

Microspheres: A Novel Approach for Delivery of Antihyperlipidaemic Drugs

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Abstract

Microsphere drug delivery is a novel approach of drug delivery for sustain and control the rate and release of active pharmaceutical ingredient. Microspheres are the choice of drug delivery for delivering the drug at specific site to release the encapsulated drug for treatment of various diseases. Microspheres drug delivery is an attractive dosage form in terms of its flexibility and sustaining the release as compare to other sustained and controlled formulation. Most of the antihyperlipidaemic drugs comes under BCS class-II which leads to poor solubility but higher permeability, oral bioavailability of statins categories drug possess less bioavailability due to poor solubility hence microspheres are the best candidate for delivering antihyperlipidaemic drugs to systemic circulation and improves the systemic bioavailability of drug for longer period of time with lesser adverse effects. Therefore microspheres can be used in various medical department like oncology, gynecology, radiology, pulmonary, cardiology, diabetes, vaccine therapy etc. this review article focuses on recent different methods of preparation. The microspheres formulated can be later evaluated and characterized by different procedure.

Keywords: - *Coronary Heart Disease, Low-Density Lipoprotein, High-Density Lipoprotein, Microspheres, improves systemic bioavailability, controlled drug release.*

INTRODUCTION

Cardiovascular Diseases, particularly coronary heart disease (CHD) and stroke, are the leading reason behind death and permanent incapacity within the Western world. hyperlipidaemia which is an leading cause for development of atherosclerosis and major factor for cardiovascular disease. Alterations in the lipid profile are both qualitative and quantitative which represents the high level of plasma triglycerides, low level of High-Density lipoprotein (HDL), cholesterol, low-density lipoprotein and other fatty molecules responsible of cardiac diseases.^[1,3]

ESTIMATION OF CARDIOVASCULAR RISK

Several charts and algorithms are developed in try the arrange to accurately estimate the entity of coronary and cardiovascular risk on the idea of individual risk factors and conditions; most charts include, as determinants of cardiovascular morbidity or mortality, age and gender, smoking status, cardiovascular disease and polygenic disorder, additionally to plasma cholesterol. The adult treatment panel-III (ATP-III) of the National sterol Education Program, stratifies completely different risk levels with regard to LDL cholesterol

goals for treatment (160 mg/dL for low-risk, a 130 mg/dL for medium-risk and 100 mg/dL for bad patients); this is often supported a categorical analysis of risk conditions, the presence or absence of CHD or diabetes and, in borderline cases, on an estimation of absolute cardiovascular risk in keeping with the Framingham score.

The presence of the metabolic syndrome is additionally underscored as a high-risk condition. A newer revision of the ATP-III panel places even larger stress on the association of CHD and diabetes, distinguishing a 'very high-risk' population with an LDL cholesterol target of 70 mg/dL^[1,3]

PATHOGENESIS OF HYPERLIPIDAEMIA

Induration of the arteries happens once Low Density Lipoprotein (LDL) get alter in plasma and become oxidized-LDL, this oxidized-LDL get engulfed by the macrophages that results in type foam cells that get adhere to the epithelial tissue layer of blood vessels and inflicting accumulation of the platelets. Additionally macrophages lead to secrete cytokines and protein which is responsible for the deposition of the animal tissue which leads to form fatty tissue 'plug' which is

that the reason behind narrowing the blood vessels and elevation of blood pressure so increase in the chances of cardiovascular diseases e.g.- Heart attack, Angina pectoris and stroke etc.^[2]

TREATMENT STRATEGIES

Diet control by increase intake of fruits, vegetables, & unsaturated fatty acids (e.g. fish oil, & olive oil) since they're not oxidized easily like saturated fatty acids. Additionally to exercise, weight reduction, avoid alcohol & smoking.

Antihyperlipidemic Drugs "lipid lowering drugs"

1. Statins (e.g. lovastatin, fluvastatin, pravastatin, simvastatin, atorvastatin, & rosuvastatin), which are HMG-CoA reductase inhibitors that modify cholesterol synthesis.
2. Ezetimibe, which is cholesterol absorption inhibitor.
3. Niacin "nicotinic acid," that decreases secretion of lipoproteins.
4. Fibrates (e.g. clofibrate, fenofibrate, & gemfibrozil), that Increase peripheral clearance of lipoproteins.
5. Resins (e.g. cholestyramine, colestipol, & colesevelam), that are steroid sequestrants that scale back steroid absorption.^[2,3]

MICROSPHERE DRUG DELIVERY

Microspheres are characterized as spherical small particulate and free flowing powders consisting of perishable polymers mostly. They ideally have a particle size starting from 1µm to 1000 µm].^[4, 5, 8, 25] Microspheres are often loaded with drug and use for targeted drug delivering. Because the drug is loaded in polymeric microspheres it shows therapeutic action on targeted tissue only. Microspheres are designed to reinforce the therapeutic effectiveness of drug and bring home the better higher bioavailability thereby minimizing the toxicity and stripped aspect effects.

