Carbon Nanotubes Based Systems for Targeted Drug Delivery : A Review

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Abstract

The development of new and efficient drug delivery systems is of fundamental importance to improve the pharmacological profiles of many classes of therapeutic molecules. Currently numerous types of drug delivery systems are available which have been successful in selective cases yet there are major drawbacks involved which are damage of healthy tissue and painful methods of inducing the drug. Nanotechnology harnesses current progress in chemistry, physics, materials science, and biotechnology to create novel materials that have unique properties because their structures are determined on the nanometer scale. Carbon Nanotubes have been recognized as the quintessential nano-materials and have acquired the status of one of the most active fields of nano-science and nanotechnology.Carbon nanotubes have potential therapeutic applications in the field of drug delivery, diagnostics, biosensing and tissue engineering by acting as scaffolds. Functionalized carbon nanotubes can also actas vaccine delivery systems.They can pass through membranes, carrying therapeutic drugs, vaccines and nucleic acids deep into the cell to targets previously unreachable by conventional drug delivery systems.

1. Introduction.

The development of new and efficient drug delivery systems is of fundamental importance to improve the pharmacological profiles of many classes of therapeutic molecules and its unlimited potential in to improve human health. Nanotechnology on the other hand deals with engineering of functional systems at a molecular level. Biomaterials with nano-scale dimensions can be used as controlled release reservoirs for drug delivery. Drug delivery systems can be synthesized using controlled composition, shape, size and morphology. Their surface properties can be manipulated to increase solubility, immune compatibility and cellular uptake to match the requirements of the target area of delivery. The limitations of current drug delivery systems include suboptimal bioavailability, limited effective targeting and potential cytotoxicity. [1]Carbon Nanotubes (CNTs) are the potential nanomaterials which can be used for trans locating and transporting materials [2]. The unique intrinsic physical and chemical properties of CNTs have been explored over the past few years. From unique electronic properties and a thermal conductivity higher than diamond to mechanical properties where the stiffness, strength and resilience exceeds any current material, carbon nanotubes offer tremendous opportunities for the development of fundamentally new material systems [3] CNTs exhibit less toxicity and are not immunogenic after functionalization which paves way for tremendous applications in the field of Nanobiotechnology and Biomedicine.

2. Structure of CNTs.

The bonding system between the carbon atoms in carbon nanotubes is similar to the arrangement in graphite. CNTs are hollow nanostructured materials consisting of carbon atoms bonded with three neighbor atoms via sp^2 bonds which are stronger than the sp^3 bonds present in diamond giving the structure it unique strength. CNTs are considered to be rolled up graphene sheets. Carbon occurs in many forms and the properties of the substance depend mainly on the structure and arrangement of the atoms. The structural arrangement of the carbon tubes in CNTs is responsible for the unique mechanical and chemical properties.

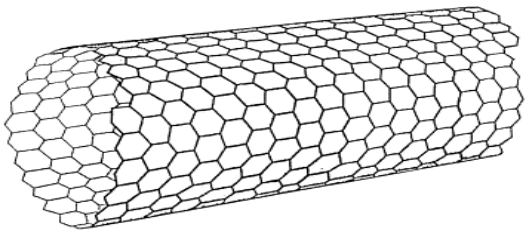

Figure 1: A part of a zigzag single wall carbon nanotube (SWNT) [4]

There are two types of CNTs: Single wall Carbon Nanotubes (SWCNT) and Multi Wall Carbon Nanotubes (MWCNT). One graphene sheet rolled up will form a SWCNT, whereas the arrangement of more than one concentric graphene sheets gives a MWCNT. The MWCNTs have more strength when compared to SWCNTs whereas the SWCNTs can be easily twisted and are more liable than MWCNTs.

