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MICROSPHERES: A NOVAL APPROACH FOR DRUG DELIVERY SYSTEMS

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ABSTRACT:
Microspheres constitute an important part of drug delivery system by virtue of there small size and efficient carrier characteristics. Microspheres in general have to the potential to be used for targeted and control released drug delivery: by coupling of mucoadhesive, oral, floating etc properties of microspheres. Novel drug delivery systems have several advantages over conventional multi dose therapy. Much research effort in developing novel drug delivery system has been focused on controlled release and sustained release dosage forms. Now considerable efforts are being made to deliver the drug in such a manner so as to get optimum benefits. There are various approaches in delivering a therapeutic substance to the target site in drug delivery systems. Microspheres are potential candidates for the protein drug delivery. These systems such as biodegradable microspheres are capable of delivering drugs over longer time periods than conventional formulations. Microspheres received much attention not only for prolonged release, but also for targeting of anticancer drugs to the tumor. The intent of the paper is to highlight method of preparation and novel drug delivery systems for microspheres.

Introduction:
Microspheres can be defined as solid, approximately spherical particles ranging in size from 1 to 1000 µm. They are made from polymeric, waxy, or other protective materials such as starches, gums, proteins, fats and waxes and used as drug carrier matrices for drug delivery. Natural polymers as albumin and gelatin are also used in preparation of microspheres. In addition, some related terms are used as well. For example, "microbeads" and "beads" are used alternatively. Sphere and spherical particles are also employed for a large size and rigid morphology. 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 are characteristically free flowing powders consisting of proteins or synthetic polymers which are

biodegradable in nature and ideally having particle size less than 200 μm [1]. Solid biodegradable microspheres incorporating a drug dispersed or dissolved throughout particle matrix have the potential for the control release of drug [2]. Microspheres have been extensively studied for use as drug delivery systems, where they have been shown to protect sensitive macromolecules from enzymatic and acid degradation, and allow controlled release and tissue targeting of the formulated drug [3]

Microspheres Morphology: The morphology of prepared loaded microspheres is analyzed by scanning electronic microscope (SEM) after palladium/ gold coating of the samples on aluminum stub [4]

Microspheres size distribution: The mean sizes of microspheres are determined using several methods like laser diffractometry method.

Bulk density measurement: Bulk density prepared microspheres was measured using different methods like dipping method. Measurement of glass transition temperatures (Tg) by differential scanning calorimetry (DSC): Glass transition temperature (Tg) is measured by DSC for the blank (unloaded) and the prepared loaded microspheres.

Degree of hydration: is measured to evaluate water uptake by the system as a first step in biodegradation.

Drug content determination: Microspheres thoroughly triturated and suspended in minimal amount of alcohol. This suspension is diluted with water and this is then filtered to separate shell fragments and drug content was analyzed by UV Spectrophotometer at 366.4 nm [5].

Types of Microspheres

- Nonbiodegradable
  - ceramic particles
  - polyethylene co-vinyl acetate
  - polymethacrylic acid/PEG

In non-biodegradable polymer: the drug is released by dissolution into the polymer and then diffusion through the polymer wall. Eg: levonorgestrel (Norplant) 5 year contra¬ceptive delivery system.

- Biodegradable (preferred)
  - gelatin
  - polylactic-co-glycolic acid (PLGA)
  - Biodegradable microspheres have the advantage over large polymer implants in that they do not require surgical procedures for implantation and removal. They are degraded in the body to biocompatible materials. Biodegradable microspheres are used to: Control drug release rates.
  - Conserve the stability of some drugs as proteins and peptides.
  - Also to target drugs to specific sites in the body, thereby optimizing their therapeutic response, decreasing toxic side effects, and eliminating the inconvenience of repeated injections.
  - They are also used in gene delivery and in diagnostic materials.

Examples on polymers used in microspheres CDDS:

- Chitosan MS.
- Gelatine MS.
- Polyadipic anhydride MS
- Gellan- gum MS.
- Polypeptide MS.
- Albumin MS.
- Poly lactic acid (PLA) MS.
- Poly lactic - co- glycolic acid (PLGA) MS etc.

METHODS OF PREPARATION OF MICROSPHERES:

Microspheres have been prepared by three basic methods as well as other modified methods:

- Solvent extraction / evaporation method (single and double emulsification)
- Coacervation or phase separation.
- Spray drying.
- Modified methods [6].

