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Review Article

MICROSPHERES: NOVEL APPROACH FOR CANCER TARGETING

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Abstract

Microspheres are characteristically free flowing powders consisting of proteins or synthetic polymers which are biodegradable in nature and ideally having a particle size less than 200 μm. A well designed controlled drug delivery system can overcome some of the problems of conventional therapy and enhance the therapeutic efficacy of a given drug. There are various approaches in delivering a therapeutic substance to the target site in a sustained controlled release fashion. One such approach is using microspheres as carriers for drugs. Microspheres received much attention not only for prolonged release, but also for targeting of anticancer drugs to the tumour. A major disadvantage of anticancer drugs is their lack of selectivity for tumor tissue alone, which causes severe side effects and results in low cure rates. Thus, it is very difficult to target abnormal cells by the conventional method of the drug delivery system. Microsphere technology is probably the only method that can be used for site-specific action, without causing significant side effects on normal cells. Cancer microsphere technology is the latest trend in cancer therapy. It helps to formulate the product with maximum therapeutic value and minimum side effects. Microspheres show a promise in cancer therapy by selectively gaining access to tumours due to small size and modifiability and also targeting drugs by physical trapping in blood vessels and sustain the action of a therapeutic agent through controlled release with enhanced permeability and retention effect.

Keywords: Microspheres, Cancer, TheraSpheres, SIR-Spheres.

Introduction

Cancer is a leading cause of death worldwide. It is a complex and critical disease, occurring as a result of progressive accumulation of genetic and epigenetic changes. The most important defining feature of cancer is the

rapid growth of abnormal cells, which can invade adjoining parts of the body and spreads to other organs. Since the previous decade, the fast growing research on therapeutics has shown promising possibilities for achieving the dream of every oncologist. Despite outstanding recent advancement in the therapeutic cock-tail, significant challenges still remain present in the field of `cancer therapeutics. Unfortunately, commonly used cancer chemotherapy have presented unsatisfactory results, as the therapy is deleterious to patient health by making patients more susceptible to other diseases and often cause death by weakening the immune system of the patient body.

Targeted delivery of anticancer drugs is one of the most actively pursued goals in anticancer chemotherapy. A major disadvantage of systemic anticancer drugs is their lack of selectivity for tumor tissue, which causes severe side effects and results in low cure rates. Any strategy by which a cytotoxic drug is targeted to the tumor, thus increasing the therapeutic index of the drug, is a way of improving cancer therapy and minimizing systematic toxicity. Modern medicine is successful in achieving disease-free survival in a good number of cancer patients. However, in a majority of cases, medical intervention is only successful in prolonging the life of a patient from a few months to a few years. Cancer is essentially a pathology with various mechanisms at its disposal to avert its own destruction. Thus, multimodal therapy is required, with or without surgical intervention. Novel therapies are constantly being discovered, and improved, with neoplasia targeting, and given high priority[1, 24].

Major challenges in cancer chemotherapy are related to toxicity on healthy proliferating cells and multi-drug resistance (MDR) against anticancer agents. The life threatening side-effects caused by nonspecific tissue distribution of the anticancer agents have restricted the systemic high dose strategy. Cancer cells except those having intrinsic resistance are sensitive to chemotherapy in the beginning; but, often develop acquired resistance upon repeated chemotherapy cycles. The resistance initiated by an anticancer agent extends cross-resistance to a wide range of drugs having different chemical structures and cellular targets. Once the resistance develops, systemic high dose administration of anticancer agents becomes ineffective and the resistance is further stimulated. Thus, there is a crisis in fighting against cancer. The effectiveness of a cancer therapy is understood by its ability to reduce and eliminate tumors without damaging healthy tissues. Therefore, a distinct capacity to target tumors with limited effect on healthy tissues is the most essential for the success of cancer therapy and the ultimate goal of it is

to maximize survival period and quality of life of cancer patient. These facts put the accent on the need for new generation of more effective and safe therapies for the treatment of various cancers.

In the U.S., acording to the National Cancer Institute in 2010, the most common cancers (excluding non-melanoma skin cancers) are listed below in table no.1.

Targeting of drugs to special cells and tissues of the body without their becoming a part of systemic circulation is a very novel idea[6].

Table no.1- According to the National Cancer Institute in 2010, the most common cancers in US (excluding non-melanoma skin cancers).

Cancer type	Estimated new cases	Estimated deaths
Bladder	70,530	14,680
Breast (female-male)	207,090-1,970	39,840-390
Colon and rectal (combined)	142,570	51,370
Endometrial	43,470	7,950
Kidney (renal cell)	53,581	11,997
Leukemia	43,050	21,840
Lung(including bronchus)	222,520	157,300
Melanoma	68,130	8,700
Non-Hodgkin lymphoma	65,540	20,210
Pancreatic	43,140	36,800
Prostate	217,730	32,050
Thyroid	44,670	1,690

In traditional drug delivery systems such as oral ingestion or intravascular injection, the medication is distributed throughout the body through the systemic blood circulation. For most therapeutic agents, only a small portion of the medication reaches the organ to be affected. Targeted drug delivery seeks to concentrate the medication in the tissues of interest while reducing the relative concentration of the medication in the remaining tissues. This improves efficacy while reducing side effects.

