Review Article

Gold Nanoparticles in Early Detection and Treatment of Cancer: Biodistribution and Toxicities

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Accepted on: 19-03-2013; **Finalized on:** 31-05-2013.

ABSTRACT

Targeted nanoparticle delivery with drugs at the specified site has the potential to provide safer, effective and efficient therapies for cancer. The nanoparticlate assisted applications predominantly in assessing toxicities, early detection, diagnostics and therapeutics. Nanomedicine has the potential to increase the specificity for the treatment of cancer cells while leaving healthy cells intact through the use of novel nanoparticles. The detection and treatment of early tumor development is of major interest in order to evaluate the efficacy of therapeutic agents. Gold nanoparticles are being used effectively in laboratory based clinical diagnosis and *in vivo* characterization allows visualization and characterization of tumor development in early stages prior to manual palpation. Gold has been used in various occasions to assess the biodistribution, accumulation and cellular uptake of nanoparticles. The focus on the distribution of particles following exposure is evident within the available literature, which can be useful in assessing appropriate in vitro and in vivo toxicity of nanoparticles. In general, the availability of *in vivo* and *in vitro* information allows to detect the tumor at an early stages. The importance of controlling the size and shape of gold nanoparticles used to minimize any potential toxic side effects is also discussed in this paper. This review focused on recent achievements in utilizing gold nanoparticles for early detection and therapeutic activity of cancer and also gives a detailed analysis of *in vitro* and *in vivo* studies on biodistribution and toxicity following the exposure of gold nanoparticles.

Keywords: Gold nanoparticles, Cancer, early diagnosis, therapeutic, biodistribution, toxicity.

INTRODUCTION

Tumor diagnosis, treatment and prevention of cancer in early stages are of great importance because high death rate and the frequency of reoccurrence of the disease even after treatment. Hence, there is a need of technologies applicable for the detection of cancer at the initial stages. It has always been one of the most important issues in diagnosis[1]. Current cancer treatment procedures consist of compounds that are non-specific and highly toxic[2]. Also, the inability of conventional diagnosis tools to detect cancer in early stages.

Nanotechnology might have a deep impact in solving many of the problems associated with conventional anticancer drugs. The nano-formulated drugs can be made as relatively safe, nontoxic and conventionally injectable formulations[3]. Nanotechnology not only has the potential to conjugate the required targeting moiety, but also has the ability to carry the moiety for site-specific delivery without compromising its activity[4]. The application of nanotechnology is rapidly progressing, and has tremendous potential to make a revolutionary impact in healthcare with profound effects on current treatment paradigm for various diseases[5]. Early detection will greatly increase survival rates with the reasonable assumption that an in situ tumor will be easier to eradicate [6-8].

Nanoparticles potentially have better access to tumor sites as compared to conventional drug delivery carriers[9].

One of the most extensively used nanoparticles for cancer treatment is the gold nanoparticle and the domain in which nanoparticles used in medicine is called as nanomedicine[10,11]. The use of nanoparticles in medicine improves the resolution of tumor area comparing with the nearby normal tissue (Figure 1). It is important to understand the hazards, potential and accessing toxicities for the exposure of nanomaterial on the delivery site. This review has focused on the in vivo and in vitro detection of cancer, biodistribution and the toxicity of gold particles.

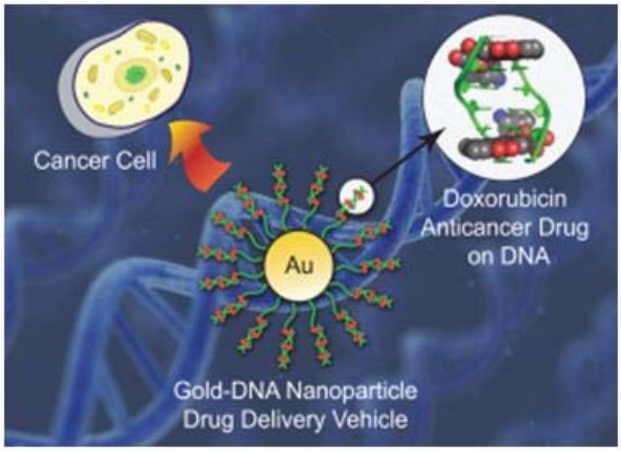

Figure 1 Gold Nanoparticles use DNA to Deliver Doxorubicin, Improving Cancer Treatment (Courtesy: Syracuse University)

