Review Article

A Review on Cyclodextrin Nanosponges

Abeena P B*, Praveen Raj R, Daisy P A
Department of Pharmaceutics, St. Josephs College of Pharmacy, Cherthala, Kerala, India-688524.
***Corresponding author's E-mail:** abeenapb1@gmail.com

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ABSTRACT

Cyclodextrin based nanosponges are hyper crosslinked polymeric structures having nanometers size range. Cyclodextrin based nanosponges have high complexing ability with many molecules than its parent cyclodextrins. Solubility of many poorly soluble compounds can be enhanced by inclusion and non-inclusion behaviour of β cyclodextrin nanosponges. The structure of polymeric mesh and release of entrapped drug can be manipulated accordingly by varying the type of cross linker and degree of cross linking. Beta cyclodextrin nanosponges offers site specific targeting with the unique advantage of controlled release and are proven to be biologically safe. A number of both hydrophilic and lipophilic molecules can be complexed with cyclodextrin based nanosponges. The nanosponges can be used to increase solubility, modulate the drug release, protein/peptide drug delivery, cosmetics and for production of various materials for diagnostic applications.

Keywords: β cyclodextrin, nanosponge, melt method, solvent method, ultrasound assisted nanosponge synthesis.

INTRODUCTION

Nanoparticles based drug delivery system has been used to produce controlled and sustained delivery of drugs for prolonged period of time. The nanocarriers are able to modify the physicochemical properties of drugs, hence the desired pharmacokinetics and biodistribution of drugs can be achieved.

Nanosponges are a class of materials able to load both hydrophilic and hydrophobic drug molecules. Nanosponges are tiny sponges having three-dimensional network structure. The outer surface of the nanosponges are porous in nature, which offers controlled release of the active constituents from the dosage forms. Drug molecules having molecular weight between 100-400Da, solubility pattern of less than 10mg/ml in water, melting point below 250^0c and consists of less than five condensed rings are better suited to formulate as nanosponges. There are various types of nanosponges such as titanium nanosponges, silicon nanosponges, hyper cross-linked polystyrene nanosponges, cyclodextrin based nanosponges….

The cyclodextrin based nanosponges are hyper crosslinked polymeric structures having nanometer size range. Drug complexation with α, β, γ – cyclodextrin and their derivatives has been investigated. The structure of which consists of cyclic oligosaccharides in a truncated cone structure. Due to low cost and suitable cavity size for varying compounds, beta-cyclodextrin is more preferred. Cyclodextrin based nanosponges has high complexing ability with many molecules than its parent cyclodextrins. These have a high solubilizing capacity for poorly soluble drugs by their inclusion and non-inclusion behaviour. It can be used for oral and parenteral routes and hence provide an effective carrier for drug delivery. These are non-allergenic, non-mutagenic and biodegradable. In cyclodextrin nanosponges several cross-linking agents are used to combine the cyclodextrin molecules together. The cross linkers like carbonyldiimidazole, pyromellitic dianhydride and diphenyl carbonate are commonly used.[1]

Chemicals used for the synthesis of nanosponges

The important materials used in the synthesis of nanosponges are

1. Polymers

Hyper cross-linked polystyrenes, methyl β-cyclodextrin, alkyloxycarbonyl cyclodextrin, 2-hydroxy propyl β-cyclodextrin, α-cyclodextrin, β-cyclodextrin.

2. Co-polymers

Poly(valerolactone-allylvalerolactone), poly (methyl methacrylate) (PMMA), poly vinyl alcohol, hydroxyl propyl methyl cellulose, ethyl cellulose.

3. Aprotic solvents

Ethanol, methanol, dimethylformamide, dimethyl sulfoxide, dimethylacetamide.

4. Cross-linkers

Diphenyl carbonate, diaryl carbonate, hexamethylene diisocyanate, carbonyldiimidazole, carboxylic acid dianhydride, toluene-2,4-diisocyanates, epichlorhydrin, pyromellitic anhydride, dichloromethane, polyamidoamine.[2]

Synthesis of cyclodextrin nanosponges

There are mainly four methods used to prepare cyclodextrin nanosponges, they are:

- Melt method

- Solvent method
- Ultrasound-assisted synthesis
- Microwave assisted synthesis

1. *Melt method*

In this method crosslinking agent and cyclodextrins are melted together. All ingredients are homogenized finely and heated at 100^0c after placing in a 250ml flask. It is then subjected to magnetic stirring for 5 hrs. The homogenised mixture is cooled and the product obtained is broken down using a spatula. The product is washed repeatedly with suitable solvents to free of unreacted excipients and byproducts.

