have prolonged circulating time in blood. However, too much PEG insertion can be disadvantageous. This has been shown in the case of doxorubicin where excessive peglylation hinders the uptake of drug by the tumor cells by reducing the binding on the cells [80].

Pegylated nanoparticles are able to enhance preferential uptake into the tumor. The nanoparticles can be uptaken by passive targeting via diffusion across the leaky blood vasculature. RES competes with the tumor for the uptake of nanoparticle. Thus the longer stay in the blood circulation allows higher chance of the nanoparticle getting into the tumor tissue.

b) "Smart" Drug Delivery System

Novel nanotechnology serves as platform in the design of smart drug delivery system. Smart drug delivery system aims to further improve drug delivery efficiency by (1) additional triggering the controlled release in response to local environmental stimuli (2) specific targeting of tumor tissue and/or (3) prolonging the circulation time.

Heat Sensitive Nanoparticle Carriers

The first report of controlled release drug delivery system was described by Dominici and Macroni in 1965 [81]. Heat sensitive or pH sensitive nanoparticle carrier has been designed to control release of the drug. The early effort in the design of temperature sensitive drug carriers was mainly focused on developing injectable and a controlled release drug carrier. Thermosensitve polymer has gained a lot of interest in drug delivery since it has demonstrated prolonged and better control of drug release. The block (diblock, triblock, multiblock) copolymer consisting of biodegradable polyesters and PEG demonstrate controlled biodegradability and good biocompatibility. For example, PEG-PLGA-PEG can form polymeric micelle and induce micelle growth at body temperature and serve as drug reservoir [82]. At body temperature, the polymeric micelle can release the drug by surface or bulk degradation over time or diffusion via polymeric matrix [83]. Acidic environment or high water content within the polymer triggers the degradation [83]. The composition and molecular weight also determines the degradation rate [84]. In past decades in drug delivery, tumor targeting was one important issue as it could provide one potential solution for reducing the systemic toxicity of cancer drugs and improving the therapeutic efficiency. Recent efforts have attempted to develop thermosensitive nanoparticle carrier for tumor targeting [85-87] after systemic administration. Nakayama *et al* has designed polymer consisting of hydrophobic block and a thermo-responsive poly(*N*-isopropylacrylamide–*N*,*N*-dimethylacrylamide) (PNIPAAm–PDMAAm) hydrophilic block [88]. This block copolymer has lower critical solution temperature (LCST) at around 40°C [88]. Rapid drug release was observed in response to the thermal stimuli by local tumor heating to or above the LCST temperature [88]. Local heat is produced by the insertion of hyperthermia applicator within tumor [89]. However, the applicator is best used for tumors at abdomen, pelvis and extremities possibly due to the ease of applicator accessibility to the tumor and insufficient heating effect at deeper tissue. Another exciting approach is to functionalize the nanoparticle carrier with magnetite (Fe_3O_4) for magnetic targeting and controlled release the drug locally in tumor site triggered by pH and heat [85].

pH Sensitive Nanoparticle Carriers

The pH sensitive drug delivery system uses the change in pH in the intra-cellular compartment to switch on the drug release. The carriers are often biodegradable liposomes, polymers or hydrogels. The therapeutic drug release can be triggered by swelling in the lysosomal environment or by the acidic or alkali catalyzed hydrolysis degradation, resulting in the entrapped drug payload into the cytosol. The pH sensitive liposome containing PHC (lipid palmotyl homocysteine) has long been proposed as a useful application for targeting drug delivery in a region that is slightly below the physiological pH (between pH 6 to pH 7.4) such as tumor, inflammatory and infection sites [86]. The pH sensitive polymer has also been developed for the escape of the intracellular barrier. Liposome coated or complexed with proton sponge Polyethyleneimine (abbreviated as PEI) enters the acidic lysosome/endosome compartment via endocytosis. The amino group is able to sequester protons and trigger the entry of chloride ions and water molecule into the lysosome/ endosome compartment, leading to swelling and the eventual rupture of the lysosome/endosome. Recently, long circulating and biodegradable pH sensitive polymeric nanoparticle carrier composing of sulfonamide was made which could respond to even sharper pH changes (between pH 6.6 to pH 7.4 normal blood pH) [64]. This design is characterized with shielding of polymeric nanocarrier by cell penetrating peptide TAT peptide at physiological pH, and unshielding at around or below pH 6.8 by detachment of TAT peptide. Another example is the formation of nanoparticles by direct

conjugation of cisplatin with hydrophobic segment of di-block polymers via hydrozone bond. The cleavage of the hydrozone bond mediates the sharp response of drug release corresponding to slight change of environment acidity [90].