Emphasis is placed on surface area, particle sizes, animal and cell model, route of administration, early detection and treatment of cancer, assays of evaluating gold

particle toxicity. This is particularly useful when assessing cell targets and evaluating particle toxicity. It is most necessary to reveal the correlations between the in vitro and in vivo findings. The primary focus of this review is to discuss the key challenge in early diagnosis and treatment of cancer using gold nanoparticles and also to analyze the biodistribution and cytotoxicity of AuNPs.

Early detection and treatment of cancer

Gold nanoparticles (AuNPs) are currently playing a significant role in the field of clinical diagnosis and treatment of cancer as well as several biomedical applications. AuNPs offer significant targeting properties at cancerous cells, leaving healthy cells without doing any harm[12]. In recent years, especially in cancer research, the light emission and scattering properties of gold nanoparticles have been studied extensively and used it for locating the tumor site and facilitated the targeted delivery at the tumor site (Figure 2). Nanoparticle surface increase its specificity to bind with the cancer- cell surface, which has advantages for both therapy and imaging.

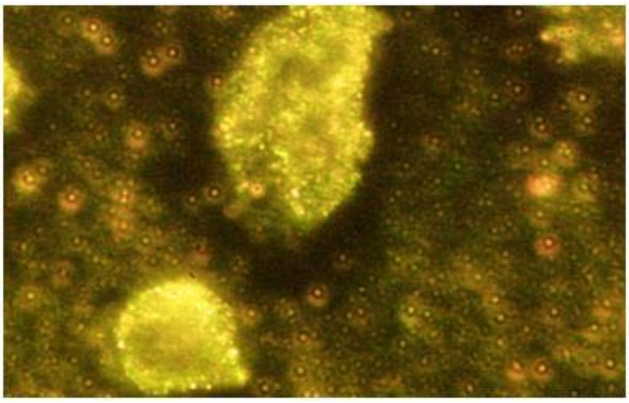

Figure 2: Gold nanoparticles accumulation at the tumor site and could treat prostate cancer with fewer side effects than chemotherapy (Courtesy: Georgia institute of Technology).

AuNps *in vivo* and *in vitro* characterization allows visualization and characterization of tumor development in early stages prior to manual palpation (Table 1). Nanotechnology based cancer treatment approaches potentially provide the earliest detection and localized targeted therapies (Figure 3). This reduces side effects and improves patient's quality of life.

The gold nanoparticle biomarkers and optical contrast agent provide excellent signal from cancer tissues and it is used to detect them from complex environment. AuNPs make them most promising probes for cancer detection[13,14]. Thus, gold nanoparticles based imaging modalities have made a significant entry in to a cancer research as a highly sensitive probe for cancer detection[15]. The new era of molecular imaging, cellular and molecular level aims at measurement of biological processes and quantifying molecular changes. Recently, there is a lot of study going on to design novel gold nanoparticle that is useful in diagnosing cancer at its earliest stages and pinpointing its location within the human body[16]. AuNPs act as good promising probes to detect the cancer cells. Cancer cell line of SiHa cells, over expressing the transmembrane glycoprotein, epithelial growth factor receptor (EGFR) can be imaged by immunotargeted AuNPs in case of pericervical cancer[17]. Cho et al.,[18] investigated the 19 nm Iron gold (Fe-Au) NPs prepared by reverse micelle method. This nanomaterial act as a good magnetic resonance contrast agent. The absorbance and scattering properties of AuNPs can be change in accordance with their size parameter[19-22].