There are 2 Types of microspheres, Reservoir type and Matrix type.

a) Reservoir type: In this type, the drug is entrapped as a core within a water insoluble polymer, that controls the speed of drug unleash. The usually used polymers in such devices are ethyl cellulose or poly vinyl acetate. This sort is additionally referred to as microcapsules.

b) Matrix type: In this type, the drug is homogeneously distributed during a polymeric matrix, which controls the drug release rate. The commonly used polymers for matrix types are sodium alginate or hydroxyl propyl methyl cellulose. This

type is additionally referred to as micromatrices.

There are two mechanism of drug release from the microspheres,

Dissolution, during which the drug dissolution rate is controlled by the polymer decreasing its wettability or by itself getting dissolve in GI fluid in slower rate.

Diffusion, during which the drug diffuses from a neighborhood of upper concentration to lower concentration of drug.

ADVANTAGES

Better Patience compliance

As microspheres provides slow release of drug for extended period of your time , there is reduction in dosing frequency thanks to which it's better for patient compliance mainly pediatrics, geriatrics, psychology patients e.g.- Karan Razdan et al. formulated sustained release microspheres of cefixim.^[4,6,39]

Enhance bioavailability

Microspheres are micron in size i.e. less size give more area to extend the solubility of poorly soluble drugs and hence increasing the systemic bioavailability of the drugs. e.g.- Karan Razdan et al.

formulated microspheres of cefixim with enhanced bioavailability.^[4,6,39]

Constant drug plasma concentration

Microspheres show controlled release of drug for a protracted time as a result there's no fluctuation of drug concentration in circulation and a continuing Cmax is achieved.^[39]

Reduce adverse effects

Biodegradable polymeric microspheres are biocompatible with body environment, they are doing not required to be removed surgically. Because the drug is controlled released the systemic toxicity is additionally reduce.^[39]

Enhance stability

Liquid drugs are often converted into solid microspheres to extend the steadiness of drug and also for maintaining its clinical time period.^[4, 6, 39]

Parenteral formulation

Microspheres are spherical in shape a high dose of drug are often given as microsphere parental depot.^[53]

Targeted drug delivery

Microspheres are designed to target disease sites specifically tumor tissues, while the concentration remains low at

remaining normal tissues.^[5,11,12,36]

DISADVANTAGES

Production cost

The cost of producing of controlled drug dosage form is much more as of conventional dosage forms.^[5]

Reproducibility

Microspheres are difficult to formulate as specialization and technologies are required for manufacturing microspheres.

Potential toxicity

As microspheres are loaded with high concentration of Drug, there are chances of dose dumping resulting in potential toxicity.

Polymeric toxicity

Polymeric additives like plasticizers, stabilizers, antioxidants, etc. also are used counting on the formulation design these polymers can undergo hydrolysis, oxidation or react to biological agents causing toxicity.

Swallowing

The microsphere intended for oral administration should be swallowed and not chewed or crushed because they're designed for prolong release of medicine.

Maintaining conditions

The processing conditions of microspheres like pH, temperature, agitation, solvent evaporation, heating, etc. can influence the steadiness of the drug to be encapsulated.

COMPOSITION OF MICROSPHERES

Polymers

In microsphere formulation most ordinarily biodegradable and non-biodegradable sort of polymers are employed by researchers. The polymers which are utilized in microspheres preparation are classified into different types as

- A. Natural
- B. Synthetic

Before choosing the polymer for the microsphere formulation we would like to think about few parameters like, nontoxicity, biocompatibility, biodegradability and straightforward availability of polymers. It should be biocompatible, biodegradable, nontoxic and simply available. These polymers which pass all parameters for its selection have many advantages like they increase the duration of drug within the body due to which we recover bioavailability of drug compared to standard drug delivery system. Polymers that are utilized in microsphere formulation are of two types.

A. Natural polymers

Carbohydrates, protein and chemically modified carbohydrates these are the various sources from wherever we tend to get natural polymers.

