Current Approaches and Pharmaceutical Applications of Colloidosome Drug Delivery Systems

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ABSTRACT

Recently a number of lipid based systems like lipospheres, liposomes, niosomes, ethersomes, and transferosomes have been developed. The purpose of this review article on colloidosome drug delivery was to compile the focus on the types, properties, fabrication techniques, characterization and stability of colloidosomes. This system also solves the problem of insolubility, instability, rapid degradation and is widely used in specialized areas like protein delivery, gene delivery, targeting to the brain and tumor targeting. In a series of vascular systems, colloidosome represents an advanced tool in drug delivery. Colloidosomes are an emerging vesicular system in drug delivery. Vesicular drug delivery reduces the cost of therapy by improved bioavailability of medication, especially in the case of poorly soluble drugs. Colloidosomes have a great encapsulation efficacy with a wide control over size, permeability, mechanical strength and compatibility.

KEYWORDS: Colloidosomes; emulsion droplets; fused colloidal particles; microcapsules

Introduction

Colloidosomes are microcapsules characterized by a coating, or shell composed of self-assembled colloidal particles that can range in size from nanometers to microns (Dinsmore et al. in 2002). As a result, the surface porosity, namely, the size of the ‘voids’ or surface pores, can range over similar orders of magnitude (see Figure 1). In the past three decades, a lot of advancement in drug delivery system has been made. As a result, new techniques have developed in drug delivery systems (Chein et al., 1982). These techniques are capable of controlling the rate of drug delivery, sustaining the duration of therapeutic activity and targeting the delivery of a drug to a cell/tissue (Chein et al., 1989). This advancement led to the development of several novel drug delivery systems of medication and provides a number of therapeutic benefits. The capsules are fabricated by the self-assembly of colloidal particles onto the interface of emulsion droplet. These assemble particles are locked together to form hollow, elastic shells the resultant structure is known as colloidosomes.

A capsule surface is composed of one packed layer of colloidal particles, linked together to form a solid shell. The interstices between the particles form an array of uniform porous whose size is easily adjusted over the parameter to micro meter scale to control permeability. The colloidosomes are produced by assembly of polymer latex colloidal particles into shells around water-in-oil emulsion drops followed by partial fusion of the shell and centrifugal transfer into water to yield stable capsules in which the shell permeability can be controlled by adjustment of partial fusion conditions. Further advantages could be obtained from colloidosomes and capsules with a non-spherical geometry, or with multiple compartments (Daeyeon Lee et al., 2009). Size ranges of colloidosomes from 5nm to several microns in diameter.

Colloidosomes have several advantages over their liposome and polymersome delivery agent counterparts. These include:

1. Mechanical stability of the colloidal shell (Dinsmore et al., 2002).
2. Control of shell pore size (Kim et al., 2007).
3. The ability to trigger release in response to external fields or changes in environmental conditions.
4. 100% encapsulation efficiency, eliminating the need for complex recovery steps of expensive encapsulants.
5. Production capability of highly monodisperse colloidosomes, in large numbers by using microfluidic devices (Kim et al., 2007).
6. Possible tracking in vivo by doping the shell with metallic particles observable in such methods as X-Ray or MRI.

Fig. 1 Colloidosome structure.
Above are sketches of colloidosomes, namely, hydrogel cores coated by a shell composed of colloidal particles (left), and the surface pores formed in the shell by the packing of the particles on the hydrogel surface (right).

**Classification of Colloidosomes**

Colloidosomes are classified on the basis of emulsion used (Gibbs et al., 1999; Saraf et al., 2011). They are classified as follows:
1. Water-in-oil emulsion based colloidosomes
2. Oil-in-water emulsion based colloidosomes
3. Water-oil-water emulsion based colloidosomes.

The colloidosomes are also classified on the basis of the nature of colloids. They are classified as follows:
1. Aqueous or oily gel core colloidosomes
2. Hairy colloidosomes
3. Nanoparticle colloidosomes
4. Layer by layer colloidosomes
5. Non-spherical colloidosomes

**Core Material**

The core material can include a polymer gel. It consists of a cross linked network of long polymer molecules (Kim et al., 2009). This can provide physical strength. The gel can include water (i.e., a hydro gel) and/or organic liquids. Swelling and deswelling in water is a characteristic of hydro gel.

The gel can be formed as a particle, such as a spherical particle, an ovoid particle, a cylindrical particle, or some other three-dimensionally shaped particle. The gel particle can have at least one dimension that is microns to millimeters in size. The particle size of the core material is dependent on the intended application of the resultant colloidosomes. Generally, the core material diameter is in the range of about one or more microns to millimeters.