The large AuNPs show relatively high-scattering properties and it is mostly applicable for biomedical applications, whereas those having relatively high-absorption properties are widely used in colorimetric detection and biological analysis by changing refractive index of AuNP's environment[23-25]. AuNPs conjugating with anti-EGFR antibody, it gave the ability to distinguish between cancer and noncancer cells from the strong scattering images by observing the scattering light on a dark field microscope. EL- Sayed et al.,[26] reported the absorption and surface Plasmon resonance scattering properties of anti- EGFR antibody conjugated AuNPs are useful for the detection of oral cancer.

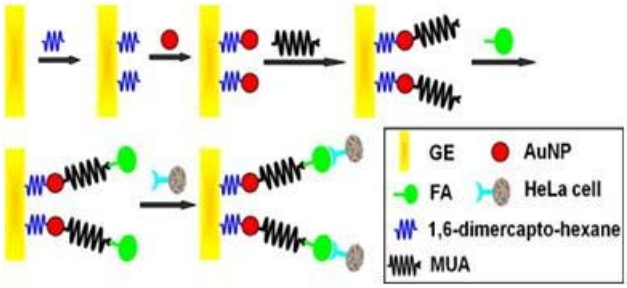

Figure 3: Highly sensitive detection of cancer cells by electrochemical impedance spectroscopy[13]

Gold nanoparticles have a unique physiochemical properties including Surface Plasmon Resonance (SPR) and able to bind amine and thiol groups that allow surface modification for biomedical application[27]. Nanoparticle functionalisation is the subject of interest with newer application and progress. This is towards the development of multifunctional and biocompatible particles for use in cancer diagnosis and treatment[28-31]. Gold nanoparticles are especially attractive for imaging and therapeutic activity due to their SPR, adsorption and enhanced light scattering properties. In clinical diagnostic gold nanoparticles are used as a template agent in place of Gadolinium (Gd) or iodinated solution as a chelating agent for use as MRI contrast agents[32,33]. Reuveni et al.,[34] demonstrated that a small tumor, which is currently undetectable through anatomical computed tomography, which is clearly visible and enhanced the visibility by the molecularly-targeted gold nanoparticles. Thus, the noninvasive and nonionizing molecular cancer imaging tools facilitate early cancer detection in vivo.

Table 1: Summary of early detection of cancer and therapeutic activity of Gold nanoparticles

Reference	Kind of study & animal used	Type of AuNps	Cell lines used	Particle Size (nm)	Outcome of studies
Reuveni et al[34]	In vivo, Nude mice	AuNps	Human squamous cell carcinoma head & neck cancer	30	Capable of producing contrast enhancement facilitate early cancer detection based on molecular markers rather than anatomical structures
Hainfeld et al[73]	In vivo, mice	AuNps	SCC VII (Murine squamous cell carcinoma)	1.9	Significant increase in apoptosis
Change et al[74]	In vivo, mice	Citrate AuNps	B16F10 (Melanoma cell line)	13	Retarded tumor growth,prolonged survival, increase of apoptotic signals detected inside tumors
Popovtzer et al[75]	In vitro	AuNR - UM-A9 antibodies	Squamous cell carcinoma (SCC) Head and neck cancer	45 X15	Inducing distinct contrast in CT imaging. Enhance imaging and early diagnosing property.
Kim et al[37]	In vivo, Hamster cheek pouch model	PEG-AuNps	Oral cancer	71	Enhanced imaging quality- early diagnosis of cancer
Chen et al[38]	Invivo, Mice	Gold nanocages	Bilateral tumor model U87wtEGFR	15	Inhibit the progression of ovarian cancer and tumor growth