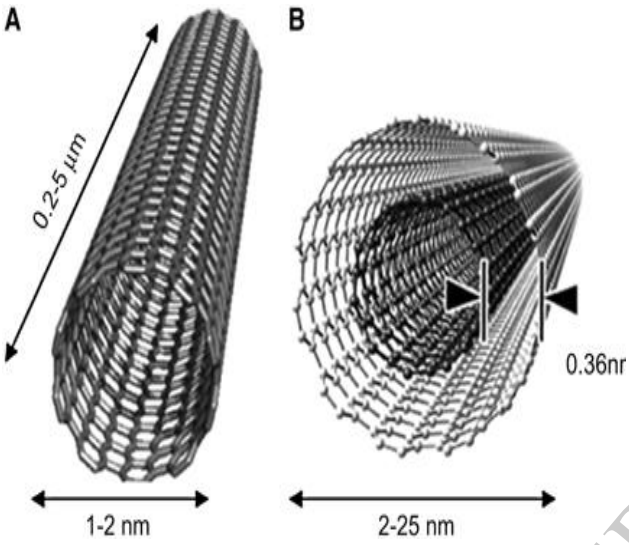

Figure 2: Conceptual representation of Single wall Carbon Nanotubes (SWCNT) (A) and Multi Wall Carbon Nanotubes (MWCNT) (B). [5-6]

3. Fabrication of carbon nanotubes.

3.1 Arc discharge.

The carbon arc-discharge method is a high temperature process that can be used for the production of nanotubes.Mass production of fullerenes was first achieved using arc discharge with the Kratschmer–Huffman method [7]. The derived product and the yields are mainly dependent on the atmosphere and catalysts utilized. This method is probably one of the simplest methods for synthesizing nanotubes on a large scale. In the carbon arc-discharge method, an arc is ignited between two graphite electrodes in a gaseous background (usually argon/hydrogen) [8] Single-walled carbon nanotubes synthesis: a direct comparison of laser ablation and carbon arc routes. [9-10]

The arcing evaporates the carbon and meanwhile it cools and condenses that some of the product forms as filamentous carbon on the cathode. The optimization of metals being included in the anode led to the growth of single-walled carbon nanotubes [12-13].

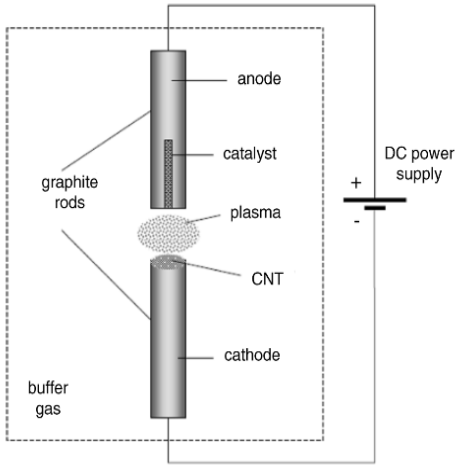

Figure 3: A Schematic representation of Arc Discharge Process [11]

3.2 Laser ablation.

In the laser ablation process, a pulsed laser is made to strike at graphite target in a high temperature reactor inthe presence of inert gas such as helium which vaporizes a graphite target.[14]The nanotubes develop on the cooler surfaces of the reactor, as the vaporized carbon condenses. A water-cooled surface is also included in the most practical systems to collect the nanotubes [15].

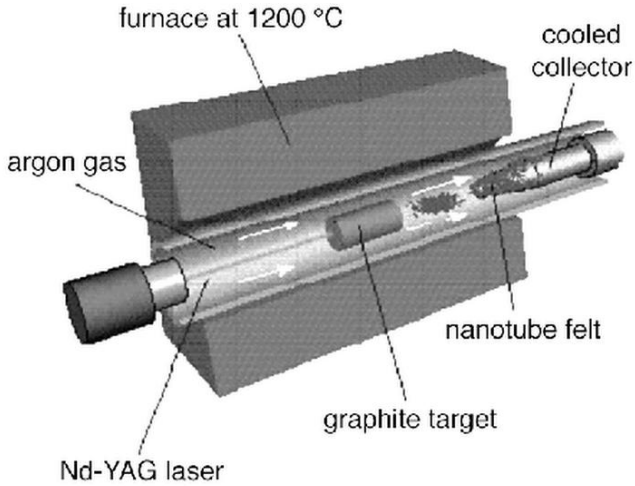

Figure 4:Schematics of a laser ablation set-up[16]

The laser ablation method yields around 70% and produces primarily single-walled carbon nanotubes with a controllable diameter determined by

the reaction temperature. However, it is more expensive than either arc discharge or chemical vapor.