For all forms of therapies, a common thread is the need for targeting to avoid side-effects of drugs. In the past few years, microspheres have been shown to be selective for tumor vascular endothelial cells. In addition, a handful of articles highlight the ability of targeting these vesicles to tumors in various parts of the body, by using advanced **Microsphere** drug delivery systems[6].

Microspheres

They are the colloidal drug delivery system. Microspheres are characteristically free flowing powders consisting of proteins / synthetic polymers that are biodegradable and ideally have a particle size less than 200 μm.

Biodegradable microspheres can be utilized to direct drugs to organ(s) by lodging them in the end organ vessels. Its success depends on the size of the microsphere used and on the mode of administration (intravenous / intra-arterial) [2, 4,6].

Advantages:

The use of microspheres as a drug-delivery system has certain advantages [2, 4, 6, 7]:

- augmented effectiveness
- reduced toxicity of the incorporated agents to non-targeted cells and tissues
- Microspheres provide constant and prolonged therapeutic effect,which will reduce the dosing frequency and thereby improve the patient compliance.
- They could be injected in to the body due to the spherical shape and smaller size.

Disadvantages:

- microspheres are denatured within several weeks
- relatively unstable
- not easily mass-produced

Microspheres in cancer therapy

Liver cancer

Microspheres are used as a bridge to surgery or transplantation or used in radiation treatment against liver cancer. The development of liver metastases from any solid malignancy heralds a poor prognosis, unless the disease is amenable to surgical resection. For patients diagnosed with colorectal carcinoma, the majority of deaths are attributable to hepatic metastasis.

The cytoreductive therapies in cancer treatment can be broadly categorized as those applied via the transcapsular or trans-vascular routes. The myriad of therapies that exploit the trans-arterial route are based on the premise that metastatic tumors receive their blood supply from the arterial rather than the portal circulation, unlike normal hepatocytes. Hepatic artery injection allows preferential delivery of the material to the peritumoral vascular plexus. A suspension of particles injected via the hepatic artery, such as microspheres of an appropriate diameter, will preferentially lodge in the peritumoral vessels, a process termed as embolization. Radiation can destroy the tumor if sufficient tumor doses can be delivered selectively without damaging the adjacent normal tissue in the process. Brachytherapy, wherein a radioactive material is placed directly inside or next to the tumor, circumvents the limitation of non-selectivity of extracorporeal radiotherapy. The utilization of this effective technology, however, is largely limited, by the frequent requirement of direct visualization of the liver that is traditionally achieved intraoperatively and is technically prohibitive in the presence of multifocal disease. From this discussion, it is evident that the altered arterial supply to hepatic tumors can potentially be exploited to deliver lethal doses of radiation. A high energy radiation source, combined with an appropriately sized trans-hepatic arterial administered embolic microscopic particle, would allow radiation to be delivered preferentially to the tumor. A β-emitter, such as yttrium-90, would create a zone of high radiation exposure confined to the vicinity of the tumor, while maintaining a non-tumorous hepatic parenchymal exposure to tolerable levels. This forms the premise for radioembolization. Millions of microspheres, measuring about 30 μ in diameter, incorporating yttrium-90, are injected via a hepatic arterial catheter to the arterial supply of the tumor. Radioemobilization is a technique that allows high average doses of radiation (200 to 300 Gy) to be given to liver tumors with minimal serious effect on the non-tumorous liver. The dose determination for glass microspheres is based on a nominal average target dose (150 Gy/kg), and the

patient's liver mass is determined from the CT data and assumes the uniform distribution of the microsphere throughout the liver volume to be:

$$A(GBq)glass=D(Gy)\times M(kg)/50$$

In this equation, A is the activity, D is the nominal target dose, and M is the mass of the targeted liver tissue. Resin microspheres are received in a vial as a 3 GBq dose, one of the two methods for activity determination; the Body Surface Area method and the Empiric Method are used [8,9,11,21].

Yttrium-90 microspheres (SIR-Spheres and TheraSpheres)

The microspheres are typically made of glass or polymers (resin) and contain yttrium-90, which is either bound to their surface or forming part of the microsphere structure. Currently two different Yttrium-90 (Y-90) microsphere products are available as shown in table no.2

Table no.2- Two different microspheres used in liver cancer with their characteristics.

BRAND NAME	DESCRIPTION	ACTION
TheraSphere	Contains millions of tiny radioactive glass beads filled with Y-90	Emit beta particles that penetrate a mean of 2.5 mm into the surrounding tissue.
SIR-Spheres	Composed of millions of tiny polymer beads filled with Y-90	Emit beta particles that penetrate a mean of 2.5 mm into the surrounding tissue

- **TheraSphere**
- **SIR-spheres**

TheraSphere contains millions of tiny radioactive glass beads filled with Y-90and **SIR-Spheres** are composed of millions of tiny polymer beads filled with Y-90, which emit beta particles that penetrate a mean of 2.5 mm into the surrounding tissue.