Targeted AuNps are now being developed to improve imaging character with MRI and CT scan. Gobin et al.,[35] indicated that gold nanoshells were intravenously administered in to mice and nanoshells were shown to selectively accumulate in tumor site. Signal intensity in tumor tissue is enhanced by 56% and also act as a good contrast agent for tumor tissue compared to normal tissue. Visaria et al[36] reported that 60 % survival was observed in the group treated with free TNF-α alone, this is a sign of systemic toxicity. For the group treated with free TNF-α and hyperthermia, there was redness, inflammation, on the tumor bearing part. Over days tumor became worse and causing the animal to limp. After the injection of Gold nanoparticle, the 100% animal survival was observed. This is concluded that this may be safe to use against tumor. AuNPs are promising in vivo contrast agents. Kim et al.,[37] reported that Syrian hamster (Mesocricetus auratus) was treated topically with 0.5% (v/v) 9, 10-dimethyl- 1, 2-benzanthracene for five months to induce cancer. 200 µl of the anti-EGFR antibody conjugated PEGlated AuNP solution was applied to the hamster's cheek pouch for 10 min by dropping it directly in to the 1-cm- diameter aperture of the ring shaped clamp. In this study concluded that this delivery of antibody conjugated PEGlated gold nanoparticles enhanced images of oral dysplasia in a hamster model.

Chen et al.,[38] studied ablate murine models with subcutaneous glioblastoma in vivo using gold nanoparticles. The tumor metabolism had declined 70% after 14 hrs, the mice were treated with gold nanocages with near infrared laser therapy. Compared to normal cells, El- Sayed et al demonstrated that oral cancerous cells targeted with gold colloidal nanospheres were destroyed with 2-3 times lower laser power[39]. Laser power needed to kill the cancer cells was approximately 20 times less than that needed to destroy normal cells. The efficient energy conversion process from photo energy to local heat energy is involved in photo thermal cancer therapy[40]. Our data suggest that this review could be used to identify multifunctional tumor-targeting gold nanoparticles that can serve as imaging agents to detect cancers or monitor clinical response, as well as to specifically deliver therapeutic agents to the tumor. Breast cancer is a leading cause of mortality among women and there is a need for improved methods for early detection, new therapies that are more effective without side effects. AuNPs modified with PEG and conjugated with monoclonal antibody (MAb) and Herceptin (HER) enhance recognition of breast cancer cells.

Biodistribution of gold nanoparticles

Nanotechnology might have a deep impact in solving many of the problems associated with anticancer drugs because nanoformulated drugs can be made as relatively safe formulations. One of the major strengths of a nanomedicine approach is the ability to alter the pharmacokinetics and biodistribution of the drug. Different routes of administration may result in varying effects on the biodistribution pattern of drug carriers[41,42]. Many researchers believe that nanoparticles based technique could prove to be more effective than current chemical or radiation based treatments, with fewer adverse side effects. The distribution pattern of NPs on tissue appears to be dependent on the size of the particles and exposure route.

AuNPs have also been used on a number of occasions to evaluate its distribution and cellular uptake of particles. Previous studies highlighted that the gold nanoparticles can potentially distributed on the exposure site based on ingestion, inhalation and dermal penetration. All the parameters are necessary to consider whether the

particles are able to pass from exposure site in to blood and become distributed within the body. In addition, the number of investigation has been focused on understanding the biokinetics and distribution of gold nanoparticles following exposure from the variety of routes. Toxicological studies suggest that nanoparticles may cause adverse health effects, but the fundamental cause-effect relationships are ill defined. This review also focused on assessing the in vivo distribution and accumulation of gold nanoparticles (Table 2) following exposure and also provide useful information regarding toxicity of gold nanoparticle *in vitro* (Table 3).

Gold sulfide (GS) nanoparticles are used against xenografted tumor mouse model. This suggests that the longer circulation time of the GS nanoparticles caused maximal accumulation at tumor site with a optimal tumor accumulation time of 24 hrs[43]. Notably the nanoparticles are rapidly eliminated from the blood and simultaneously accumulate in the reticuloendothelial system and renal tissues. It is possible that during the entire observation period nanoparticles were predominantly excreted by the kidneys. Many studies have been reported that gold nanoparticles enhance the toxicity of radiation on cancer cells and decrease side effect. Kong et al indicated that Glu-GNPs enhance the radiation sensitivity in cancer cells but not in nonmalignant cells, though the cell lines MCF-7 and MCF-10A uptake the same level of Glu-GNPs[44]. EMT6 murine breast tumor in mice was targeted with gold nanoparticle, and then the mice were imaged with 22-kVp mammography. Eventhough there is a high initial blood concentrations of AuNps (10 mg ml^{-1} blood), there were no hematological or biochemical abnormalities were detected at 11 or 30 days post-injection[45].