One concern of this is that the encapsulated drug has to be released at the free drug level to reach the therapeutic dose. Indeed, the carrier can be collapsed by the pH gradient in acidic environment such as lysosome/ endosome compartment and releases the drug in a controlled manner. In addition, the rate of drug release from pH sensitive carrier also depends on the encapsulation method and drug types. For example, cisplatin entrapped in the liposome showed minimal antitumor activity despite the accumulation in the tumor tissue [91] while encapsulated doxorubicin in liposome demonstrates antitumor activity [92].

Antibody Conjugated Nanoparticle Carrier

Free cancer therapeutics or nanocarrier constitute a platform for conjugating with monoclonal antibody that is specific to antigen expressing on the tumor cells or tumor endothelial cells. A variety of antibodies and growth factor receptors, such as nerve growth factor (NGF), vascular endothelial growth factor (VEGF) and epidermal growth factor (EGF) [93-98], which are overexpressed in the tumor have been used to improve cancer drug delivery specificity. It can also reduce the systemic toxicity since the antibody conjugates can be developed to shield the free anti-cancer drugs. The solubility is further enhanced due to the hydrophilic nature of the antibody. An example of this is Taxane conjugate, an antibody conjugate with Paciltaxol. It has been reported to enhance the solubility and tumor targeting drug delivery [99].

Magnetic Directed Drug Delivery System

Magnetic nanoparticles and magnetic nanoparticle carriers including liposomes and dendrimers have been developed for cancer drug delivery. The carriers are made either by encapsulating drugs in the magnetic nanoparticles or as a composite with the magnetic nanoparticle carrier. The use of magnetic nanoparticle carrier has been developed to direct drug delivery to the tumor site. One way of magnetically directed cancer drug delivery is by locally applying magnetic field at the tumor target site using MRI (magnetic resonance imaging). The approach should be feasible in most tumor sites since the magnetic field strength of MRI is sufficient enough to apply to most soft tissue and mineralized tissue. The magnets are often iron oxides or gadolinium containing nanoparticles. The magnet can be either surface bound or encapsulated into the nanoparticles. If the magnet is bound on the surface of the carrier rather than being entrapped in the carrier, it will increase the systemic overexposure to gadolinium. Overexposure to gadolinium increases the risk of systemic toxicity associated with more tissue and blood vessel deposition. Another way in magnetic drug targeting is to complex with pH sensitive polymeric matrix carrier and this has been suggested to be useful in the acidic gastrointestinal tract environment [100]. Targeting diagnostic system based on magnetic nanoparticle can be used to identify the location of the tumor by functionalizing with ligand binding specifically to the tumor receptor.

CURRENT STATUS OF NANODRUG THERAPY

There are currently several nanodrugs available in the market for cancer treatment such as Doxil®, Abraxane®. Doxil® is liposomal encapsulated doxorubicin. With the so called stealth liposome using pegylation, dose frequency can be minimized, resulting in a longer treatment cycle repeated every 4 weeks. Due to the long circulating feature, the doxorubicin encapsulated in stealth liposome demonstrated more favorable pharmacokinetic. Clearance and volume of distribution decrease by at least 250 fold compared with free doxorubicin [101]. Clinical study also shows that it is associated with modest toxicity and side effects in patients [102]. However, targeting strategy could further be improved. This may be done effectively by using active targeting, for example conjugation with tumor specific antibody or ligands resulting a tumor receptor mediated internalization.

TARGETING CANCER STEM CELLS: THE NEXT STEP

In recent years, cancer stem cells have received tremendous attention. Increasing evidence have showed that cancer stem cells can initiate and sustaining tumor growth and can be associated with tumor recurrence [103, 104]. Recent work has also suggested that cancer stem cells could be responsible for cancer drug resistance [105]. Identification of their location and targeting them will require novel therapeutic design in cancer treatment [106].

FUTURE HORIZONS

The ideal drug delivery system needs to enhance the efficiency of delivery and payload of the drug specifically in the tumor. These depend upon the accumulation, retention and release of the drug in the tumor tissue. The conjugation with PEG as described previously is one promising option to increase the uptake by the tumor over the RES. The conjugation of ligand to tumor specific receptor such as EGFR is an active targeting strategy that can increase binding to the tumor cells. Thus the retention and the intracellular uptake of the drug can further be improved. Thus, the approach of combining conjugation of both PEG and tumor specific ligands can potentially further increase the accumulation as well the bioavailability of drug in the tumor. More work in development is currently underway.

Given that nanoparticle carrier get into the tumor and perform effective anti-tumor activity, it is important to develop an non-invasive in situ detection for investigating tumor recurrence and tracking the location of metastasized tumor cells. For example, the carrier that can deliver the anti-tumor drug but also covered with particles for tumor surveillance and imaging would be ideal. If the drug demonstrates good imaging potential, it would be possible to combine the therapeutic and diagnostic approaches in the same nanoparticle carrier. Thus, "theragnostics" (therapeutic + diagnostic) may carry the next potential in nanomedicine development for cancer.

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Received: September 14, 2010 Revised: November 27, 2010 Accepted: December 05, 2010