Conventional methods of preparing a core particle include capillary-based micro fluidic techniques, precipitation polymerization techniques and inverse suspension polymerization techniques (Angew et al., 2005).

The gel possesses at least one property that exhibits an attractive interaction with the colloidal particles. For example, the gel can possess electrical charges, magnetic materials, and the like that are capable of attracting the colloidal particles toward the core. Magnetic material can be included in the gel that attracts colloidal particles to the core. For example, the gel can entrain certain magnetic materials (as solids or in solution) within the cross linked network that are attractive to magnetic metal particles that serve as the colloidal particles.

**Factors Affecting the Property of the Gel or Core Material**

**External stimulus**

The core material is responsive to an external stimulus that causes a change in a property in the core material (Saraf et al., 2011). The properties of the core material are altered upon application of external stimuli, such as a temperature, electric field, magnetic field, pH and ionic strength.

The core material responds to the external stimulus by undergoing a volumetric change. When a gel undergoes a dramatic change in volume, it is sometimes referred to as a phase change. At the extremes, the gel particle can change from a fully expanded (swollen) gel state to a collapsed (deswollen) solid state.

**Temperatue**

Temperature responsive phase transition material (or “thermally responsive” material) undergoes a phase transition and/or alters the size of the material in response to thermal energy. The core material i.e. synthetic polymers may change its volume from an expanded state at a lower temperature to a collapsed state at higher temperatures. Examples of synthetic polymers are starch, xanthan gum, agar, gelatin, hyaluronic acid, Arabic acids and alginate.

**pH**

pH responsive phase transition material (or “pH responsive” material) undergo a similar phase transition and/or alter the size of the material in response to a change in pH. For example, the core material may change its volume from an expanded state to a collapsed state at higher or lower pH values. pH sensitive hydro gels may include polypeptide hydro gels made of hydrophobic (e.g., leucine) and hydrophilic (e.g., glutamine) amino acids, poly(N-isopropylacrylamide), poly(acrylamide), and poly(acrylic acid).

**Electrical effect**

Electrically-responsive material can include materials containing electrically responsive elements that can deform or alter the size of the material in response to an applied electric field. For example, the core material may change the volume of the gel from a collapsed state to an expanded state upon application, removal, increase, or decrease of an electric field. Exemplary materials that can be utilized include N-isopropyl acrylamide, vinyl alcohol, vinyl amine, acrylic acid, gelatin, urethane and vinylsulfonic acid.

**Colloidal Particles**

The colloidal particles can include a metal, a semiconductor, a polymer, an inorganic material, nanometer sized particles and/or micrometer sized particles (Kim et al., 2009). The colloidal particles have a particle size that is smaller than that of the core material. The colloidal particles possess an attractive interaction with the core material. For example, the colloidal particles possess electrical charges, magnetic properties, and the like that are attractive to the core material. The colloidal particles may be negatively charged when the core material is positively charged.

Types of colloidal particles are:
1. Semiconductor colloidal particles include silicon, germanium, gallium arsenide, and cadmium selenide.

2. Polymer colloidal particles include polystyrene, polymethyl methacrylate, poly (ε-caprolactone), poly (lactic acid), and poly (lactic acid-co-glycolic acid).

3. Inorganic colloidal particles include gold, silver, copper, cobalt, palladium, platinum, manganese-zinc, nickel-zinc, iron-platinum, silica, Titania, iron oxide, zinc oxide, and nickel oxide.

4. Magnetic colloidal particles include gold, silver, copper, iron oxide, manganese-zinc, nickel-zinc, nickel oxide, cobalt, and iron-platinum, CoFe₂O₄. The colloidal particles can possess magnetic properties that can be attracted to a gel having a magnetic material. For example, the gel can contain certain magnetic particles within the cross linked network and the colloidal particles can include a magnetic material.

5. The colloidal particles can possess additional properties, such as fluorescence that are suitable for certain biological chemical uses. For example, the colloidal particles might be sensitive to certain diagnostic tools (e.g., MRI, ultrasound, x-ray, etc.) to determine the location of the colloidosomes, e.g., colloidosomes that encapsulate a drug or other therapeutic agent, in the human body.

**Tuning the Properties of the Colloidosome**

When the colloidosomes can be exposed to one or more external stimuli it alters the properties of the colloidosomes (Kim et al., 2009). For example, permeability, mechanical properties and morphology of the colloidal layer can be altered.