1. **Solvent extraction/evaporation method:** Oil phase (polymer + solvent) is injected into the aqueous phase (water + surfactant), the solvent dissolves into the aqueous phase and evaporates at the air-liquid interface. This method was successfully used for numerous of water insoluble and slightly soluble drugs encapsulated in microspheres such as lidocaine, naletrxone, bupivacaine, 5-aminosalicylis acid, flurbiprofen,

all-trans retinoic acid and testosterone. This method was successfully used for numerous of water insoluble and slightly soluble drugs encapsulated in microspheres such as lidocaine, naletrxone , bupivacaine, 5-aminosalicylis acid, flurbiprofen, all-trans retinoic acid, testosterone This method is suitable for such kind of drugs where the water soluble drugs may infiltrate to the aqueous phase and decrease its entrapment efficiency [7].

2. **Coacervation phase separation:** This method is based on dispersion of drug as solid or organic solution in organic polymeric solution, then addition of the second solvent in which the polymer is insoluble where phase separation occurs and polymer loaded with drug precipitate as microspheres. BSA is an example prototype for this method [7].

3. **Spray drying:** In this technique the drug and polymer are mixed in a solvent system, and then the solvent is evaporated by spraying the solution leaving the polymeric particles loaded with the drug. This method generates heat so it is not suitable for heat sensitive drugs. Fluconazole and tetracycline hydrochloride are examples on drugs prepared by this method.

MECHANISMS FOR MICROSPHERES RELEASE OF DRUG FROM MICROSPHERES:

In vitro drug release

The in vitro release pattern of macromolecules from ABA microspheres was influenced by the molecular mass of the solute and showed continuous release profiles above threshold level of Ca 20 kDa where as PLG microspheres yielded biphasic release profile independent of the molecular mass of the solute.

In vitro release profile of tinidazole microspheres from the preparations was examined in pH 1.2 buffers from 0-2 hr, in pH 4.5 phosphate buffer from 2 to 4 h and in phosphate buffer pH 7.2 from 4 to 12 h using the rotating basket method specified in USP XXI at 100 rpm. Microspheres equivalent to 100 mg of drug were placed in the basket and the medium was maintained at 37±0.50C. An aliquot of 10 ml were withdrawn periodically at intervals of one h and same volume of fresh medium was replaced. The concentration of the drug released at different time intervals was determined by measuring the absorbance at 366.4 nm [8]. Microspheres belong to the monolithic system which refers to a rate controlling polymer matrix through which the drug is dissolved or dispersed. In contrast, a reservoir device consists of shell like dosage form with the drug contained within a rate controlling membrane [9].

Biodegradable polymers: The in vivo elimination time is determined by, the nature of the polymer chemical linkage, the solubility of the degradation products, the size, shape and density of the device, the drug and additive content, the molecular weight of the polymer, and the implantation site. [10-11]

Microsphere Releases

- Hydrophilic (i.e. gelatin)
  - Best for burst release
- Hydrophobic (i.e. PLGA)
  - Good sustained release (esp. vaccines)
  - Tends to denature proteins
- Hybrid (amphipathic)
  - Good sustained release
  - Keeps proteins native/active

Naltroxone (Vivitrol TM) microspheres (PLA-PLGA) are the first approved alcohol dependence medication in USA.

Mechanism: the release pattern of naletroxone as a result of: absorbing water and swelling immediately after injection where the near-surface drug is released first. As water absorption continues, hydrolysis starts and after several days physical erosion begins further drug diffused to the surrounding resulting in sustained release of medication with the elimination of water and carbon dioxide as degradation products of the polymer matrix. [12]

- DURIN TM It has successfully achieved controlled, zero-order drug release for up to 6 months in vivo. Because of the broad range of physical properties and degradation times that can be designed into biodegradable polyesters, DURIN implants can deliver a wide variety of drugs including both hydrophobic and hydrophilic compounds as well as small and large molecules.
- 3- Different patterns of release could be achieved through other modifications as: coating of microspheres with other polymer for further prolongation of release time. Eg: algenate-polyethylenimine coated alginate microspheres loaded with furosemide. The membrane acted as a

physical barrier to drug release from the beads. Alginate coating of algenate-polyethylenimine beads further prolonged the release of the drug by increasing membrane thickness and reducing swelling of the beads possibly by blocking the surface pores.