2. *Solvent method*

In this method, the melting step is avoided. The solvents such as dimethyl formamide or dimethyl sulfoxide (DMF/DMS) is used to solubilise the crosslinking agent. The polymer is treated with a polar aprotic solvent and add this mixture excessively in to the above cross-linker solution. The reaction temperature varying from 10^0c to the reflux temperature of the solvent can be used. The reaction time is ranging from 1 to 48 hrs. The cooled solution is added to an excess of double distilled water. The product obtained is separated by filtration under vacuum and purified by prolonged soxhelet extraction with absolute ethanol (99.9%v/v). The suitable crosslinking agent used in this method are carbonyl compounds such as diphenyl carbonate, dimethyl carbonate or carbonyldiimidazole. The spherical solid nanoparticles obtained have very high solubilising capacity by forming either inclusion or non inclusion complexes. The obtained hyper crosslinked cyclodextrins are ready for drug loading by incubation. The size of the prepared nanosponges can be reduced by high pressure homogenization technique. For this, an aqueous suspension of nanosponges are homogenised with an ultra turrax at fixed rpm for 10 min. then it is subjected to cycles of homogenisation and produces nanosponges with narrow sizes.

3. *Ultra sound assisted synthesis*

Here, the reaction is carried out in the absence of solvents. In this method, anhydrous β-cyclodextrin are mixed with diphenyl carbonate(cross linking agent) in a 250ml flask. The flask is then kept in an ultrasound bath previously filled with water and heated to 90^0c. It is then sonicate for 5 hrs. Subsequent crystallisation and purification steps are same as that of melt/solvent method. The main advantage of this technique is no use of organic solvents which may be harmful. Ultrasonication can be replaced effectively by high energy processes like probe sonication.

4. *Microwave-assisted synthesis*

It is the simplest method for the synthesis of nanosponges. Microwave irradiation is used for cross linking the cyclodextrins molecules. Higher degree of crystallinity are the main property of these nanosponges. Compared to conventional methods, about fourfold reduction in reaction time are observed. It also provides homogeneous sized particles with uniform crystallinity.[3]

Loading of drug into nanosponges

Drug candidates can be inserted in to the nanocavities of β-cyclodextrin. The presence of crosslinking in the beta-cyclodextrin nanosponges provide more interactions with the guest molecules. The hydrophobic cavities of cyclodextrins are surrounded by hydrophilic nanochannels of polymeric mesh makes interactions with molecules of different structures and lipophilicities.

The drug can be loaded in to the nanocavities of cyclodextrins by mainly two methods. First is freeze drying method in which the nanosponges are suspended in to the drug dispersions followed by freeze drying using a lyophilizer. Second method is solvent evapouration technique in which nanosponges are added in to the drug dispersed in a suitable organic solvent and then it is triturated continuously until the solvents get evaporated.

Characterization of nanosponges

To study the nature and properties of nanosponges loaded with drugs, it is evaluated by the parameters described below.

1. Particle size and polydispersity index.

The average diameter and polydispersity of the particles are determined using a particle size analyser. It works on the principle of dynamic light scattering also known as photon correlation spectroscopy. It correlates the variation of intensity of scattered light with the particle size by the autocorrelation function. PCS/DLS gives the hydrodynamic diameter since it considers all the particles are spherical under measurement. By taking in to consideration of the effective viscosity, temperature and refractive index of the dispersing medium, DLS/PCS measures the particle size. So the particle size measured would be as a result of taking all factors in to consideration. It is always recommended to perform qualitative analysis as well. For this, the sample is dispersed in water or suitable organic solvents and the analysis is carried out using SEM, TEM or ESEM.

2. Zeta potential

zeta potential determination is the measure of surface charge of the particles. Surface charge affects drug distribution in the body and interaction with biological membranes. It mainly considers diffusion coefficient and electrophoretic mobility. These are transformed in to zeta potential using smoluchowski equation or stokes equation. The P^H and electrolyte concentration are the parameters which considered during zeta potential measurement. The zeta potential of the samples can be determined by placing in an electrophoretic cell after diluting with Kcl solution. An electric field of about 15v/cm was applied across the electrodes.

3. SEM and TEM analysis

The particle shape and morphology of nanosponges can be determined by SEM and TEM analysis. SEM involves applying conductivity to the particles under vacuum using a focused electron beam. Environmental scanning electron microscopy (ESEM) can be employed for moist samples. TEM study involves the investigation of surface morphology of the particles. For this the nanosponge sample was diluted with HPLC water and applied on carbon coated copper grid. It is then stained with proper reagent, dried and scanned with transmission electron microscope at various magnifications.