While both are called microspheres, these products differ in microsphere size profile, base material (resin in SIR-spheres versus glass in TheraSpheres), and size of the commercially available doses. These physical characteristics of the active and inactive ingredients affect

- the flow of microspheres during injection,

- their retention at the tumor site,
- their spread outside the therapeutic target region,
- and the dosimetry calculations.

Targeting of tumors is achieved in part by injecting the microspheres directly into the branches of the hepatic artery feeding the tumor(s). The spheres provide local radiotherapy of liver tumors with simultaneous embolization of the vasculature nourishing the tumors (blocks small arteries originating from the hepatic artery). After injection, the microspheres deliver over 95% of their total radioactive dose in two weeks, as Yttrium-90 has a half-life of 64 hours. The goal of radioemobilization, which is a form of brachytherapy, is to prolong patient survival by selectively destroying tumor tissue.

SIR-Spheres are used for the treatment of unresectable metastatic liver tumors from primary colorectal cancer, with adjuvant intrahepatic artery chemotherapy of floxuridine. TheraSpheres are used in irradiation treatment or as a neoadjuvant to surgery or transplantation in patients with unresectable hepatocellular carcinoma, who can have placement of appropriately positioned hepatic arterial catheters. They are not biodegradable and should not redistribute to other organs of the body. Patients typically receive only two treatments to each lobe of the liver, given at approximately two month intervals. Both glass and resin microspheres produce heterogeneous high dose regions in the tumor. These are evident from an analysis of four explanted livers previously treated with Y-90 microsphere agents (glass or resin). In analyzing the practicality and utility of radioemobilization for patients with PET / CT positive liver metastases via use of Yttrium-90 coated microspheres in a community hospital setting, it has been observed that the median time to liver tumor progression was four months, and the median survival was 9.4 months, after treatment. Outpatient radioemobilization with Y-90 labeled microspheres in patients with hepatic metastases from a variety of tumors is well tolerated and efficacious, with modest toxicity, compared to chemoembolization techniques used previously. In a study on the safety and efficacy of Y-90 resin microsphere treatment in unresectable liver metastases from neuroendocrine cancer, a symptomatic response with median overall survival of 24.4 months has been seen.

The safety and efficacy of Y-90 microsphere treatment in patients with primary and metastatic liver cancer has been established using FDG-PET. In their study, Delbeke et al. have reported that for FDG-PET a lower sensitivity (91

versus 97%), but higher specificity (95 versus 50%) results in superior overall diagnostic accuracy compared to CT portography. In the study of Topal et al. PET has been shown as being capable of detecting liver metastases with 99% sensitivity. Several studies have compared the accuracy of FDG-PET and CT in the detection of hepatic metastases. Overall, FDG-PET was Found to be more accurate than CT.Using this technique, the anatomical and physiological determinants of radiation dose distribution and the dose response of tumor and liver toxicity in patients with liver malignancies, who underwent hepatic arterial Y-90 resin microsphere treatment, has been evaluated. It was concluded by the study that doses up to 99.5 Gy administered to uninvolved liver are tolerated with no clinical veno-occlusive disease or liver failure. The lowest tumor dose producing a detectable response is 40.1 Gy. It is also possible to evaluate a patient-specific, single photon emission computed tomography-based method of dose calculation, for the treatment planning of Y-90 microsphere selective internal radiotherapy. The tumor dose calculated with this patient-specific method is more predictive of response in liver directed Y-90 SIRT. Commercially available resin microspheres and SIR-Spheres are labeled with metallic positron emitters and evaluated as PET imaging surrogates of Y-90 SIR-Spheres. The in-vivo stability of radiolabeling is evaluated in rats by micro-PET imaging after the intravenous injection of labeled microspheres. The different resin microspheres and radionuclides evaluated in the study have all shown good radiolabeling efficiency and in-vitro stability. However, only resins labeled with 86Y and 89Zr have proved to have the in-vivo stability required for clinical application

The microspheres are typically made of glass or polymers (resin) and contain yttrium-90, which is either bound to their surface or forming part of the microsphere structure. Currently two different yttrium-90 (Y-90) microsphere products are available. The TheraSphere contains millions of tiny radioactive glass beads filled with Y-90, and SIR-Spheres are composed of millions of tiny polymer beads filled with Y-90, which emit beta particles that penetrate a mean of 2.5 mm into the surrounding tissue. While both are called microspheres, these products differ in microsphere size profile, base material (resin in SIR-spheres versus glass in TheraSpheres), and size of the commercially available doses. These physical characteristics of the active and inactive ingredients affect the flow of microspheres during injection, their retention at the tumor site, their spread outside the therapeutic target region, and the dosimetry calculations. Targeting of tumors is achieved in part by injecting the microspheres directly into