Size dependent distribution of gold nanoparticles

Nanoparticle biodistribution is influenced by the size and surface characteristics. The addition of PEG increases the hydrodynamic particle size, which prevents filtration by excretory organs. This ultimately increases the circulation time of the particles. After intravenous administration of 20 nm PEG- coated gold colloidal nanosphers had slower clearance, fewer uptakes by RES cells, and higher accumulation in the tumor compared to 40 and 80 nm particles. The 80 nm particles were found to have lower PEG density than the 40 nm particles[46]. Perrault et al., evaluated that the increased blood half life by the addition of PEG was more prominent in smaller versus larger total diameter particles[47]. Jiang et al reported that 20-50 nm particles have more effective PEG coverage and consequently longer blood half-lives, and they are also in the optimal size range for cellular uptake[48].

Etame et al.,[49] found that the permeation of PEGylated AuNps was size dependent. Short PEG chain length in combination with smallest core size led to optimum permeation of a blood-brain barrier model system. This finding may pave the way to optimized therapy of malignant brain tumors. Nance et al demonstrated that a dense poly(ethylene Glycol) coating improves the penetration of large polymeric nanoparticles within brain tissue (Figure 4). Semmler-Behnke et al.,[50] reported that the intravenous administration of AuNPs within 24 hrs, were completely removed from the blood and were preferentially accumulated in the liver and spleen. But 14 nm nanoparticles were remained within the blood at 24 hrs and small level of nanoparticles was accumulated in the liver and spleen. In this study, the nano particles size influences the behavior of particles within the body. Smaller particles below 60 nm have more successfully dispersing throughout the tumor. Thus, tumor accumulation and distribution also depends on the particle size and time. Gold nanoparticles (15, 50, 100 and 200 nm) were intravenously administered in to mice and evaluated the distribution of those particles in tissue. After 24 hrs the highest accumulation was observed in lung, kidney and spleen. 15 nm and 50 nm NPs were observed in brain, indicating that these NPs have the potential to cross the blood-brain barrier. Sonavane et al.,[51] investigated that the size of NPs are responsible for the extent of particle distribution.

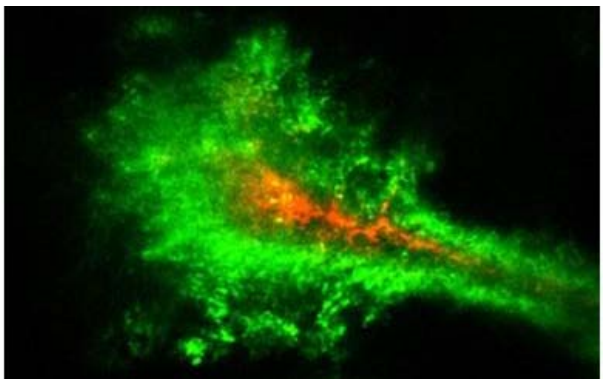

Figure 4: A Dense Poly(Ethylene Glycol) Coating Improves Penetration of Large Polymeric Nanoparticles Within Brain Tissue[68]

Among from various studies indicated that size of the particles mostly responsible for the distribution of nanoparticles following intravenous exposure. In contrast, small particles are likely to avoid recognition and as a consequence become more widely distributed within the body. The distribution of nanoparticles in the organism is known to depend on multiple factors, including the size and coating. Therefore, an extended study with the use of different size with various characteristics is needed to explain this phenomenon.