4. Drug loading and entrapment efficiency

For this, drug loaded nanosponges are placed in a solvent in which drug is soluble. It is then sonicate for 5 min at room temperature. So that the drug gets dissolved in to the solvent. Then the amount of drug present is found out by using techniques like uv-visible spectrophotometer and high performance liquid chromatography. Encapsulation efficiency and drug loading are calculated with the help of calibration curve.

% drug encapsulation efficiency = amount of drug encapsulated/total amount of drug *100.

5. Saturation state interaction

To find out the saturated solution interaction, uv spectroscopy is used. For this purpose, to the fixed concentration of drug solution increasing concentration of nanosponges are added. The sample mixture are kept for overnight. Then to find out interaction absorbance of the filtered solution was measured in the uv range and observed for shift of the λmax compared to pure drug.

6. Phase solubility studies

The phase solubility studies gives an idea about the effect of nanosponges on drug solubility. Phase solubility constants can be determined by adding saturated drug solutions in to increasing concentrations of blank nanosponges. The study is conducted till the equilibrium is attained. Nanosponge concentration v/s drug concentration is plotted on a graph and is defined as per Higuchi and conors classification. The stability constant value gives an indication ofinteraction between drug and nanosponges. The dissolution rate and solubility of the drug increases with increased interaction of drug and nanosponges.

7. In-vitro release studies

Drug release from nanosponges can be determined by in-vitro release study.for this purpose multi-compartmentn (n=6) rotating cells can be used. The donor phase and receptor phase of the rotating cells are separated by a dialysis membrane (Sartorius cut off 12000 Da). An aqueous dispersion of nanosponge containing drug was placed in donor compartment. The receiving phase is filled with phosphate buffer of appropriate ph and add 0.5%w/v of sodium lauryl sulphate (1ml) to maintain proper sink conditions. After fixed time intervals the receiving phase is completely withdrawn and filled with fresh medium. It is then diluted suitably and the amount of drug is determined by using a suitable analytical method. The drug release is determined and calculate the release pattern.[4]

8. Porosity

The porosity study was conducted to determine the extent of nanocavities and nanochannels formed. Because of the ability of helium gas to penetrate inter and intra particular channels of material, helium pycnometer is used to assess the porosity of nanosponges. The extent of helium displacement gives the true volume of material. The following equation is used to determine the percent porosity.

%porosity (E) = bulk volume-true volume/bulk volume *100.

9. Water uptake and swelling studies

The swelling and water uptake studies are conducted for nanosponges prepared with swellable polymers like polyamidoamines. It can be carried out simply by direct soaking in water.. The following equation gives the swelling index and water uptake respectively.

Percent swelling = St/So*100.

Where, St = marking of the cylinder at specific time point after soaking.

So = initial marking of cylinder before soaking.

Percent water uptake = Mt/Mo *100

Mt = mass of the hydrogel after specific time

Mo = initial mass of dry polymer.

10. Fourier transform-infrared spectroscopy (FTIR)

FTIR- spectroscopy reveals the functional groups present in the structure. During polymer synthesis monomers are linked together to form polymer. The presence of functional group peaks in the FTIR spectrum gives the formation of bonds between the monomer units of polymer. A vibration spectrum can be seen with crystalline structure. The FTIR spectra of pure drug, polymer, dried nanosponges and drug loaded nanosponges were observed for any possible interaction. The range of 4000 to650 $c.m^{-1}$ are taken for observation. FT-IR also gives the hydrophilic and hydrophobic sites of the system. The absence of any functional group peak in the spectra of hydrophobic drug is an indication of its inclusion in cyclodextrins nanosponge cavity.

11. Powder-X-ray diffraction (PXRD)

The chemical decomposition and complex formation of a mixture of compounds are given by the diffraction peaks. Complexation of drug with nanosponges varies the diffraction patterns and hence changes the crystallinity of the drug. Scatte

ring angle is taken as a parameter for the determination of XRD pattern. Appearance or disappearance of a new

peaks, sharpening of the existing peaks or shifting of certain peaks indicates the complex formation. The method can be possible to detect inclusion complexation in the solid state. The liquids have no diffraction pattern, when complexed with nanosponges shows a diffraction pattern purely different from an uncomplexed nanosponges. This variation in the diffraction pattern shows complex formation. In case of solid drug molecules the diffraction pattern of expected complex is compared between the diffractogram of the mixture of drug and polymer molecules. The interaction between the molecules can be identified and an exact geometrical relationship can be recognised.