Another approach of modifying the release time of drug from microspheres is by using blend of polymers with different properties like the use of blend of PLGA and polyoxyethylene. Also, PLGA microspheres made of different molecular weights of the polymer have been prepared. Three molecular weights (6,000, 30,000 and 41,000) were blended to achieve long term release where the higher molecular weights degrade more slowly. Zero order release pattern with very low or no burst effect was achieved [13].

DRUG DELIVERY SYSTEM FOR MICROSPHERES

Multi-particulate drug-delivery systems are used such as:

Microspheres As Potential Oral Drug Delivery Systems

Oral controlled release (CR) dosage forms (DFs) have been developed for the past three decades due to their considerable therapeutic advantages. [14] Biologically adhesive delivery systems offer important advantages over conventional drug delivery systems. Polymer microspheres made of biologically erodable polymers, which display strong adhesive interactions with gastrointestinal mucus and cellular linings, can traverse both the mucosal absorptive epithelium and the follicle-associated epithelium covering the lymphoid tissue of Peyer's patches. The polymers maintain contact with intestinal epithelium for extended periods of time and actually penetrate it, through and between cells. Thus, once loaded with compounds of pharmacological interest, the microspheres could be developed as delivery systems to transfer biologically active molecules to the circulation. We show that these microspheres increase the absorption of three model substances of widely different molecular size: dicumarol, insulin and plasmid DNA. [15]

Microspheres As A Potential Nasal Drug Delivery System

All types of microspheres that have been used as nasal drug delivery systems are water-insoluble but absorb water into the sphere's matrix, resulting in swelling of the spheres and the formation of a gel. The building materials in the microspheres have been starch, dextran, albumin and hyaluronic acid, and the bioavailability of several peptides and proteins has been improved in different animal models. Also, some low-molecular weight drugs have been successfully delivered in microsphere preparations. The residence time in the cavity is considerably increased for microspheres compared to solutions. However, this is not the only factor to increase the absorption of large hydrophilic drugs. Microspheres also exert a direct effect on the mucosa, resulting in the opening of tight junctions between the epithelial cells. Starch and dextran microspheres have been administered repeatedly and can be classified as safe dosage forms. [16]

The rapid mucociliary clearance mechanism in the nasal cavity can be considered as an important factor when low bioavailability are obtained for drugs given intranasally. A nasal delivery system in the form of bioadhesive microspheres has been developed. Studies in human volunteers using gamma scintigraphy showed great differences in clearance times between 3 microsphere systems and two controls. The half life of clearance for starch microspheres was found to be in the order of 240 min as compared to 15 min for the liquid and powder control formulations. The microspheres form a gel-like layer in contact with the nasal mucosa that is cleared slowly from the nasal cavity. In vitro studies using model compounds (cromoglycate and Rose bengal) showed high degrees of loading capacities for the various microsphere systems. Using various physical and chemical approaches, it was possible to a certain degree to control the release of the compounds from the microsphere systems [17].

Microspheres As A Controlled Drug Delivery System

The concept of controlled drug delivery has been traditionally used to obtain specific release rates or spatial targeting of active ingredients. The phenomenon of bioadhesion, introduced by Park and Robinson [18], has been studied extensively in the last decade and applied to improve the performance of these drug delivery systems. Recent advances in polymer science and drug carrier technologies have promulgated the development of novel drug carriers

such as bioadhesive microspheres that have boosted the use of "bioadhesion" in drug delivery. This article presents the spectrum of potential applications of bioadhesive microspheres in controlled drug delivery ranging from the small molecules, to peptides, and to the macromolecular drugs such as proteins, oligonucleotides and even DNA. The development of mucus or cell- specific bioadhesive polymers and the concepts of cytoadhesion and bioinvasion provide unprecedented opportunities for targeting drugs to specific cells or intracellular compartments. Developments in the techniques for in vitro and in vivo evaluation of bioadhesive microspheres have also been discussed [19].

Mucoadhesive microspheres include microparticles and microcapsules (having a core of the drug) of 1-1000 mm in diameter and consist either entirely of a mucoadhesive polymer or having an outer coating of it, respectively. Microspheres, in general, have the potential to be used for targeted and controlled release drug delivery, but coupling of mucoadhesive properties to microspheres has additional advantages, e.g. efficient absorption and enhanced bioavailability of the drugs due to a high surface to volume ratio, a much more intimate contact with the mucus layer, specific targeting of drugs to the absorption site achieved by anchoring plant lectins, bacterial adhesions, antibodies, etc. on the surface of the microspheres. Mucoadhesive microspheres can be tailored to adhere to any mucosal tissue including those found in stomach, thus offering the possibilities of localized as well as systemic controlled release of drugs. The application of mucoadhesive microspheres to the mucosal tissues of gastric epithelium is used for administration of drugs for localized action. Mucoadhesive microspheres are widely used because they release the drug for prolong period, reduce frequency of drug administration and improve the patient compliance [20].