3.3 Chemical Vapor Deposition.

The catalytic chemical vapor deposition of carbon was reported in 1959[17], but only in 1993 carbon nanotubes were formed in this way [18]. During CVD, a substrate covered with metal catalysts, like nickel, cobalt, iron, ora combination is heated to approximately 700°C. The growth starts after two gases are passed through the chamber, a carrier gas like nitrogen, hydrogen or argon, and some hydrocarbon gas like acetylene (C_2H_2) or methane (CH_4). The synthesis production yield, which indicates the amount of carbon nanotubes in the converted carbon, reaches 90% [19]. CVD is commonly used for the industrial purposes because the method is already well investigated and offers acceptable results on the industrial-scale.

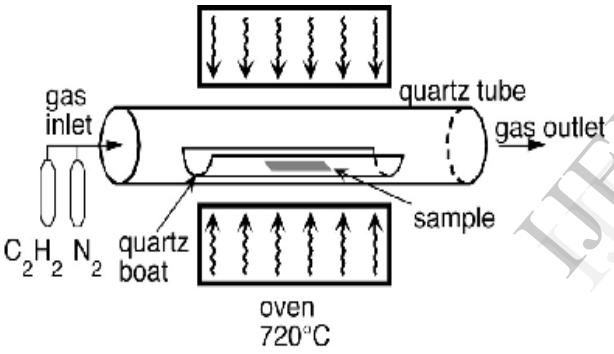

Figure 5: Schematics of a CVD Deposition Oven[20]

3.5 Flame synthesis method.

Hydrocarbon flames with metal aerosol catalysts provide a unique combination of the chemical and catalytic factors that are conducive to initiation and growth of carbon nanotubes [21]. Gases (CO, CH_4, C_2H_2, C_2H_4 and C_2H_6) present in the post flame environment form a diverse source of gaseous carbon [22]. Catalysts in appropriate form (substrate or aerosol) provide the reaction sites for deposition of solid carbon. The geometry and characteristics of the catalysts play an important role in the structural properties of the carbon nanotubes. Single-walled nanotubes have been observed in the post-flame region of a premixed acetylene/oxygen/argon flame operated at 50 Torr with Iron Pentacarbonyl vapor as the catalyst.The fuel gas is burnt in a controlled environment to a temperature of ~800^oC to enable the formation of CNTs on the small metal particles.As optimization parameters the fuel gas composition, catalyst, catalyst carrier surface and temperature can be controlled. [23]

3.5 Silane Solution Method.

Carbon nanotubes were produced using a silane solution method, in which a substrate such as carbon paper or stainless steel mesh was immersed in a silane solution of a metal catalyst, preferably Co: Ni in a 1:1 ratio; and a feedstock gas containing a carbon source such as ethylene was fed through the substrate and the catalyst deposited thereon while the substrate was heated by applying an electrical current thereto [24]. Thus, a reaction occurs between the catalyst and the gas to yield CNTs supported on the conductive substrate.

4. Purification of CNTs.

Nanotubes generally contain a large amount of impurities such as metal particles, amorphous carbon and multishell. There are different stages in purification of nanotubes.

4.1 Oxidative treatment.

Oxidative treatment of the SWNTs is a good way to remove carbonaceous impurities or to clear the metal surface [25].

The main disadvantages of oxidation are that not only the impurities are oxidized, but also the SWNTs. Luckily the damage to SWNTs is less than the damage to the impurities [26].

These impurities have relatively more defects or a more open structure. Another reason why impurity oxidation is preferred is that these impurities are most commonly attached to the metal catalyst, which also acts as oxidizing catalyst. Altogether, the efficiency and the yield of the procedure are highly dependable on a lot of factors, such as metal content, oxidation time, environment, oxidizing agent and temperature. The fact that metal acts as oxidizing catalyst, the metal content should certainly be taken into consideration, when looking at the oxidizing time. For example, when the temperature is raised above 600 °C, SWNTs will also oxidize, even without catalyst60. This is the case with thermal54, fixed air and pure oxygen oxidations. These can easily oxidize all the components, so the temperature and the time should be in good control [25].