the branches of the hepatic artery feeding the tumor(s). The spheres provide local radiotherapy of liver tumors with simultaneous embolization of the vasculature nourishing the tumors (blocks small arteries originating from the hepatic artery). After injection, the microspheres deliver over 95% of their total radioactive dose in two weeks, as yttrium-90 has a half-life of 64 hours. The goal of radioemobilization, which is a form of brachytherapy, is to prolong patient survival by selectively destroying tumor tissue.SIR-Spheres are used for the treatment of unresectable metastatic liver tumors from primary colorectal cancer, with adjuvant intrahepatic artery chemotherapy of floxuridine. TheraSpheres are used in irradiation treatment or as a neoadjuvant to surgery or transplantation in patients with unresectable hepatocellular carcinoma, who can have placement of appropriately positioned hepatic arterial catheters. They are not biodegradable and should not redistribute to other organs of the body. Patients typically receive only two treatments to each lobe of the liver, given at approximately two month intervals. Both glass and resin microspheres produce heterogeneous high dose regions in the tumor. Outpatient radioemobilization with Y-90 labeled microspheres in patients with hepatic metastases from a variety of tumors is well tolerated and efficacious, with modest toxicity, compared to chemoembolization techniques used previously. In a study on the safety and efficacy of Y-90 resin microsphere treatment in unresectable liver metastases from neuroendocrine cancer, a symptomatic response with median overall survival of 24.4 months has been seen.

Several regional therapeutic techniques have been developed to produce localized tumor destruction and increase the rate of potentially curative treatments. These techniques include chemotherapy administered through the arteries using infusion pumps, selective chemoembolization, radiofrequency ablation, cryoablation, alcohol ablation, and radiolabeled Y-90 microspheres. It is important to evaluate the treatment efficacy of these techniques, so as not to miss the opportunity for an early intervention. In this context, positron emission tomography (PET) is emerging as a useful tool in the management of various cancers. It is an effective tool to detect metastasis and to monitor the response to systemic and local therapies. PET scanning with the tracer fluorine-18 fluorodeoxyglucose (FDG), called FDG-PET, is widely used in clinical oncology. The safety and efficacy of Y-90 microsphere treatment in patients with primary and metastatic liver cancer has been established using FDG-PET.

Using this technique, the anatomical and physiological determinants of radiation dose distribution and the dose response of tumor and liver toxicity in patients with liver malignancies, who underwent hepatic arterial Y-90 resin

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Breast Cancer

In the breast, cytotoxin-loaded microspheres are delivered through a catheter, surgically implanted directly into either the subclavian artery or into a branch of the subclavian artery, usually the thyrocervical trunk. More selective perfusion, however, can be obtained by the angiographic placement of catheters directly into the internal mammary artery. When administered intra-arterially these microspheres are carried by the blood flow to the capillary bed where they embolize and release their therapeutic pay-load into the target organ. Cancer cells of the breast have been targeted by delivering a single pulse of adriamycin-loaded albumin microspheres through a radiologically placed internal mammary artery catheter. Animal studies have shown that adriamycin-loaded albumin microspheres can suppress tumor growth to a greater degree than free drugs in a solution [16, 17, 18, 21].

Radiation therapy should be considered for women at high risk for local-regional tumor recurrence (women with advanced primary tumors, or four or more positive lymph nodes). Mitoxantrone (MXN)-loaded albumin microspheres for localized intratumoral chemotherapy of breast cancer localize the activity of mitoxantrone, greatly reduce systemic toxicity compared to the intravenously delivered free drug, and significantly improve median survival in the murine mammary adenocarcinoma model [21].

During the past few years the analysis of microRNA expression patterns has led to completely new insights into cancer biology. Furthermore, these patterns are a very promising tool for the development of new diagnostic and prognostic markers. However, most human tumor samples, for which long-term clinical records are available, exist only as formalin-fixed, paraffin-embedded specimens. Therefore, a study has been conducted to examine the feasibility of microRNA profiling studies and to derive comprehensive microRNA expression patterns in routinely processed formalin-fixed, paraffin-embedded human breast cancer specimens, using fluorescence-labeled bead technology. The study shows that routinely processed human formalin-fixed, paraffin-embedded breast cancer specimens are suitable for large scale as well as small-scale microRNA profiling projects using fluorescence-labeled bead technology. Therefore, this methodology can now be used for large retrospective studies, utilizing stored archival formalin-fixed, paraffin-embedded samples, together with the corresponding clinical and histopathological records.

Local, sustained delivery of cytokines into a tumor can enhance induction of antitumor immunity and may be a feasible neoadjuvant immunotherapy for breast cancer. The ability of intratumoral poly-lactic-acid-encapsulated microspheres (PLAM) containing interleukin 12 (IL-12), tumor necrosis factor (TNF-), and granulocyte-macrophage colony stimulating factor (GM-CSF) have generated a specific antitumor response in a murine model of breast cancer. A single intratumoral injection of IL-12 and TNF--loaded PLAM into a breast tumor leads to infiltration by polymorphonuclear cells and $CD8^{+}$ T-cells, with subsequent tumor regression. In addition, this local therapy induces specific antitumor T-cells in the lymph nodes and spleen, resulting in a memory-immune response [16, 17, 18, 21].