12. Thermal analysis

Thermoanalytical methods are used to determine whether the drug substances subjected to any change before thermal degradation of the prepared delivery system. The techniques like differential scanning calorimetry (DSC), differential thermal analysis (DTA), thermogravimetric analysis (TGA) are extremely used. Broadening of existing peaks, shfting and appearance or disappearance of certain peaks can be observed by DTA and DSC thermogram. The influence of temperature on the properties of nanosponges due to melting, decomposition, evaporation, oxidation or polymorphic transition indicating complex formation. The disappearance of melting peak in the crystalline structure of the drug in the DSC thermogram is an indication of molecular dispersion of drug within the polymer. There is a significant difference between the melting peak of complexes and the drug which indicates its interaction with the nanosponge structure.[5]

13. **Raman spectroscopy**

Raman spectroscopy is a useful technique in describing the behaviour of cyclodextrin-nanosponges, When they change from the dry to swollen state. The width, intensity as well as wave number of the Raman peaks are sensitive to the conformational changes of molecules and to intermolecular interactions. Hence it can be used for studying the molecular structure. It also provide an information about the state of water and the solute dissolved inside the nanoporous structure. By analysing the vibration modes of decoupled OH and CH groups from bulk water background, the dynamics of hydration can be examined.

14. Moisture analysis

It is used to confirm the non-hygroscopic nature of nanosponges and their ability to retain the crystalline structure during absorption and desorption of moisture. For this purpose, dynamic vapour sorption studies are conducted.

15. NMR spectroscopy

The technique is used for assessing the chemical environment of the hydrogen and carbon atoms. ^{13}C NMR, ^{1}H NMR, 2D-NMR and high resolution magic angle spinning NMR techniques have been used for studying the structure of cyclodextrins nanosponges. The nanosponge structure can be predicted from the characteristic peaks nd splitting pattern of the hydroxyl and carbonate groups. The changes in chemical shift values (δ) in NMR is occurs due to the exchange of proton between the reacting species. It indicates the formation of nanosponges. Using the high resolution magic angle spinning (HR MAS) NMR, the diffusion coefficients of dissolved solutes and water within the nanosponges can be measured. The individual signals on the NMR time scale and presence of two states of water confirms the existence of two different molecular environments.

16. Stability studies

The nanosponges have been subjected to stability studies under accelerated conditions and photodegradation studies. The formulation is evaluated periodically for 3 months. The changes on physical appearance, size and nature of drug are studied. The photodegradation study is carried out under uv-lamp for 1 hr stirring under dark. The nanosponges are placed at a distance of 10 c.m from the lamp. The sample is withdrawn and analysed by HPLC.[6]

Applications of cyclodextrin nanosponges

Cyclodextrin based nanosponges have many applications in the pharmaceutical sector. It can be used to encapsulate various drugs and provide controlled release of drug for long time. It is an efficient carrier having number of advantages such as increased product performance, improved product elegancy, extended release, non irritating, high physical, chemical and thermal stability…Due to the porous nature, these are able to adsorb flavours and hence unpleasant flavours can be masked. These are solid particles and can be formulated as different dosage forms such as oral, parenteral, topical or inhalation dosage forms. It has also wide applications in environmental control, in the field of agrochemistry and as diagnostic agent. The following are some specific applications of cyclodextrins based nanosponges.[7]

1. Improvement of drug solubility

Cyclodextrins based nanosponges are very good in improving the solubility and dissolution rate of poorly soluble drugs. The cross-linking and cavities formed in the structure improves interaction with the drug molecules and get solubilised in the nanocavities. The formation of inclusion complexation with cyclodextrins increase drug solubility by reducing the drug crystallinity. The formed complex hides the hydrophobic groups in the interior of the cavity and hydrophilic groups exposed to the environment hence increase the solubility of the molecule. It is also reported to be as release enhancers even when there is no complexation.

2. Modulating drug release

The major drawback of frequent administration of conventional dosage forms can be overcome by modified drug release dosage forms like nanosponges. The drug loaded in the cross linked nanosponge structure are

retained and slowly released over time. Both hydrophilic and hydrophobic cyclodextrins nanosponges may provide sustained release for water soluble drugs.

3. Drug delivery

Due to the presence of spherical shape and nanometric in size it can be formulated as different dosage forms like tablets, capsules, aerosols, topical and parenteral preparations. By dispersing the nanosponge complex in to a matrix of excipients, tablets and capsules can be prepared. Sterile water, saline or other aqueous solutions are used as carrier for nanosponges in parenteral administrations. It can also be incorporated in to topical hydrogel for topical drug delivery system.

4. Protein delivery

There are several challenges associated with protein administration. Such as their property to undergo denaturation, aggregation, short half life, enzymatic degradation, high molecular mass and poor bioavailability. Cyclodextrin nanosponges can encapsulate proteins and peptides, hence improves their stability and pharmacokinetic properties.