The packing uniformity, packing density and particle layering on the core material can be altered by changing the size of the core materials after assembly of the colloidal particles around it. In this manner, permeability of a colloidosome can be tuned by changing the morphology of the colloidosome. As used herein, “permeability” refers to permeation or movement of one or more species into and out of the colloidosome.

Tuning the colloidosome may occur during the manufacturing of the colloidosome, for example, to increase the packing density of the colloidal particle shell and thereby reduce the permeability of the colloidosome.

Tuning the colloidosome may occur during use, for example, to increase volume size of the gel core, thereby reducing the packing density of the colloidal particle shell and thereby increasing the permeability of the colloidosome.

Colloidosomes having a thermally responsive gel as the core material also have colloidal particles that are loosely spaced on the surface of the gel core. Upon assembly, the colloidal particles are attracted to the gel core, but may be assembled in a random manner where the interparticle distances between the colloidal particles are relatively high. The permeability into and out of the gel is expected to be very high due to the large spacing between the colloidal particles. Due to the large spacing, both large and small molecules may migrate into and out of the colloidosome.

Upon application of heat, the gel can shrink leading to a reduced spacing between colloidal particles. In one embodiment, the gel can shrink to about 90%, or about 80%, or about 60% or up to 20% of its original size. The closer spacing of the colloidal particles reduces the interstitial distances in the colloidal layer and lowers the permeability of the colloidal particle layer. It may be expected that higher molecular weight particles do not move as readily across the colloidosome boundary layer.

![Fig. 2 Scanning electron microscope image of a colloidosomes encapsulating oil in aqueous solution.](image-url)
Even further reduction in permeability may be achieved by further heating the gel or by the application of yet another external stimulus that serves to further reduce the core material volume. In some embodiments, the gel can shrink to about 80%, or about 70%, or about 50% or up to 10% of its original size. Further crowding of the colloidal particles around the reduced surface area of the core material results in reduced spacing between particles and/or overlap of particles on the core surface and is expected to exhibit lower permeability.

**Method of Preparation**

**Preparation of emulsion based colloidosomes**

1. **Water-in-oil emulsion based colloidosomes**

An aqueous solution is emulsified in oil in presence of colloidal particles to produce water-in-oil emulsion. Particles are adsorbing onto the surface of the droplets to reduce the surface energy. These particles are subsequently locked together by addition of polycations, by Vander Waals forces or by sintering the particles. These colloidal particles help in producing water-in-oil stabilized emulsion (Gibbs et al., 1999, Cayre et al., 2004). Then water in oil based colloidal dispersion is obtained. The two ways to transfer them into water are centrifugation approach and filtration approach.

In the centrifugation approach, the obtained colloidosomal dispersion is diluted with non aqueous phase (ethanol, dodecane) and then allowed to centrifuge to separate them from the supernatant. The obtained water core colloidosomes are washed with ethanol and water and redispersed in water.

The second transfer approach is to filter the oil suspension on the hydrophobic millipore membranes. At the end of filtration, a small amount of water containing a small proportion of ethanol is added to top of the membrane, removing the oil interface and resuspending the colloidosomes in water. This water core colloidosome (W/O emulsion based) seems to be a suitable encapsulating agent for the drugs, dye, and fragrances because of its mechanical resistance of shell, the tunable porosity, and the easy mass transfer potential due to absence of a chemical barrier (Velev et al., 1997).

2. **Oil-In-water emulsion based colloidosomes**

In this type, oil is emulsified in an aqueous solution containing particles in presence of a surfactant to produce oil-in-water emulsion. This colloidal particle in the presence of surfactant is used to stabilize the oil/water interface. This introduces an electrostatic driving force to take the colloidal particles to the emulsion interface.

Then Oil-in-water emulsion based colloidosomal dispersion is obtained. The obtained colloidosomal dispersion is added to the non-aqueous phase (ethanol) and allowed to centrifuge to separate them from the supernatant. The obtained oil core colloidosomes are washed with ethanol and finally redispersed in water (Cayre et al., 2004).

3. **Water-oil-water emulsion based colloidosomes**

A pendant drop of an aqueous suspension of latex particles is formed in an oil phase. A closely packed particle monolayer is adsorbed on the drop surface by multiple infusion and withdrawal of the particle suspension through the capillary in the oil phase. Finally, the pendant water drop in oil, densely coated with adsorbed particles, is transferred through a planar oil-water interface (free of particles) to form a giant pendant colloidosome, which consists of a spherical water/oil/water film supported by latex particles possibly bridging both surfaces. Nanoparticle colloidosomes with selective permeability are generated by using water-in-oil-in-water (W/O/W) double emulsions as templates (Lee et al., 2008).