Colorectal Cancer

Conventional chemotherapy is not as effective in colorectal cancer as it is in other cancers, as the drug does not reach the target site in effective concentrations. Thus, effective treatment demands increased dose size, which may lead to undue consequences. To improve this situation, pharmaceutical technologists have been working on methods to deliver the drug more efficiently to the colon, where it can target the tumor tissues. Ciftci and Groves have shown that it is possible for a colon-targeted delivery system to selectively deliver drugs to tissues, and not through tissues. It is possible that delivery of small quantities of an anti-neoplastic agent to the inner surface of the

colon could destroy small tumors that arise spontaneously in this region, reducing the need for surgery. Several strategies can be used to selectively target drug release to the colon. Drugs are commonly delivered to the large bowel by coating them with polymeric substances such as cellulose derivatives or acrylic polymers. However, the performance of such colonic delivery systems may be limited by gastrointestinal motility and pH variations. Multiparticulate systems have been developed to overcome these limitations. Other strategies are based on the assumption that the high enzymatic activity of the rich microbial flora in the colon will act as a release trigger. Guar gum microspheres are a potential system delivery of methotrexate to the colon, for chemotherapy of colorectal cancer. Results of release studies have demonstrated that microspheres are capable of retarding the release of MTX until it reaches the colon, an environment rich in bacterial enzymes that degrade the guar gum and allow drug release to occur at the desired site.

A pH-sensitive polymer Eudragit P-4135F is used to prepare microspheres by a simple oil / water emulsification process. The formulation proved its applicability *in-vitro* as a promising device for pH-dependent colon delivery of 5-fluorouracil.

TheraSphere administration is a phase-II study, to determine the safety and efficacy of TheraSphere treatment in patients with liver-dominant colorectal metastases, and provides stabilization of liver disease with minimal toxicity in patients in whom the standard systemic chemotherapy regimen fails.

In-vivo gene therapy has been attempted using poly-(D,L-lactic-co-glycolic acid) microspheres containing the interleukin-12 gene (p2CMVmIL12) in colon adenocarcinoma (CT-26)-bearing Balb / c mice. Treatment with IL-12-loaded microspheres inhibits tumor growth significantly, although the degree of tumor inhibition does not depend on the amount of IL-12 gene loaded. Combining gene therapy with 5-fluorouracil (50 mg/kg) treatment further inhibits tumor growth compared to gene therapy alone. This indicates that microsphere formulations of pDNA may provide an efficient gene delivery system.

The therapeutic effects of 5-fluorouracil microspheres on peritoneal carcinomatosis in mice, by inducing Colon 26 or B-16 melanoma, shows much better survival than an equivalent dose of aqueous 5-FU [10, 12, 21].

Lung Cancer: In Lewis lung carcinoma cells, the paclitaxel-loaded PLGA microspheres significantly inhibit lung tumor growth *in-vivo* with no clinically apparent toxicity.

In the treatment of lung and pleural diseases, acid-prepared mesoporous spheres, chemically modified with different surface molecules (lipid, a linker having a terminal amine group, a thiol group or a tetraethylene glycol), are effective vehicles for pulmonary chemotherapeutic drug delivery and are found to be non-immunogenic and nontoxic, as evaluated by differential cell counts and lactate dehydrogenase levels in bronchoalveolar and pleural lavage fluids. Conjugating camptothecin onto PEGylated microspheres prolongs the release of camptothecin *in-vitro* and enhances the anti-cancer efficacy *in-vivo* in an orthotopic lung cancer rat model [21].

Brain Tumor

A microsphere-based system has been developed to deliver therapeutic agents to brain tumors. The polymer, poly(methylidene malonate), has been used to prepare 5-fluorouracil-sustained release biodegradable microspheres, in order to treat malignant brain tumors by local delivery of anti-neoplastic agents, This polymer presents a slow degradation rate, thus leading to a long-term local delivery system [21].

Ovarian Cancer

The effects of intra-peritoneal administration of cisplatin prepared as $_{L}$ -Lactic acid and glycolic acid copolymer microspheres in rats with ovarian cancer show increased survival of rats, without any increase in the systemic toxicity of cisplatin.

A monoclonal antibody, MJ01, which recognizes human ovarian cancer antigen CA125 is encapsulated in PLGA microspheres, which is capable of inducing T3 as evidenced by the T-cell proliferation *in-vitro*, in response to the challenge by CA125. Immunotherapy of ovarian cancer by the anti-CA125 antibody in murine rat models shows a promising response for ovarian cancer therapy [21].

Bladder Cancer

A photosensitizer conjugate, chlorin e6 (Ce6), covalently bound to 1 μm diameter polystyrene microspheres, has been investigated in the photodynamic destruction of MGH-U1 human bladder carcinoma cells *in-vitro*. The markedly greater phototoxicity of Ce6-microsphere conjugates compared to unconjugated Ce6 are therefore a consequence of the high intracellular Ce6 concentration attained by phagocytosis of the conjugates and their particular sites of intracellular localization. Thus, these conjugates are an efficient system for the delivery of photosensitizing drugs to carcinoma cells.

The efficacy of paclitaxel in the intracavitary treatment of superficial transitional cell carcinoma of the bladder, by designing bio-adhesive microspheres capable of achieving controlled release of the drug at the urothelium / urine interface, have been promoted. *In-vivo* studies have been performed in Balb / c mice after inducing bladder cancer by BBN (N-n-Butyl-N-butan-4-ol-nitrosamine) in drinking water. Intravesical administration of poly (methylidene malonate 2.1.2) paclitaxel microspheres is a promising approach for intracavitary chemotherapy of superficial bladder cancer [21].