5. Gas delivery

The nanosponge formulations has the ability to store and release gas molecules slowly over time. The gases play an important role in diagnostics and treatments. Various pathologies are results from the deficiency of adequate oxygen supply. β – cyclodextrin nanosponges can act as a reservoir for huge amounts of gases such as co_2, o_2 and 1-methyl cyclopropene.

6. Nanosponges as diagnostic tools

β – cyclodextrin is commonly used for the production of various materials for diagnostic applications. The properties offered by cyclodextrin nanosponges such as high biocompatibility, prolonged blood circulation time, uniform size distribution for permeability and easy access to the target make them excellent for use as diagnostic agent.

7. Cosmetics

Nanosponges has a number of applicatiions in cosmetic industry. Nanosponges provide a good protection for cosmetic ingredients which are susceptible to photodegradation. It can entrap and prolong the release of volatile oils. It can also adsorb the bad smell from the body produced by sweating. It can release the volatile ingredients slowly hence provide a prolonged fresh feel in oral cosmetic. It can also be used in products such as rouge or lipsticks to provide a long lasting effect.

Other applications of nanosponges

Cyclodextrin based nanosponges can be used for the purification of water. It can adsorb organic and inorganic substances and remove dissolved pollutants from the contaminated water. Cyclodextrins nanosponges can also used as prosthesis and implants to get the controlled release of antibiotics for reducing the risk of infection during surgery. It has been used in the selective separation of inorganic electrolytes by applying the principle of size exclusion chromatography.[8]

CONCLUSION

Cyclodextrin based nanosponges are hyper cross linked polymeric carriers, which can entrap both hydrophilic and hydrophobic drugs by forming inclusion and non-inclusion complexes. The properties such as encapsulation efficiency, biocompatibility and solubilisation capacity for different types of molecules make it as a versatile drug carrier system. The desired particle size and release rate can be achieved by varying the cross linker to polymer ratio. They can be administered by oral, topical and parenteral routes to get at the target site. It has various applications in the field of pharmacy, medicine, cosmetics, bioremediation processes and environment. In conclusion, cyclodextrins nanosponges are a versatile nanocarrier for the delivery of drug molecules in nanomedicine.

REFERENCES

1. Anandam S, Selvamuthukumar S. Fabrication of cyclodextrin nanosponges for quercetin delivery: physicochemical characterization, photostability and antioxidant effects. Journal of material science, 49, 2014, 8140-8153.
2. Shende P, Pawar S, Trotta F. Diversity of beta-cyclodextrin based nanosponges for transformation of actives. International Journal of Pharmaceutics, 565, 2019, 333-350.
3. Shringirishi M, Prajapati s k, Mahor A, Alok S, Yadav P, Verma A. Nanosponges: A potential carrier for novel drug delivery-a review. Asian Pacific Journal of Tropical disease, 2014, 519-526.
4. Shende P, Deshmukh K, Trotta F, Caldera F. Novel cyclodextrin nanosponges for delivery of calcium in hyperphosphatemia. International Journal of Pharmaceutics, 456, 2013, 95-100.
5. Jain s, Suresh S, Singh C, Devasari N, Kushwah V, Trotta F et al. Potential of erlotinib cyclodextrins nanosponge complex to enhance solubility, dissolution rate, in vitro cytotoxicity and oral bioavailability. Carbohydrate polymers, 137, 2016, 339-349.
6. Rastegar R, Javar H A, Khoobi M, Kelishadi DP, Yousefi G H, Doosti M et al. Evaluation of a novel biocompatible magnetic nanomedicine based on beta cyclodextrin loaded doxorubicin-curcumin for overcoming chemoresistance in breast cancer. Journal of Artificial cells, Nanomedicine and Biotechnology, 46, 2018, 207-S216.
7. Swaminathan S, Trotta F, Cvalli R, Tumbiolo S, Bertinetti L, Coluccia S. Structural evidence of differential forms of nanosponges of beta cyclodextrins and its effect on solubilisation of a

model drug. Journal of Inclusion Phenomena and Macrocyclic Chemistry, 76, 2013, 201-211

8. Dharshana J, Amrita B, Tejashri G. Cyclodextrin based nanosponges for pharmaceutical use: A review. Journal of Acta Pharmaceutica, 63, 2013, 335-358.

9. Allahyari S, Trotta F, Valizadeh H, Jelvehgari M. Cyclodextrin based nanosponges as promising carriers for active agents. Journal of expert opinion on drug delivery. 6, 2019, 467-479. Doi:10.1080/17425247.2019.1591365.

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