B. Synthetic Polymers

1. Non-biodegradable polymers

Polymethyl methacrylate (PMMA) Acrolein, Glycidyl methacrylate, epoxy polymers. [4, 5, 6, 25, 34, 36]

2. Biodegradable Polymers

Lactides, their glycolides and their copolymer, polyalkyl cyano acrylate, polyanhydride. [4, 5, 6, 25, 34, 36]

Surfactant

In microsphere formation surfactant play a necessary role throughout emulsification and extrusion process. Surfactants play a important role by lowering the interfacial surface tension between Hydrophilic and hydrophobic molecules, due to which stable emulsion is formed. Use of surfactant results in formation of distinct microspheres by preventing the emulsion droplets from coalescing. HLB indicator is employed for choice of correct emulsifier. Deliquescent (Hydrophilic) surfactants have HLB value within the range of 8-18 and are used for oil in water emulsion while emulsifiers with HLB value within

the range of 3.5 to 6 are understood as lipophilic surfactants. Particle size of microspheres is decrease by increasing the concentration of surfactant due to which smaller size and size distribution of microspheres were formed.

Oil

Particle Size, size distribution and uniformity of microspheres are affected by the ratio of viscosity of oil phase to the viscosity of water phase. For e.g. it's reported that the particle size of microspheres are more which were prepared by using vegetable oil compared to microspheres which were prepared by using liquid paraffin because the viscosity of vegetable oil is higher compared to liquid paraffin . There are various sorts of oils which are utilized in the fabrication of microspheres throughout emulsification/gelation method.

Cross linkers

Most ordinarily used cross linkers for microspheres preparation are Ca²⁺, Sr²⁺ and Ba²⁺ ions. However Sr²⁺ and Ba²⁺ ions are gently ototoxic and Ca²⁺ ions are nontoxic due to which Ca²⁺ ions are wide used cross linkers for preparation of microspheres. At low concentration of Ca²⁺ ions agglomeration of microspheres takes place by increasing Ca²⁺ ions

concentration demurrer potency of microsphere slightly increases. However when optimum concentration of cross linker if additional cross linker is extra the entrapment efficiency decreases thanks to overloading of cross linker.

Solvent

Solvents principally used when microspheres are ready by exploitation solvent evaporation method, solvent act as a carrier for dispersing the polymers as well as the drug.

METHOD OF PREPARATION

Spray Drying

During this technique coating polymer is first dissolve/dispersed in organic solvent like acetone, methylene chloride and so on the drug then incorporated into polymeric solution along with high speed homogenization. The resultant mixture then atomized in stream of hot air.

Atomization results in formation of fine mist or droplets from that organic solvent evaporates like a shot which leads to the formation of microspheres during a size vary of 10um - 100um. Pavanetto et al. (1993) fabricated microspheres of vitamin D3 by using spray drying technique. [4, 6, 7, 21, 34]

Solvent Evaporation

This technique involves use of organic part as producing vehicle; this method consists of two parts. First is that the liquid innovate that the drug is incorporated along with stabilizing agent or without stabilizing agent and also the different phase is organic phase which carries polymers in organic volatile solvent admire acetone, Dichloromethane and so on then aqueous and the organic phase ought to be mixed with high speed homogenisation which results in formation of w/o emulsion then this emulsion is extra in massive aqueous phase to make w/o/w emulsion if necessary.

The resultant mixture heated along with continuous stirring which leads to evaporation of the organic phase which leads to shrinking the coating polymer across the core material and results in formation of microspheres. Sagar Balaso Sangale et al. formulated floating felodipine microspheres by using solvent evaporation technique. [4, 6, 13, 14, 22, 32]

Single emulsion technique

During this technique microsphere is ready by emulsification technique, coating polymer is dissolved in organic volatile solvent that leads to formation of polymeric solution. The resultant

polymeric solution extra into liquid part containing emulsifying agent leads to formation of o/w emulsion.

This emulsion is that the stirred for few hours underneath constant environmental condition, then filtered and dried into desiccator. Yuksel et al. (1997) fabricated polymeric microspheres containing Nicardipine by using single emulsion technique. [5, 6, 7, 16, 22, 26, 34]

Double emulsion technique

This method involves preparation of double emulsion either w/o/w or o/w/o type. The solution, which containing drug in it disseminate in organic phase which is hydrophobic. The organic part containing coating chemical compound encapsulates the drug available within the spread liquid phase and results in the formation of primary emulsion.