Radioactive Microspheres for cancer treatment:

In radiotherapy, external irradiation provides only small doses to deep-seated cancers, and often causes damage to healthy tissues. It has been reported that 20 - 30 μm diameter $17Y_2O_3$ - $19Al_2O_3$ -$64SiO_2$ (mol%) glass microspheres are useful for the *in situ* irradiation of cancers. Yttrium-89 ($_{89}Y$) in this glass can be neutron bombarded to form the β-emitter $_{90}Y$ (half-life = 64.1 hours). When injected in the vicinity of the cancer, such activated glass microspheres can provide a large localized dose of β-radiation. The Y_2O_3 content of the glass in the microspheres is limited to only 17 mol%. Chemically durable microspheres with a higher Y_2O_3 content need to be developed. Phosphorus-31 ($_{31}P$) with 100% natural abundance can also be activated by neutron bombardment to form the β-emitter $_{32}P$ (half-life = 14.3 days). Chemically durable microspheres containing high phosphorus content are expected to be more effective for cancer treatment. Pure, smooth, highly spherical polycrystalline Y_2O_3 and YPO_4 microspheres have been prepared using a high-frequency induction thermal plasma melting technique. Both the Y_2O_3 and YPO_4 microspheres show high chemical durability in saline solutions buffered at pH = 6 and 7. These microspheres are expected to be more effective than the conventional glass microspheres for the *in situ* radiotherapy ofcancer[21].

Hyperthermia (Using Magnetic Microspheres) in cancer treatment:

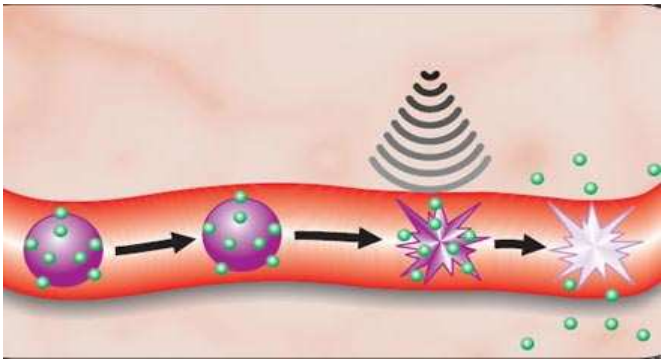
In hyperthermia therapy a focused magnetic field device is used to heat the magnetic microspheres in the organ. The magnetic treatment involves injecting thousands of beads via an incision into the artery - the main blood supply for the organ. When the microspheres are injected, they are picked up by the blood flow and eventually lodge in the malignant cells of the tumors. A magnetic field is then applied to the body to heat the magnetic microspheres and damage the cancer cells. The treatment's magnetic field is localized to an area, so the patient's body is subjected

only to a small, focused field, minimizing the possibility of side effects. Magnetically responsive albumin microspheres containing doxorubicin and magnetite (Fe3O,) are selectively targeted to Yoshida sarcoma tumors in rats, by utilizing an extracorporeal magnet. Tumor cells are inoculated subcutaneously in the tail of rats, and the tumors are allowed to grow to an average size of 9 X 45 mm, prior to initiating treatment. Drug-bearing microspheres (0.5 mg of doxorubicin per kg of body weight) are infused proximal to the tumor, through the ventral caudal artery, while the tumor is exposed to an external magnetic field of 5500 Oe for 30 minutes. The animals are treated for 12 months with a single dose; after which they exhibit total remission of the tumor, representing a disappearance of tumors as large as 60 mm in length. The experiment indicates that targeting oncolytic agents to solid neoplasms by magnetic microspheres may be a means of increasing the efficacy and decreasing the toxicity of antitumoragents.

Role of Ultrasound in anti-cancer drug delivery loaded on microspheres:

Ultrasound has been shown to enhance degradation and drug delivery from biodegradable and non-biodegradable polymeric devices. If a microsphere is partially filled with an entrapped drug substance, it is then able to transport the drug through blood vessels and release its load on being triggered by an ultrasound pulse, which cracks the shell as shown in figure 1. Cisplatin chitosan microparticles can be used. Tumor growth is delayed in Ehrlich ascites carcinoma, in Swiss Albino mice, for four to six days by the combined treatment of cisplatin chitosan microparticles and ultrasound, rather than with cisplatin chitosan microparticles alone. Ultrastructure investigations of tumor cells show severe damage in cytoplasmic organelles and cytoplasmic vacuoles.

Figure no.1 Ultrasound in Cancer treatment using Microspheres.



Some Other Microspheres: Various microspheres are now available, and are used in various cancer treatments, such as, orntide poly(d,l- lactide-co-glycolide) and poly(d,l-lactide) microspheres, a biodegradable poly(lactic acid)

microsphere formulation for *in-vivo* cytokine immunotherapy of cancer, paclitaxel loaded in PLGA microspheres, polylactic co-glycolic acid microspheres in nanofibrous scaffolds have been shown to control the release of rhPDGF-BB (platelet-derived growth factor) *in-vitro*, ethylcellulose microspheres containing 5-fluorouracil, paclitaxel-sodium alginate microsphere, yttrium silica sol-gel microspheres, and microspheres labeled with holmium-166.