4.2 Acid Treatment.

In general the acid treatment will remove the metal catalyst. First of all, the surface of the metal must be exposed by oxidation or sonication. The metal catalyst is then exposed to acid and solvated. The SWNTs remain in suspended form. If treatment is done using HNO3, the acid only has an effect on the metal catalyst. It has no effect on the SWNTs and other carbon particles [27]. IfHCl is used for treatment, the acid has a considerable effect on the SWNTs and other carbon particles. Hence HCl is considered as the ideal refluxing acid [28-29].

4.3 Magnetic Purification.

In this method ferromagnetic (catalytic) particles are mechanically removed from their graphitic shells[30]. The SWNTs suspension is mixed with inorganic nanoparticles (mainly ZrO_2) in anultrasonic bath to remove the ferromagnetic particles. Then, the particles are trapped with permanentmagnetic poles. After a subsequent chemical treatment, a high purity SWNT material will be obtained.This process does not require large equipment and enables the production of laboratory-sizedquantities of SWNTs containing no magnetic impurities.

4.4 Ultrasonication.

In this technique particles are separated due to ultrasonic vibrations. Agglomerates of different nanoparticles will be forced to vibrate and will become more dispersed [27]. The separation of the particles is highly dependent on the surfactant, solvent and reagent used [31]. The solvent influences the stability of the dispersed tubes in the system. In poor solvents the SWNTs are more stable if they are still attached to the metal. But in some solvents, such as alcohols, monodispersed particles are relatively stable [32].
When an acid is used, the purity of the SWNTs depends on the exposure time. When the tubes are exposed to the acid for a short time, only the metal dissolves, but on longer exposure, the tubes will also be chemically cut [33].

5. Functionalization.

Pristine, as-produced CNTs tend to bundle up and are insoluble in most types of solvents making it difficult to use them in biomedical applications. Moreover, some CNTs without any functionalization have been shown to exhibit toxicity. Therefore, to integrate CNTs into biological systems, CNTs need to be functionalized. Functionalization can make CNTs soluble and improve their biocompatibility properties [34]. Two types of Surface functionalization of carbon nanotubes are carried out: covalent and non-covalent.[35]

5.1 Covalent Functionalization.

Chemical reactions are carried out to form bonds with nanotube sidewalls in the process of covalent fictionalization. This functionalization strategy is through oxidation and carboxyl-based couplings. In this method, the tube cap openings are created and holes in the side walls are formed by an oxidation process in which strong acids are used. In addition to opening tube caps and creating side wall holes, the oxidation introduced to the caps and side walls with carboxylic groups enhances the solubility of CNTs in aqueous solutions. The carboxylic groups also allow for covalent couplings with other molecules through amide and ester bonds [36]

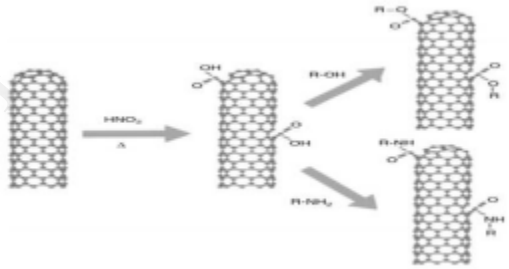

Figure 6: Functionalization of CNTs through oxidation followed by carboxyl-based couplings. The tubes are oxidized by a strong acid followed by the reaction of the carboxyl groups.[37]

Through this method, CNTs can be conjugated with various functional groups. Importantly, by bonding with suitable groups, CNTs can become soluble in aqueous or organic solvents. The presence of carboxylic groups on the sidewalls of CNTs reduces Van der Waals interactions between the tubes, enabling separation of nanotube bundles into individual, separated tubes.