Then this primary emulsion undergoes homogenizing or sonicating before adding into solution of polyvinyl alcohol (PVA) to make secondary emulsion, then ready microspheres filtered and dried in desiccator. Das et al. (2007) fabricated microspheres containing Zidovudine by using double emulsion technique. [5, 6, 7, 16, 34]

Phase Separation Coacervation Technique

This technique is usually used for fabrication of reservoir variety of microspheres. Principally this method wont to encapsulate the deliquescent drugs, during this method coating polymer is dissolved in organic volatile solvent and then aqueous solution of drug is extra to allow chemical compound to coat drug , then phase separation can initiated by ever-changing the close conditions like changing temperature, changing PH, addition of salt and so on. Arunachalam et al. (2010) fabricated gelatin microspheres of ofloxacin by using phase separation coaservation technique. [4, 6, 7, 23]

Spray congealing

During this technique drug is dissolved/ disseminate in polymeric solution i.e. oleophilic polymer like wax. Hot liquefied solution then sprayed to make fine droplets into a vessel that already cooled in CO2 ice bath. [7, 24]

Solvent extraction

This technique involves the removal of organic phase by extraction of the organic solvent via exploitation deliquescent organic solvents like iso-propyl-alcohol. Organic phase then extracted by using water, this method results in decreasing

the hardening time of microspheres.^[5,34]

Quasi emulsion solvent diffusion

By exploitation {this technique/this system} micro sponges may well be prepared. It involves two parts one is internal and also the different is external. External phase consists of polyvinyl alcohol and distilled water and internal phase carries with it polymer, drug and ethanol. Internal phase is hot to 60°C then extra to external phase main. It is then maintained at area temperature. Resultant emulsion is then homogenized up to a pair of hours and fictional into micro sponges then filtered, washed and dried in vacuum oven for twenty-four hours.^[4]

Cross linking agent method

In this method cross linking agent is employed for fabrication of the microspheres. First specific focused polymeric solution has been created in an liquid medium then extra in continuous part containing oil and specific concentration of surfactant to make w/o emulsion.

Then drop wise solution of cross linker added alongside continuous stirring and permit for rigidisation of the surface of microspheres. Resultant microspheres then washed and dried.^[4]

Hot melt microencapsulation

In this method coating polymeric solution homogenized with drug, the resultant mixture is then suspended in oleophilic solvent admire semiconducting material oil along with continuous agitation/stirring and heating the solution at 5°C up to the melting point of the polymer, when the emulsion get stabilized ,it is cooled to solidify the polymeric microspheres.^[6,28]

Ionic gelation technique

In this method suspension of deliquescent polymer alongside drug is complexed with multivalent ion i.e., salt ensuing into formation of extremely viscous gel spheres. An iridescent suspension is obtained. This suspension is centrifuged to induce uniform size of microspheres. Microspheres are then washed and dried at temperature for twenty-four hours. Selveraj et al., (2011) prepared microspheres of acyclovir by using ionic gelation technique.^[4, 20, 29, 30]

EVALUATION TECHNIQUES CHARACTERISTICS

It is a very important characteristic to develop these microparticulate carriers, which helps design a sustainable and suitable carrier for proteins, drug, or antigen delivery. Every microsphere has different microstructures. These

microstructures are determining the release and stability of the carrier.

Particle size and shape

The most common ways to picture microspheres are conventional light microscopy (LM) Confocal fluorescence microscopy and scanning electron microscopy (SEM). These techniques can be used to determine the shape and outer structure of microspheres. [4, 6, 11, 13]

Conventional light microscopy:

Conventional light microscopy provides control over coating parameters in the case of double-walled microspheres. The microsphere's structures can be visualized before and after coating and the change can be measured microscopically. SEM provides higher resolution in distinction to lightweight microscopy.

Scanning microscopy: Scanning lepton microscopy permits investigations of the surfaces of the microspheres and when particles are cross-sectioned, it can even be used for the investigation of double-walled systems. [31, 38]

Confocal microscopy: Confocal visible radiation microscopy is employed to characterize the structure of multiple walled microspheres. Optical laser light

scattering and multi-size colter counter except instrumental methods, which might be used for the characterization of size, form and morphology of the microspheres.

Angle of contact

The angle of contact determines the wetting property of microspheres in terms of hydrophobicity and hydrophilicity. This physical science property is such that to a solid substance and suffering from the presence of an absorbed component. The angle of contact is measured at solid/air/water interface. The increasing and decreasing angle of contact is measured by inserting a drop during a circular cell mounted higher than the target of an inverted microscope. The Contact angle is measured at two hundred degree Centigrade at intervals a second of deposition of microspheres.