Drug eluting microspheres

Chemoembolization with drug-eluting particles has been recently introduced in the field of interventional oncology. Drug Eluting Microsphere-Transarterial Chemoembolization (DEM-TACE) is a new delivery system to administrate drugs in a controlled manner, useful for application in the chemoembolization of cancer metastases. DEM-TACE is focused on obtaining higher concentrations of the drug to the tumor with lower systemic concentrations than traditional cancer chemotherapy.

In the setting of locoregional hepatic intra-arterial infusion, the drug carriers should present some essential qualities, such as, precise delivery and controlled and sustained release, as well as, high intra-tumoral concentration, for a sufficient time, without damaging the surrounding hepatic parenchyma. These may comprise on non-biodegradable polymers, such as polyvinyl alcohol (PVA), or biodegradable materials such as polylactide-coglycolides (PLGA).

Sodium acrylate polyvinyl (SAP) microspheres had been first developed by Hori and Osuga, and used for several years in Japan, for embolization of hepatocellular carcinoma and arteriovenous malformations, under the appellation of SAP. They are now CE-approved HepaSphere and FDA-approved QuadraSphere microspheres (Biosphere Medical, Rockland, MA) for the treatment of primary and metastatic liver tumors. SAP microsphere is a spherical embolic agent made of polyvinyl alcohol-sodium acrylate copolymer. It is not only able to adsorb a given drug through an ionic interaction process, but also to absorb drugs in a solution. SAP microspheres are supplied as dry particles, in several calibrated sizes 50 - 100, 100 - 150, and 150 - 200 μm, corresponding to an expanded size range of 200 and 800 μm, which can rapidly absorb aqueous medium up to 64 times their initial dry state volume, while maintaining their spherical shape. The user of SAP microspheres has to take in account two features: (1) The expansion rate is mostly dependent on ionic concentration and the drug loading capacity is limited by the solubility

of the drug in saline and by the volume that has to be injected during the embolization procedure. It therefore appears that the mechanical properties of SAP microsphere are susceptible to vary as a function of the expansion rate, so that the benefit of calibration is lost. (2) The SAP microsphere can carry a chemotherapeutic agent through an ionic interaction process and release it progressively. However, as SAP microspheres also absorb drugs in a solution, the latter can be released rapidly, leading to a plasmatic peak. Pharmacokinetic studies are needed to verify the actual *in-vivo* release.

The SAP microspheres can be loaded with doxorubicin or cisplatin for drug delivery during transcatheter arterial chemoembolization. Initial *in-vitro* and *in-vivo* studies show encouraging results, leading to their CE, marking approval for transcatheter arterial chemoembolization of unresectable hepatocellular carcinoma in combination with doxorubicin.

For 30 years, non-spherical polyvinyl alcohol (PVA) particles have been widely used to perform embolization, but they are difficult to calibrate and their behavior can be unpredictable during embolization, which leads to difficulties when performing targeted embolization.

Calibrated microspheres have drastically changed the conditions of embolization, as the radiologist may adapt the size of microspheres to the size of the vessels to be occluded, so that accurate targeting can be obtained. First, trisacryl-gelatin microspheres (EmboSphere, Biosphere Medical, Rockland, MA) were approved in Europe (CE approval), in 1997, in the United States (FDA approval) for general embolization in 2000, and specifically for uterine fibroid embolization, in 2002. Thereafter, two PVA-based microspheres have been developed, named Contour SE (Boston Scientific, Natick, MA) and Bead Block (Biocompatibles, Farnham, UK), which were both approved recently in Europe and in the United States. There are a number of experimental studies that support the advantages of using calibrated microspheres instead of non-spherical particles for embolization purposes. Using the animal model of sheep renal arteries, Andrews and Binkert have shown that trisacryl-gelatin microspheres reduced renal blood flow more quickly and reliably than PVA. Using an animal model of uterine arteries embolization, Pelage and co-workers have shown that calibrated microspheres behave differently from non-spherical PVA. Uterine artery embolization (UAE) with PVA and EmboSphere have a different impact on fertility in sheep. PVA particles lead to a drastic decrease of ewe fertility. The ranges of calibers of microspheres have been chosen

empirically and correspond to the size ranges of the arterioles detectable by angiography and accessible to catheters and micro-catheters. The industry standard today is to make calibrated size ranges that typically span a 200 μm range: 100 to 300 μm, 300 to 500 μm, 500 to 700 μm, 700 to 900 μm, and 900 to 1200μm. This calibration is mainly obtained by sieving, and for each size range a Gaussian distribution of EmboSphere is obtained, with 90 to 95% of the microspheres distributed in the size range. Examination of the pathological specimens, after embolization, can provide useful information on the final location of the microspheres in the vasculature. In the case of particular tumors such as nasopharyngeal angiofibromas and paragangliomas, it has been shown that calibrated EmboSpheres are located in their majority in intratumoral vessels, and the size of the occluded vessels increased significantly with the size of the EmboSpheres, and there was a size threshold of 500 μm for the penetration of the Embosphere in the intratumoral vasculature of these tumors.