5.2 Non-covalent functionalization of carbon nanotubes.

The advantage of non-covalent functionalization is that it does not destroy the conjugated system of the CNTs sidewalls, and therefore it does not affect the final structural properties of the material [38]. The non-covalent

functionalization is an alternative method for tuning the interfacial properties of nanotubes. The CNTs are functionalized non-covalently by aromatic compounds, surfactants, and polymers, employing π-π stacking or hydrophobic interactions for the most part. In these approaches, the non-covalent modifications of CNTs can do much to preserve their desired properties, while improving their solubilities quite remarkably. Functionalization can be carried out using surfactants and polymers. The distinct structural feature of a surfactant originates from its duality, namely the hydrophilic region of the molecule or the polar head group, and the hydrophobic region or the tail group that usually consists of one or few hydrocarbon chains [39]. Those amphiphilic molecules adsorb at the interface between immiscible bulk phase, such as oil and water, air and water or particles and solution, act to reduce the surface tension [40]. Two important features which characterize surfactants, namely adsorption at interface and self-accumulation into supramolecular structures, are advantageously used in processing stable colloidal dispersions.Amphiphilic polymers or soluble polymer are often used to solubilize CNTs. The main advantage of using polymers instead of small molecular surfactants is that the polymers reduce the entropic penalty of micelle formation.Also some conjugated polymers have significantly higher energy of interaction with nanotubes than small molecules with nanotubes. The main problem for polymers is that interactions with mechanically rigid SWNTs may force them into energetically unfavorable conformation. It has been suggested that to minimize strain in their conformations some polymers can wrap around nanotubes in a helical fashion [41].

6. Properties of Carbon Nanotubes.

The Electric and thermal properties are far more enhanced compared the convectional materials used. The heightened properties of the CNTs are due to formation of the network which takes advantage of the high aspect ratio between the length and diameter of the tube. The formation of networks also improves the mechanical properties within the matrices; the basic material for example has tensile strength that is 20 times that of steel.

Another significant property of CNTs is that they can perforate cell membranes easily and pass into the cellular components without causing apparent cellular damage. The long and narrow structure of the Carbon Nanotube makes them appear closely like a miniature needle so they can function as a needle at the cellular level. This property of CNTs is utilized by medical researchers by attaching molecules that are attracted by cancer cells to the nanotubes and delivering the drugs directly to the affected cells [42].

7. Carbon Nanotubes in Drug Delivery.

Drug delivery system is designed to improve the pharmacological and therapeutic profile of a drug molecule [43]. Carbon nanotubes are very dominant in today's world of medical research and are being highly researched in the fields of efficient drug delivery and tissue engineering methods. Pure CNTs are not soluble. It was the development of techniques to functionalize the molecules with organic molecules and make them soluble that paved way for therapeutic applications. Due to their high surface area, they are capable of adsorbing or conjugating with a wide variety of therapeutic molecules. Thus, CNTs can be surface engineered (i.e., functionalized) in order to enhance their dispersability in the aqueous phase or to provide the appropriate functional groups that can bind to the desired therapeutic material or the target tissue to elicit a therapeutic effect[44-47]

The functionalization also prevents the CNTs from being toxic and varying the function of the immune cells. CNTs might help the attached therapeutic molecule to penetrate through the target cell to treat disease. The large inner volume of tubes allows encapsulation of drugs which have both low as well as high molecules [48]. CNTs can also be used for multi-drug therapy by loading the tubes with more than one drug. In addition, CNTs can also act a controlled release system by releasing drugs over a long period of time.

8. Use of Carbon Nanotubes Cancer therapy.

Cancer is a group of diseases in which cells grow and divide abnormally. It is one of the primary diseases being looked at with regards to how it responds to CNT drug delivery. The conventional treatments for cancer include Radiation, Chemotherapy and surgery. These methods are successful in many cases, however these curative methods are painful, kill many healthy cells and produce adverse effects like toxicity in the patient's body. Metastasis is the spread of cancer from one organ or part to another non adjacent organ or part, is the main cause of cancer death [49]. CNTs are considered as antitumor agents and when used in the combination with conventional drugs, can significantly enhance their chemotherapeutic effect with the help of the advanced drug delivery system of

carbon nanotubes.Carbon Nanotubes and nanohorns have been tested *invitro* conditions for delivery of drugs. Functionalized, solubilized SWNTs can transport peptides, proteins, genes, and DNA[50-54]across cell membranes with little cytotoxicity [55-56]