Attenuated total reflectance Fourier transforms infrared Spectroscopy

FT-IR determines the degradation of the chemical compound matrix of the carrier system and also the surface of the microspheres is investigated activity alternated total coefficient of reflection (ATR). A beam of infrared is gone through the ATR crystal in such some way that it reflects over and over through the sample to supply IR spectra principally of

surface material. Along ATR-FTIR provides information concerning the surface composition of the sample microspheres.[4,6,36]

Density Determination

The multi-volume pycnometer is employed to work out the density of microspheres. During a cup accurately weighed sample of microspheres is placed then the cup is placed within the multi-volume pycnometer in a cup is placed into the multi-volume pycnometer. At constant pressure, noble gas is introduced in the chamber and allowed to expand. Enlargement of the noble gas gas decreases the pressure at intervals the chamber and two consecutive readings of reduction in pressure at completely different initial pressure are noted. From these two pressure readings, the amount additionally the density of the sample microsphere is determined. [5, 11]

Electron spectroscopy for chemical analysis

Electron spectroscopy for chemical analysis (ESCA) determines the surface chemistry of the microspheres. ESCA also determines the atomic composition of the surface of microsphere and layer degradation of the perishable microspheres by making spectra by the

assistance of electron spectroscopy. It's also known as X-ray photoelectron spectroscopy (XPS). [4, 36]

Surface carboxylic acid residue

Radioactive glycine is employed to live surface acid residue. Hot glycine conjugates are ready by the reaction of C14 - glycine alkyl radical organic compound complex with the sample microspheres. The glycine residue shaped is coupled exploitation the soluble compression 1- ethyl- 3 (3-dimethyl amino propyl) carbodiimide (EDAC). Emission of the conjugate is then measured employing a liquid counter tube technique. Thus, the carboxylic acid residue is compared and correlated. Free carboxylic acid residue can be wont to measure hydrophobic or deliquescent or the other derivatized variety of the microspheres. [4]

Isoelectric point

To work out isoelectric point equipment called micro electrophoresis is employed that measures the electrophoretic mobility of microspheres. Mean velocity at every pH values starting from 3-10 is calculated by activity the time of particle movement over a distance of one mm. Electrical mobility of the particle is determined exploitation this data. The electrophoretic mobility can be regarding these three

parameters admire surface contained charge, ionisable behavior or ion absorption nature of the microspheres. [4, 36]

Surface amino acid residue

The radioactive c14-acetic acid conjugate is used within the determination of Surface associated amino acid residue. The amino acid residue is decided indirectly by first determinant radical acid residue through liquid scintillation counter. EDAC is employed to condense the amino and also the c14 - acetic acid carboxylic acid residue. The indirect estimation technique is completed to work out the free amino or the free carboxylic acid residues, by activity the emission of the c14 glycine alkyl radical organic compound complex having acetic acid or the glycine conjugates.[4,5,36]

Entrapment Efficiency

Entrapment efficiency of the microspheres is decided by permitting washed microspheres to the lysate. The lysate is then subjected to the determination of active constituents as per the necessity of a monograph. The percent encapsulation efficiency is calculated by exploitation the following equation:

$$\% \text{ Entrapment} = \frac{\text{Actual content}}{\text{Theoretical content}} \times 100. \quad [11, 20, 36, 38]$$

APPLICATIONS OF MICROSPHERES IN PHARMACEUTICAL INDUSTRIES

Oral drug delivery

Oral route is simple and convenient route for administration of the drug with higher patient compliance. There are massive numbers of pharmaceutical merchandise administered via oral route. Principal behind oral absorption is completely depends on solubility and permeableness of the drug. Microsphere drug delivery offers sustained and controlled manner drug unleash for extended amount of your time results in scale back dosing frequency and improve patient compliance. [27,51]

Ocular drug delivery

Microspheres are sensible carrier for the ocular drug delivery. By exploitation microspheres drug delivery bioavailability of the drug has been improved as compare to the liquid ocular preparations. Because of their sustained or controlled unleash mechanism microspheres are used for long lasting release of drug that results in scale back the dosing frequency. [6, 10, 35, 42]

Intranasal drug delivery

This route is especially most well-liked for the delivery of proteins and also the peptides. Typical formulations are simply

get drained far from nasal mucosa. Bioadhesive microspheres offer higher bioavailability by exerting its sustained/controlled mechanism. [6, 9, 43]

Gene Therapy

In this technique microspheres are fictional with viral vectors in gene drug delivery. This method offers ease of preparation, web site targeting, and enormous scale production and shows low immunogenic response as compare to the direct viral vector drug delivery. [6, 44]

Buccal drug delivery

Mucoadhesive microspheres function as reservoir for drug; it releases the drug from the applied site for extended period of time. Mucoadhesive polymers reside on the mucous membrane of the cavity and act as reservoir; it additionally improves the bioavailability of the drug by avoiding first pass metabolism. [6, 37, 40]

Percutaneous and Topical Drug Delivery

Polymers having sensible film forming ability used for delivery of drug through skin. For instance Chitosan, Alginate and PLGA loaded microspheres are used as Percutaneous Drug Delivery. It also used for delivering the drug for topical application. E.g., Asiatic side loaded

microspheres for wound healing showing acceleration in re-epithelization still as promoting the angiogenesis. [45, 54]