Over the past two decades, calibrated microspheres have deeply revolutionized the field of embolization, as they have allowed the performance of an embolization that is targeted with the caliber of the microspheres and controlled by the amount of microspheres injected. One may predict that they will completely replace non-spherical particles in the near future, as microspheres of the next generation will be easily detectable by imaging and will be loaded with different types of medications to perform a targeted drug release and perhaps also be resorbable.

A List of Microspheres under investigations is given in table no.3.

Table-3: List of Microspheres under investigations.

Type of cancer	Microsphere	Basic material	Subjects
Hepatocellular	SIR-Spheres (yttrium-90)	Resin	Humans
Hepatocellular	TheraSpheres (yttrium-90)	Glass	Humans
Hepatocellular	Phosphorus-32	Glass	Humans
Hepatocellular	Mitomycin	Poly-lactide-co-glycolide	ACI-rats
Hepatocellular	cisplatin and doxorubicin	Degradable starch and iodized oil (Lipiodol)	Humans
Hepatocellular	Mhomycin C	Albumin	Humans
Hepatocellular	Mitoxantrone	Albumin	Murine mammary rats
Hepatocellular	IL-12, TNF-α	Poly-lactic-acid	Murine mammary rats
Colorectal	Methotrexate	Guar gum	Balb / c mice
Colorectal	5-fluorouracil	Eudragit P-4135F	Rat model
Colorectal	IL-12 gene	Poly-(D,L-lactic-co-glycolic acid	Balb/c mice
Colorectal	5-florouracil	Poly-(D,L-lactic-co-glycolic acid	B-16 melanoma in mice
Lung	Paclitaxel	Poly-(D,L-lactic-co-glycolic acid	Lewis lung carcinoma cells
Lung	Camptothecin	PEGylated	Orthotopic lung cancer rat model
Brain tumor	5-fluorouracil	Poly-methylidene malonate	Rat model
Ovarian	Cisplatin	L-Lactic acid and glycolic acid	Human
Ovarian	Monoclonal antibody MJ01	PLGA	*In-vitro* human ovarian cancer antigen CA125
Bladder	Chlorin e6 (Natural porphyrin)	Polystyrene	*In-vitro* MGH-U1 human bladder carcinoma cells
Bladder	Paclitaxel	Poly-methylidene malonate 2.1.2	Balb / c mice
Pancreatic	Paclitaxel	Poly-methylidene malonate 2.1.2	Balb / c mice
Various cancers	Y^2O^3 and YPO^4 (Radioactive)	Glass	*In situ* irradiation on Humans
Various cancers	Doxorubicin and magnetite (Magnetic)	Albumin	Yoshida sarcoma tumors in rats
Various cancers	Cisplatin	Chitosan	Ehrlich ascites carcinoma in mice
Metastasis	Antigens (Cancer vaccine)	Hydrophobic ion pairing proteins	Humans
Immunotherapy of cancer	Cytokine	Orntide poly (d,l- lactide-co glycolide)	Humans

Future Trend

Microspheres offer a unique carrier system for many cancer drugs and can be tailored to adhere to any cancerous tissue, including those found throughout the respiratory, urinary, and gastrointestinal tract. The microspheres can be used not only for controlled release, but also for targeted delivery of the drugs to specific sites in the body. Recent advances in medicine have envisaged the development of polymeric drug delivery systems for protein / peptide drugs and gene therapy. Although significant advances have been made in the field of microspheres, there are still many challenges ahead in this field. The most significant are the development of the universally acceptable standard evaluation methods and development of newer site-directed polymers. Polymeric science needs to be explored, to find newer microspheric polymers, with added attributes of being biodegradable, biocompatible, and bioadhesive for specific cells or mucosa, and which can also function as enzyme inhibitors for the successful delivery of proteins and peptides. A multidisciplinary approach will therefore be required to overcome these challenges and to employ microspheres as a cutting edge technology for site-targeted, controlled release drug delivery of new as well as existing drugs. The future direction of microspheres lies in potent and various other vaccine formulations that adhere to tissues and result in immunity. There is a need to look forward to further improvements in the formulations and drug delivery by these mechanisms, giving better tools to care for patients.

Conclusion

Microsphere technology, although in its nascent stage, has a great potential to cure cancer, with least side effects. . It causes less side effects as compared to other formulations as it goes directly to the site of action. It can accommodate the problems associated with API handling like hygroscopicity as the drug can be entrapped within the microsphere. Microspheres have the advantage that they are used for sustained release formulations. The release pattern in sustained release can be tailor made as per the requirement by controlling the level of polymer coating onto the microspheres. It is a technology that will grow in the years to come, and probably, the human race will have a 100% cure for cancer. Microsphere provides the biggest advantage for targeted drug delivery system. It provides a very feasible approach to different types of formulations as it can easily be formulated into tablets, capsules and it can also be given through i.v. route.

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