Accumulation of Gold nanopaticles

AuNps have high-level accumulation in the liver and spleen, and these accumulations can induce the gene changes and liver necrosis[52,53]. James et al.[54] studied gold-silica nanoshells (13 nm) were intravenously administred to mice and noted a peak accumulation of the nanoshells at 24 hrs. De Jong et al. reported that different size nanoparticles (10, 50, 100 and 250 nm) are accumulated in the liver and suggested that this was caused by the high perfusion of blood through this organ[55]. The large size of nanoparticles and the shape contribute to

difficulties in tumor delivery. Cho et al. administered AuNps stabilized with polyethylene glycol in mice through intravenous injection[56]. The distribution of those particles was recorded from 5 mins to 7 days post injection and primarily observed in liver and spleen. An increased level of apoptosis was also observed within the liver. The primary NPs accumulations have been consistently demonstrated within the liver and spleen following intravenous administration. The gold nanoparticles with small size can penetrate kidney tissue and have promise to decrease in vivo toxicity by renal clearance.

Methods of nanoparticle delivery to target organs and tissues are currently being actively developed. Zagainova et al, shown that the AuNPs were used to produce neither acute nor long term pathomorphologic alteration in the organs of healthy mice. The biodistribution and the effects of AuNps in different organs and tissues of animals after intravenous administration were investigated using transmission electron microscopy. A significant number of nanoparticles were detected in blood and urine 4 hrs after administration. Gold nanoparticles are shown to accumulate primarily in the spleen and kidneys with a peak of accumulation being reached on day 7 after injection. Single nanoparticles are detected in the brain, heart and liver. Examining the nanoparticles in living animals following intravenous administration, nanoparticles removed from the circulation by macrophages in the liver and spleen with only a mild acute inflammatory response and an increase in oxidative stress in the liver[57].

Recent researchers are interested in modifying existing drugs to improve pharmacokinetics properties and reducing non-specific side effects and enabling higher dose delivery to target tissues (Figure 5). James et al. and Torentyuk et al, reported that only marginal accumulation in the kidney[58]. In Passive tumor targeting, the leaky nature of the tumor vasculature, allowing AuNps to reach the tumor, and are more rapidly cleared from the tumor and accumulated in the kidney, liver, and spleen[59,60].

Cytotoxicity effect of gold nanoparticles

In vitro cytotoxicity assays are a simple way to evaluate the basic toxicity of a material. The interaction of nanoparticles with biological systems including living cells has become one of the most urgent areas of collaborative research in material science and biology. Recent studies have suggested that nanoparticle toxicity and pharmacokinetics can depends on the size, shape and surface charge of particles as well as surface modifications (Table 3).

Nanoparticles are of similar size to typical cellular components and proteins and thus may bypass natural mechanical barriers, possibly leading to adverse tissue reaction[61]. Tunable nanoparticles are the cytotoxic agent that preferably promotes apoptosis or secondary necrosis depending predominantly on their size. This is particularly useful in assessing relevant tissue and cell targets that should be useful for evaluating particle toxicity in vitro[62].

This section discusses the current understanding of in vitro studies of gold nanoparticle toxicity.

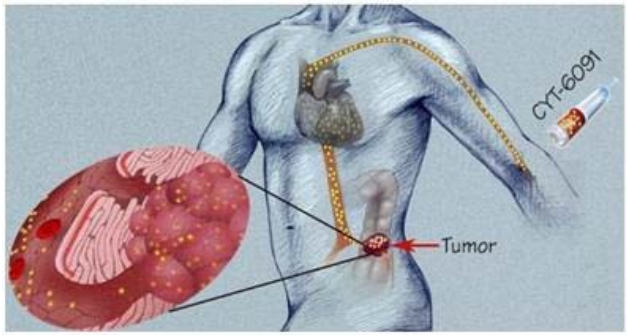

Figure 5: Gold particles could deliver cancer drugs (Courtesy: CytImmune)

Gold nanoparticles are naturally taken up by many cell types and do not cause any detectable cytotoxicity. This fact has been extensively exploited for delivery of DNA inside cells. Antisense oligonucleotide conjugated AuNps has been shown to be spontaneously taken up by cells and bring down the expression level of target genes[63]. Of the various gold nanoparticles discussed, gold colloidal nanopheres have induced little toxicity in vitro, with an approximate 15% reduction in cell viability observed for the concentration of 200 mg L^{-1} after 24 hrs[64,65]. Gold nanoparticles are typically less toxic than their metal precursors, with over 90% cell death reported after exposure to 250µm of gold solution[66]. The cytotoxicity of gold nanoparticles depended primarily on their size and not on ligand chemistry. Pan et al., reported that 15 nm size nanoparticles are nontoxic up to 60 and 100 fold higher concentrations. The cellular response nanoparticle is size dependent, in that 1.4 nm particles cause predominantly rapid cell death by necrosis within 12 hrs while 1.2 nm in diameter effect predominantly programmed cell death by apoptosis.