Gastrointestinal drug delivery

Microspheres are used for delivery of the potent drug to the precise web site (GIT). Eudragit, alkyl radical cellulose, carbopol and alginate microspheres are used for delivery of the drug at specific site in GI Tract. It prevents the first pass metabolism of the drug and will increase the bioavailability of the drug. [6]

Intra-Tumoral and local drug delivery

Anti-tumor medicine ought to be delivered at the tumor site in applicable concentration parentetically paclitaxel loaded microspheres. Film forming polymers are wont to sustain the discharge at local site i.e. oral cavity. [44, 47, 52]

Colonic drug delivery

Microspheres are used for delivering the drug at specific site in bowel i.e. colon hypoglycemic agent loaded into chitosan microspheres targeted to release its drug at colon. [6, 48]

Vaginal drug delivery

Microspheres drug delivery used for treating the vaginal infections admire

mycotic infection of the reproductive organ tract. Chitosan, Gelatin, PLGA polymers are used for fabricating the microspheres to treat vaginal infections. [6,49]

Radioactive applications

Radioactive isotopes loaded microspheres are used to treat many diseases like liver and spleen tumor. For instance Yttrium 90 loaded microspheres are used to treat cancer. [50]

Medical applications

Microspheres also are employed in nutraceuticals to deliver proteins, peptides over extended amount of time. Magnetic microspheres are used to extract the stem cells and bone marrow purging.

Vaccine drug delivery

Microsphere Drug Delivery has been showing economical in vaccine delivery; Thiolated Eudragit Microspheres would be a higher candidate for oral vaccine delivery. It also evokes the general and tissue layer immunity. [36]

CONCLUSION

Microspheres drug delivery system has been developed for improvement of bioavailability and sustaining the release of drug. Antihyperlipidaemic drugs suffer

from poor bioavailability and solubility hence need of novel drug delivery system becomes essential. Microsphere drug delivery system shows promising tool for delivery of such drugs. Subsequently various methods of preparation of microspheres were discussed along with their application.

REFERENCES

1. Bertolotti M, Maurantonio M, Gabbi C, Anzivino C, Carulli N. Review Article: Hyperlipidaemia and Cardiovascular risk. *Aliment pharmacol ther* 2005; 22: 28-30.
2. Tripathi.K.D. Essentials of medical pharmacology. 7th edition, jaypee brothers medical publishers: New Delhi; 2013:634-41.
3. Toth PP. Drug treatment of hyperlipidaemia drugs 2010 July 1; 70(11):1363-79.
4. Prasad BS, Gupta VR, Devanna N, Jayasurya K. Microspheres as drug delivery system-a review article. *J glob trends pharm sci*.2014; 5:1961-72.
5. Batra Deepa, Kakar Satinder, Singh Ramandeep, Nautiyal Ujjwal. Magnetic microspheres as a targeted drug delivery system: an overview, *journal of drug delivery research*: 2-3.

6. Sharma M, Dev SK, Kumar M, Shukla AK. Microspheres as suitable drug carrier in sustained release drug delivery: an overview. *Asian J Pharm Pharmacol.* 2018; 4:102-8.
7. Virmani T, Gupta J. Pharmaceutical application of microspheres: An approach for the treatment of various diseases. *Int J Pharm Sci Res.* 2017; 8:3252-60.
8. Lengyel M, Kállai-Szabó N, Antal V, Laki AJ, Antal I. Microparticles, microspheres, and microcapsules for advanced drug delivery. *Sci Pharm.* 2019; 87:20.
9. Farraj NF, Johansen BR, Davis SS, Illum L. Nasal administration of insulin using bioadhesive microspheres as a delivery system. *J Control Release.* 1990; 13:253-61.
10. Genta I, Conti B, Perugini P, Pavanetto F, Spadaro A, Puglisi G. Bioadhesive microspheres for ophthalmic administration of acyclovir. *J Pharm Pharmacol.* 1997; 49:737-42.
11. Chandna A, Batra D, Kakar S, Singh R. A review on target drug delivery: magnetic microspheres. *J Acute Dis.* 2013; 2:189-95.
12. Zhang J, Zhang S, Wang Y, Zeng J. Composite magnetic microspheres: Preparation and characterization. *J Magn Magn Mater.* 2007; 309:197-201.
13. Sangale SB, Barhate SD, Jain BV, Potdar M. Formulation and Evaluation of Floating Felodipine Microsphere. *Int J Pharm Res Dev.* 2011; 3:163-70.
14. Srivastava AK, Ridhurkar DN, Wadhwa S. Floating microspheres of cimetidine: Formulation, characterization and in vitro evaluation. *Acta Pharm.* 2005; 55:277-85.
15. Chapter in a book: Urs HÄfeli. Radioactive microspheres for medical applications. In: Marcel De Cuyper Jeff W. M. Bulte, editors. *Physics and Chemistry Basis of Biotechnology. Volume 7.* Springer: Dordrecht; 2001. P. 213-48
16. El-Helw AM, Al-Hazimi AM, Youssef RM. Preparation of sustained release phenobarbitone microspheres using natural and synthetic polymers. *Med Sci.* 2008;15:39-51
17. Cai Y, Chen Y, Hong X, Liu Z, Yuan W. Porous microsphere and