However, evaluating the *in vivo* efficiency of CNTs loaded with anti-cancer drugs is critical. Several previous studies have demonstrated *in-vivo* targeting of tumors with carbon nanotubes in animal models, but with no drugs delivered to the target area [57-58]. Other problems faced in carbon nanotube drug delivery systems are lack of solubility, clumping occurrences, and half-life [59].Advancements are being made to the structure of CNTs to overcome these disadvantages.Because of the tube structure, carbon nanotubes can be fabricated with or without end caps. In the case of CNTs without end caps drug is held inside would be more accessible. Also, drug encapsulation has been shown to enhancewater dispersability, better bioavailability, andreduced toxicity in Single Walled Carbon Nanotubes. Encapsulation of moleculesalso provides a material storage application aswell as protection and controlled release ofloaded molecules over a long period of time.

9. Targeted cancer cell destruction.

Biological systems are highly transparent to 700- to 1,100-nm near-infrared (NIR) light [60]. The strong absorbance of SWCNTs in this special spectral window [61], an intrinsic property of SWCNTs, can be used for optical stimulation of nanotubes inside living cells to afford multifunctional nanotube biological transporters. For oligonucleotides transported inside cells by nanotubes, the NIR laser pulses trigger endosome rupture Trans locating the oligonucleotides into the cell nucleus.However, continuous NIR radiation can cause cell death because of excessive local heating of SWNTs *in vitro*. Selective cell destruction can be achieved by functionalization of SWNTs with a folate moiety, selective internalization of SWNTs inside cells labeled with folate receptor (FR) tumor markers, and NIR-triggered cell death, without harming receptor-free normal cells [62]. The intrinsic physical properties of SWNTs can thus be exploited to afford new types of biological transporters with useful functionalities in innovating new methods of drug delivery and cancer therapy.

One of the main disadvantages in using Carbon Nanotubes for drug delivery in the human body is the difficulty in controlling the breakdown of the nanotubes, which can cause unwanted toxicity and tissue damage. Recent studies show how CNTs can be broken down biologically into harmless components.

Carbon nanotubes were once considered bio persistent, meaning that they did not break down in body tissue or in nature. Research has shown that laboratory animals exposed to carbon nanotubes via inhalation or through injection into the abdominal cavity develop severe inflammation. This and the tissue changes (fibrosis) that exposure causes lead to impaired lung function and perhaps even to cancer. Alarming reports by other scientists suggested that carbon nanotubes are very similar to asbestos fibers, which are themselves bio persistent and which can cause lung cancer (mesothelioma) in humans a considerable time after exposure.

Certain studies conducted by a team of researchers at the Karolinska Institute, show that endogenous MPO can break down carbon nanotubes. This enzyme is expressed in certain types of white blood cell (neutrophils), which use it to neutralize harmful bacteria. Now, however, the researchers have found that the enzyme also works on carbon nanotubes, breaking them down into water and carbon dioxide. The researchers also showed that carbon nanotubes that have been broken down by MPO no longer give rise to inflammation in mice [62].

10. Conclusion.

Carbon nanotubes work beyond anticipations and their simple mechanism with long lasting life makes it more reliable to use. The use of Carbon Nanotubes in cancer treatment can guarantee up to 85% of the cure which other treatments cannot afford and having 100% site target with its body friendly nature adds to its advantage. The versatile properties of CNTs are the advantages which make them applicable in various fields. Organic functionalization has opened new horizons in the study of the biological properties of CNT. The biocompatibility of carbon cylinders has been determined. As pure CNT are highly toxic, and insoluble, the main factor was to verify the solubility of CNT in physiological media. Functionalized CNT possess higher tendency to cross the cell membranes, which is enabled by the needle like structure of CNTs. CNTs can be charged with biologically active moieties, which can then be delivered to the cell's nucleus. The chemistry of CNT offers the possibility of introducing more than one function on the same tube, so that targeting molecules, contrast agents, drugs, or reporter molecules can be used at the same time. The major problem involved in CNT application in drug

delivery is the lack of control over the degradation. However recent researchers have found that MPO (Myeloperoxidase), a particular enzyme expressed in certain type of white blood cells (neutrophils) which can breakdown Carbon Nanotubes into water and carbon dioxide thus declaring it as body friendly.

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