Several groups have examined the cellular uptake and cellular toxicity of gold nanoparticles. This approach concluded that the gold proposes a novel platform therapy with minimal toxicity and increased efficacy profiles for the destruction of hepatic cancer cells. Tomuleasa et al.,[67] evaluated the in vitro antitumor efficacy of gold nanoparticles conjugated with conventional chemotherapy drugs for the treatment of liver cancer. In the presence of the anti-cancer drugs the cellular proliferation rates are very low by the AuNps than those of cells exposed to the cytostatic drugs alone. This study indicating and facilitated that AuNps increased susceptibility of cancer cells.

Cytotoxicity is also depends on the type of cells used. CTAB capped goldnanorods were found to be cytotoxic, as judged by the MTT assay with a different cell line, HeLa cells[69]. The nanocytotoxicity was able to reduce by over coating the nanorods with polyethylene glycol (PEG), which is well known to reduce nonspecific binding of biological molecules to surfaces. Huff et al[70] reported that phosphatidylcholine is also a coating molecule that reduces the cytotoxicity of CTAB-coated gold nanorods.

Table 2: Summary of *In vivo* distribution and accumulation of gold nanoparticles

Reference	Size (nm)	Exposure route	Animal	Accumulation	Conclusion
De Jong et al[55]	10,50, 100,250	Intravenous	Rat	Most nanoparticles found in liver and spleen	No toxicity and wider organ distribution for small particles
Semmler Behnke et al[50]	4 and 18	Intratracheal	Rat	Liver, spleen, kidneys, skin	Translocation of 4 nm NPs. No toxicity and wider organ distribution for small particles.
Sonavane et al[51]	15, 50, 100,200	Intravenous	Mice	Liver, lungs, kidney, spleen and brain.	No toxicity and wider organ distribution for small particles.
Sonavane et al[76]	15, 102, 198	Intravenous	Rat	Skin and Intestine	Able to permeate through excised skin and intestine. No toxicity and wider organ distribution for small particles
Cho et al[56]	13	Intravenous	Mice	Primary sites of accumulation in the liver and spleen	Nanoparticles induced inflammation and apoptosis in the liver tissue.