- its applications. *Int J Nanomed.* 2013; 8:1111.
18. Zhang CZ, Niu J, Chong YS, Huang YF, Chu Y, Xie SY et al. Porous microspheres as promising vehicles for the topical delivery of poorly soluble asiaticoside accelerate wound healing and inhibit scar formation in vitro & in vivo. *Eur J Pharm Biopharm.* 2016; 109:1-3.
 19. Li S, Nguyen L, Xiong H, Wang M, Hu TC, She JX et al. Porous-wall hollow glass microspheres as novel potential nanocarriers for biomedical applications. *Nanomed Nanotechnol.* 2010; 6:127-36.
 20. Uyen NT, Hamid ZA, Ahmad NB. Synthesis and characterization of curcumin loaded alginate microspheres for drug delivery. *J Drug Deliv Sci Tec.* 2020; 58:101796.
 21. L. Mua, S.S. Fenga. Fabrication, characterization and in vitro release of paclitaxel (Taxol[®]) loaded poly (lactic-co-glycolic acid) microspheres prepared by spray drying technique with lipid / cholesterol emulsifiers. *J Control Release.* 2001;76: 239-254
 22. Yi-Yan Yanga, Hui-Hui Chiab, Tai-Shung Chunga. Effect of preparation temperature on the characteristics and release profiles of PLGA microspheres containing protein fabricated by double-emulsion solvent extraction/evaporation method. *J Control Release.* 2000; 69: 81-96.
 23. Bhattacharya S, Alam M, Dhungana K, Yadav S, Chaudhary KR, Chaturvedi KK et al. Preparation and Evaluation of Diclofenac Gelatin Microspheres Using Coacervation. *Int J Pharm Res Innov.* 2020;13:14-21
 24. Bertoni S, Albertini B, Passerini N. Different BCS Class II Drug-Gelucire Solid Dispersions Prepared by Spray Congealing: Evaluation of Solid State Properties and In Vitro Performances. *Pharmaceutics.* 2020; 12:548.
 25. Gurung BD, Kakar S. An overview on microspheres. *Int J Health Clin Res.* 2020; 3:11-24.
 26. Baimark Y, Srisuwan Y. Preparation of polysaccharide-based microspheres by a water-in-oil emulsion solvent diffusion method for drug carriers. *Int J Polym Sci.* 2013;2013:1-6
 27. Kim JU, Shahbaz HM, Lee H, Kim T, Yang K, Roh YH et al.

- Optimization of phytic acid-crosslinked chitosan microspheres for oral insulin delivery using response surface methodology.
- a. Int J Pharm. 2020; 588:119736.
28. Mathiowitz E, Langer R. Polyanhydride microspheres as drug carriers I. Hot-melt microencapsulation. J Control Release. 1987; 5:13-22.
 29. Khanam N, Alam MI, Sachan AK, Gangwar SS. Fabrication and evaluation of propranolol hydrochloride loaded microspheres by ionic-gelation technique. Der Pharm Lett. 2012; 4:815-20.
 30. Patel N, Lalwani D, Gollmer S, Injeti E, Sari Y, Nesamony J. Development and evaluation of a calcium alginate based oral ceftriaxone sodium formulation. Prog Biomater.2016;2: 117-33
 31. Fujii S, Okada M, Sawa H, Furuzono T, Nakamura Y. Hydroxyapatite nanoparticles as particulate emulsifier: fabrication of hydroxyapatite-coated biodegradable microspheres. Langmuir. 2009; 25:9759-66.
 32. Trivedi P, Verma AM, Garud N. Preparation and characterization of aceclofenac microspheres. Asian J Pharm. 2014;2:110-15
 33. Pradeesh TS, Sunny MC, Varma HK, Ramesh P. Preparation of microstructured hydroxyapatite microspheres using oil in water emulsions. Bull Mater Sci. 2005; 28:383-90.
 34. Naveen HP, Nesalin JA, Mani TT. A Modern Review on Microsphere as Novel Controlled Drug Delivery System. Asian J Res Pharm Sci Biotech.2014; 2: 62 - 69.
 35. Thiel MA, Morlet N, Schulz D, Edelhauser HF, Dart JK, Coster DJ et al . A simple corneal perfusion chamber for drug penetration and toxicity studies. Br J Ophthalmol. 2001; 85:450-3.
 36. Rastogi V, Shukla SS, Singh R, Lal N, Yadav P. Microspheres: a promising drug carrier. J Drug Deliv Ther. 2016; 6:18-26.
 37. Remuñán-López C, Portero A, Vila-Jato JL, Alonso MJ. Design and evaluation of chitosan/ethylcellulose mucoadhesive bilayered devices for buccal drug delivery. J Control Release. 1998; 55:143-52.
 38. Masaeli R, Kashi TS, Dinarv R, Tahriri M, Rakhshan V, Esfandyari-Manesh M.