Microspheres as Floating Drug-Delivery Systems

Gastric emptying is a complex process, which is highly variable and makes in vivo performance of the drug- delivery systems uncertain. In order to avoid this variability, efforts have been made to increase the retention time of the drug-delivery systems for more than 12 h. The floating or hydrodynamically controlled drug- delivery systems are useful in such applications. In recent years, the multiparticulate drug-delivery systems are used in the oral delivery of drugs. One of the approaches toward this goal is to develop the floating microspheres so as to increase the gastric retention time. Such systems have more advantages over the single-unit dosage forms. The development of floating microspheres involves different solvent evaporation techniques to create the hollow inner core [21].

Microspheres of biodegradable polymers as a drug-delivery system

Microspheres of biodegradable polymers were evaluated as a potential controlled-release drug-delivery system in the vitreous and others. The microspheres were prepared with polymers of poly (lactic acid) or copolymers of glycolic acid and lactic acid. The release of 5- fluorouracil (5-FU) from the microspheres was studied in vitro. Poly (lactic acid) microspheres released 70-85% of total 5-FU over 7 days. Microspheres of polymers with a smaller molecular weight released the drug more rapidly. Copolymer microspheres released 98% of 5-FU over 2 days. The rate of drug release was controllable by changing the molecular weight of the polymers or using a matrix of copolymer. The intra vitreal kinetics of the microspheres was studied in ten rabbits in vivo. A suspension of microspheres was injected into the vitreous cavity of five normal eyes and five vitrectomized eyes. By 48 +/- 5.2 days after injection, the microspheres disappeared from the vitreous cavity in the five normal eyes. Clearance from the vitreous cavity was accelerated in the five rabbits that underwent vitrectomy (14 +/- 2.4 days; P less than 0.001). No difference was found in the b waves of electroretinograms before and after injection of the microspheres. The histologic study showed no abnormal findings as a result of the injection [22].

Microspheres as a Multiparticulate Colonic Delivery System

A drug delivery system prepared by combination of a hydrophobic polymer and a polysaccharide has the capability to be applied as a colonic delivery system. There have been considerable researches in the field of colonic drug delivery for many purposes:

a) Development of new therapeutic agents for the treatment of colonic diseases has required colon-specific delivery systems to maximize the effectiveness of these drugs [23]

b) Introduction of once a day sustained release formulations has required a better understanding of the transit of dosage forms through the colon, and of the colonic absorption of the drug present within them[24]

c) The colon itself is susceptible to many disease states including constipation, irritable bowel syndrome and more serious diseases such as Crohn's disease, ulcerative colitis, Carcinomas and infections [25].

ADVANTAGES OF MICROSPHERES IN DRUG DELIVERY:

Controlled release delivery Biodegradable microspheres are used to control drug release rates thereby decreasing toxic side effects, and eliminating the inconvenience of repeated injections. Biodegradable microspheres have the advantage over large polymer implants in that they do not require surgical procedures for implantation and removal. PLGA copolymer is one of the synthetic biodegradable and biocompatible polymers that have reproducible and slow-release characteristics in vivo. [26] Products in the market for control drug delivery: Luproline (Lupron Depot) (depote susp.7.5 mg, 4 months, prostate cancer) [27] Goseriline acetate (Zoladex) in the United States (prostate cancer and breast CA) leporine acetate (Enantone Depot), Bromocriptine (Pariodel LA) in Europe [28-29-30-31-32].

Protein/Peptide Stability:

The key to the success of proteins to be prepared as pharmaceutical products is to have in place an efficient drug delivery system that allows the protein drugs to gain access to their target sites at the right time and for the proper duration. Four factors must be considered to fulfill this goal: route of administration, pattern of drug release, method of delivery, and fabrication of formulation. Microspheres help to protect proteins because they are not allowed to react with anything until the polymer is degraded, thus minimizing the contact with solutions that could cause the proteins to react [33].

Examples:

- Albumine (prototype).
- Vaccines (HI), diphtheria toxoid (DT), tetanus toxoid (TT), and pertussis toxin (PT) in poly (lactic acid) and PLGA microspheres were prepared by spray drying; all antigens were found to maintain their immunogenicity.
- Lyzozymes.