Table 3: Summary of *In vitro* cytotoxicity studies on Gold nanoparticles

Reference	Size (nm)	Cell line	Toxicity	Effect
Lowery et al[77]	3 nm	SK-BR-3 breast carcinoma cells	Selectively induce cell death.	Increased localization of nanoshells within the tumour. Tumour specific antibody increases the specificity of the nanoshell therapy.
Zhang et al[46]	30	DU145	13.5% Loss of cell proliferation.	Enhanced radiation sensitivity
Kong et al[44]	10.8	MCF7 and $MCF10_a$	5% Loss of cell proliferation.	Enhanced radiation cytotoxicity
Rahman et al[71]	1.9	BAEC	30% loss of cell proliferation	Greatest radiosensitization was observed.
Liu et al[78]	6.1	CT-26 EMT-6	Low cytotoxic activity.	Targeting ability to liver cancer cells BEL-7404 and BEL-7402 while not to the normal healthy liver cell HL-7702
Takahashi et al[79]	6×11	Human dermal fibroblast	Reduced toxicity.	Replacing CTAB with PEG on the surface of nanorod reduced the toxicity
Pernodet et al[80]	13.1	HeLa	Loss of cell proliferations at 24-144 hrs	Nanoparticles decreased cell proliferation rate, adhesion and motility
Patra et al[81]	33	BHK21, HEP2G, A549	Nanoparticles are not toxic to HEP2G and BHK21 at 2hrs but toxic to A549 at 36 hrs	Non cytotoxic to baby hamster kidney and human hepatocellular liver carcinoma cells. Cytotoxic to human carcinoma lung cell line.
Khan et al[82]	18	HeLa	Nanoparticles are not toxic at 3-6 hrs	Nanoparticles did not change gene expression patterns.
Pan et al[61]	0.8, 1.2, 1.4,1.8 and 15	HeLa, SK-Mel-28, L929 J774A1	15 nm particles is nontoxic, 1.4 nm nanoparticles are most toxic at 72 hrs.	Toxicity is not dependent on cell lines but size dependent.
Hauck et al[83]	40×18	HeLa	Polyelectrolyte coating of nanorods are not toxic compared to the CTAB-Capped nanorods	No gene expression abnormalities were observed.
Villiers et al[84]	10	Dendritic cells from C57BL/6	Nanoparticles were not toxic	Did not induce dendritic cell activation
Pan et al[85]	1.4 and 1.5	HeLa	The 1.4 nm nanoparticles induced necrosis by oxidative stress. 15 nm particles were found to be not toxic	GSH- capped nanoparticles were less toxic than TPMS capped nanoparticels at 48hrs.
Gu et al[86]	3.7	HeLa	Nanoprtivles are not toxic within the 6-72 hrs at the dose of 0.08-100 μM	Nanoparticles entered nucleus and did not induce toxicity.
Alkilany et al[87]	65×15 nm	HT-29 Human colon carcinoma cell	Over coating the CTAB Capped nanorods with either negatively or positively charged polymer substantially reduces cytotoxicity and affect their uptake	Physiochemical surface properties of nanomaterials change substantially after coming into contact with biological media
Niidome et al[69]	65±5×11±1	HeLa Cells	80% cell death with 0.05 Mm CTAM coated nanorods. Only 10% cell death at 0.5 mM for PEG-coated nanorods	PEG-modified gold nanoparticles had less cytotoxicity compared to CTAM coated nanorods.

In Vitro studies demonstrated that 5nm AuNps in mice bearing MC7-L1 murine breast cancer cells showed cytotoxicity with a 55% less in colony formation in the absence of radiation. Also it was found that no radio sensitization at concentrations up to 5 mM. Rahman et al., reported that a significant decrease in cell viability was observed when cells were incubated with 0.125 mM AuNPs. This effect increases with increase of AuNPs concentration 0.25 mM and beyond. The large numbers of *in vitro* studies have indicated that gold nanoparticles have no acute or sub acute toxicity on cells,[71] when used as nanomedicine[72]. Experimental studies were performed to determine cell toxicity to various AuNP concentrations. It is necessary to investigate the internalization of particles by cells.

CONCLUSION

Tumor targeted drug delivery nanoparticles systems must address technical and biological concerns that influence their distribution. More extensive investigation is required to understand the bio distribution and fate of gold nanoparticles after the exposure. It has been found that normally biodegradable substances are decomposed and their waste products are excreted by the kidneys and intestines. However, non-biodegradable nanoparticles have been studied and it seems that they accumulate in certain organs, especially to the liver. It is clear that AuNPs offer various advantages over bulky structures and the characteristic properties of gold nanoparticles make them ideal for diagnostic purpose and several biomedical applications. Though many of the technologies involving nanoparticles for cancer detection and treatment are mainly in preclinical stages, there is tremendous potential for nanotechnology to enable desperately needed for cancer detection in its early stages.

Acknowledgement: This work was funded by the Tamilnadu State Council for Science and Technology (TNSCST), Government of Tamilnadu, India. The authors acknowledge the management and Dr. R. Joseph Xavier, Principal, Sri Ramakrishna Institute of Technology, Coimbatore for providing facilities and support to execute the research work. The authors declare that they have no competing financial interests.

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Source of Support: Nil, **Conflict of Interest:** None.

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