- Preparation, characterization and evaluation of drug release properties of simvastatin-loaded PLGA microspheres. *Iran J Pharm Res.* 2016; 15:205.
39. Tejash P, Shah CN, Shah DP. Microspheres: as a Novel Controlled Drug Delivery System a Review. *Pharm Sci Monit.* 2016;7:37-53
40. Rathbone MJ. Human buccal absorption. I. A method for estimating the transfer kinetics of drugs across the human buccal membrane. *Int J Pharm.* 1991; 69:103-8.
41. Jiang WZ, Cai Y, Li HY. Chitosan-based spray-dried mucoadhesive microspheres for sustained oromucosal drug delivery. *Powder Technol.* 2017; 312:124-32.
42. Liu W, Lee BS, Mieler WF, Kang-Mieler JJ. Biodegradable microsphere-hydrogel ocular drug delivery system for controlled and extended release of bioactive aflibercept in vitro. *Curr Eye Res.* 2019; 44:264-74.
43. Sahu Y, Jain S, Shukla K. Mucoadhesive microspheres based formulation development of ziprasidone hydrochloride for nasal delivery. *J Drug Deliv Ther.* 2020; 10:175-81.
44. Xu Q, Leong J, Chua QY, Chi YT, Chow PK, Pack DW et al . Combined modality doxorubicin-based chemotherapy and chitosan-mediated p53 gene therapy using double-walled microspheres for treatment of human hepatocellular carcinoma. *Biomaterials.* 2013; 34:5149-62.
45. Lee S, McAuliffe DJ, Kollias N, Flotte TJ, Doukas AG. Photomechanical delivery of 100?nm microspheres through the stratum corneum: Implications for transdermal drug delivery. *Lasers Surg Med.* 2002; 31:207-10.
46. Goswami N, Joshi G, Sawant K. Floating microspheres of valacyclovir HCl: Formulation, optimization, characterization, in vitro and in vivo floatability studies. *J Pharm Bioallied Sci.* 2012; 4:S8.
47. Brett A. Almond , Ahmad R. Hadba, Shema T. Freeman , Brian J. Cuevas ,Amanda M. Yorke, Carol J. Detrisacb et al. Efficacy of mitoxantrone-loaded albumin microspheres for intratumoral chemotherapy of breast cancer. *J Control Release.* 2003;91 :147-155

48. Wang QS, Wang GF, Zhou J, Gao LN, Cui YL. Colon targeted oral drug delivery system based on alginate-chitosan microspheres loaded with icariin in the treatment of ulcerative colitis. *Int J Pharm.* 2016; 515:176-85.
49. Yang TT, Cheng YZ, Qin M, Wang YH, Yu HL, Wang AL et al. Thermosensitive chitosan hydrogels containing polymeric microspheres for vaginal drug delivery. *BioMed Res Int.* 2017; 2017.
50. Eyles JE, Spiers ID, Williamson ED, Alpar HO, Williamson ED. Tissue distribution of radioactivity following intranasal administration of radioactive microspheres. *J Pharm Pharmacol.* 2001; 53:601-7.
51. Nafea EH, El-Massik MA, El-Khordagui LK, Marei MK, Khalafallah NM. Alendronate PLGA microspheres with high loading efficiency for dental applications. *J Microencapsul.* 2007; 24:525-38.
52. Kanafi MM, Ramesh A, Gupta PK, Bhonde RR. Dental pulp stem cells immobilized in alginate microspheres for applications in bone tissue engineering. *Int Endod J.* 2014; 47:687-97.
53. Chang B, Ahuja N, Ma C, Liu X. Injectable scaffolds: Preparation and application in dental and craniofacial regeneration. *Mater Sci Eng R Rep.* 2017; 111:1-26.
54. Zhao XH, Tay FR, Fang YJ, Meng LY, Bian Z. Topical application of phenytoin or nifedipine-loaded PLGA microspheres promotes periodontal regeneration in vivo. *Arch Oral Biol.* 2019; 97:42-51.