Drug targeting:

Drug targeting could be the greatest advantage of microspheres. Most drugs are targeted in the body to give desired results either in specific tissues or organs. A good example of how microsphere technology could be implemented is targeting cancer cells in chemotherapy, as drugs and chemical agents attack cancer cells but have a toxic effect on healthy ones which could very easily cause the cells to die.

A-Passive Targeting:

Passive targeting depends on the size of microspheres. The lung's capillaries will let the passage of particles less than seven microns (micrometers) through. If one wants the drug to be released into the lungs then the correct size would be around ten microns since they would then be captured in the capillaries. Eg. Carboplatine microspheres.

B- Active targeting:

Intigrin, Lectin, immunoglobulin's, lipoproteins, monoclonal antibodies, specific peptides and receptor antagonists were all used as ligands conjugated with microspheres as leading molecules for precise targeting.

Gene delivery:

Encapsulation of therapeutic agents such as DNA in microspheres protects the agent from enzymatic degradation, enhances tissue specificity due to localized delivery, eliminates the need for multiple administrations, and allows for controlled and sustained delivery. For controlled delivery applications, formulating native hyaluronan into microspheres could be advantageous but has been difficult to process unless organic solvents are used or hyaluronan has been modified by etherification. Therefore, we present a novel method of preparing hyaluronan microspheres using adipic dihydrazide mediated crosslinking chemistry. To evaluate their potential for medical applications, hyaluronan microspheres are incorporated with DNA for gene delivery or conjugated with an antigen for cell-specific

targeting. The results show that our method, originally developed for preparing hyaluronan hydrogels, generates robust microspheres with a size distribution of 5-20mum. The release of the encapsulated plasmid DNA can be sustained for months and is capable of transfection in vitro and in vivo. Hyaluronan microspheres, conjugated with monoclonal antibodies to E- and P-selectin, demonstrate selective binding to cells expressing these receptors. In conclusion, we have developed a novel microsphere preparation using native hyaluronan that delivers DNA at a controlled rate and adaptable for site-specific targeting [34].

Microspheres in diagnostic materials:

Gamma emitters such as 99Tc and 131I have been incorporated with microspheres for diagnostic purposes. Radio labeling of microspheres is usually achieved either during or after their preparations. Although the former method is still more commonly used in medicine, the latter is preferred, especially for shorter-lived radioisotopes, because stability and logistical problems are in this way minimized. Polymer microspheres are used in diagnostics as reagents and as elements of diagnostic devices. In this paper we compare properties of microspheres with aldehyde, carboxyl and hydroxyl groups in their surface layers. Microspheres with aldehyde groups were obtained by radical emulsion copolymerization of styrene and acrolein and in sequential redox polymerization of pyrrole followed with radical polymerization of acrolein. Microspheres with hydroxyl groups were synthesized by radical emulsion copolymerization of styrene and - t-butoxy- - vinylbenzyl-polyglycidol macromonomer.¬Microspheres with hydroxyl and carboxyl groups were synthesized by radical emulsion copolymerization of methylmethacrylate, acrylic acid and 2- hydroxyethylmethacrylate. X-Ray photoelectron spectroscopy (XPS) and atomic force microscopy (AFM) revealed that microspheres had the core-shell morphology with surface layers enriched in polymeric units with reactive groups [35].

CONCLUSION

The microspheres are easy to use and allow precise control of the embolization procedure. Their physical characteristics make them a safe embolic agent [36]. Microspheres offer a unique carrier system for many pharmaceuticals and can be tailored to adhere to any mucosal tissue, including those found in eyes, oral cavity, and throughout the respiratory, urinary, and gastrointestinal tracts. The microspheres can be used not only for controlled release but also for targeted delivery of the drugs to specific sites in body such as nasal, floating etc. Recent advances in medicine have envisaged the development of polymeric drug delivery systems for protein/peptide drugs and gene therapy. These challenges put forward by the medicinal advances can be successfully met by using increasingly accepted polymers, e.g. HYAFF, polyacrylates, chitosan, and its derivatives, polyphosphazenes, etc. Although significant advancements have been made in the field of mucoadhesive, there are still many challenges ahead in this field. A multidisciplinary approach will therefore be required to overcome these challenges and to employ mucoadhesive microspheres as a cutting edge technology for site-targeted controlled release drug delivery of new as well as existing drugs.

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