**Preparation of colloidosomes basis of nature of colloids:**

1. **Aqueous or oily gel core colloidosomes**

The basics of the method are illustrated in Fig. 3 and involve the following three stages (Cayre et al., 2004):

- A hot aqueous solution of agarose is emulsified in oil in the presence of solid particles to produce water-in-oil emulsion stabilized by the solid particles and the system is cooled to set the agarose gel.
- The obtained suspension of aqueous gel microcapsules is diluted with ethanol and centrifuged to separate them from the supernatant.
- The microcapsules are washed with ethanol and water and redispersed in water. This technique allowed us to prepare giant colloidosome microcapsules of diameters varying between several tens to several hundreds of micrometers. The average size of the colloidosomes can be varied by changing the size of the solid particles.

2. **Hairy colloidosomes**

- The basics of the method are illustrated in Fig. 4 & 5 and involve the following three stages (Panav et al., 2005 and Paul et al., 2004):
  (i) A hot aqueous solution of agarose is emulsified in oil in the presence of rod like polymeric particles to produce a stable water-in-oil emulsion stabilized by the solid particles, and the system is cooled off to set the agarose gel.
  (ii) The obtained suspension of aqueous gel microcapsules is diluted with ethanol and centrifuged to separate them from the supernatant.
  (iii) The microcapsules are washed with ethanol and water and redispersed into water. This technique allows preparation of colloidosome microcapsules of diameters varying between several tens to several hundreds of micrometers. The function of the gel cores is to support the particle shell and to give the microcapsules enough stiffness to be separated from the oil phase by centrifugation.
3. Nano particle colloidosomes
Nano particle colloidosomes are prepared by using water in oil in water double emulsion as a template to provide robust and monodisperse semi-permeable nanoparticle colloidosomes with precisely tuned structure and compositions (Lee et al., 2008). Monodispersed water in oil in water double emulsions with core/shell geometry is generated using glass capillary micro fluidic devices. Hydrophobic silica nanoparticles are dispersed in
the continuous phase (water in oil emulsion). These particles self-assemble at the interface between the two immiscible liquid phases and form a colloidal shell/oil shell, to stabilize the droplets and ultimately become the colloidosome shells upon removal of the oil solvent. Then they were transferred to an aqueous phase either by centrifugation or repeated washings, to create efficient colloidosomes with particles ranging from 5nm to several microns in diameter. Size and dimensions of colloidosomes are controlled by independently controlling the flow rates of each fluid phase. Figure 5 explains the following features:

A. Micro-capillary device for double emulsion generation
B. Formation of Nano particle colloidosomes
C. Optical microscope image of double emulsion
D. Fluorescence microscope image of double emulsion

4. Layer by layer colloidosomes
This process is used to encapsulate enzymes by using biocrystals as templates for the deposition of polymer multilayers, subsequent enzyme solubilization and release, and the formation of hollow polymer capsules (Caruso et al., 2000). Polyelectrolyte layers are deposited stepwise onto the crystals by making use of the surface charge reversal that occurs upon adsorption of each layer. Each polyelectrolyte layer deposited bears an opposite charge to that already adsorbed. The unadsorbed polyelectrolyte is removed by repeated centrifugation/wash redispersion cycles before the next layer is deposited. Solubilization of the enzyme inside the polymer capsule by exposure to solutions of pH>6 or acidic solution with pH<4 results in a morphology change of the polymer capsule. Release of the enzyme by rupturing the polymer capsule is achieved by exposure to solution of pH >11. Exposure of the encapsulated enzyme to an oxidizing solution results in decomposition of the enzyme which then is expelled from the interior through the polymer walls, leaving behind the hollow polymer capsule that originally encapsulated the enzyme.

5. Non-spherical colloidosomes
The generation of nonspherical colloidosomes with multiple compartments, we use glass capillary micro fluidics to prepare W/O/W double emulsions with different morphologies (Daeyeon Lee et al., 2009). These double emulsions have a different number of internal aqueous drops in the oil drop. Hydrophobic SiO2 nanoparticles, suspended in the oil phase, and poly vinly alcohol (PVA), dissolved in the continuous aqueous phase, stabilize the double emulsions. The nanoparticles in the oil phase eventually become the shell of colloidosomes upon the removal of the oil. During the oil removal, the internal W/O interface retains in spherical shape whereas the outer O/W interface deforms; this process leads to the generation of non-spherical colloidosomes with multiple compartments.

Fig. 5 Schematics of the method for formation of nano particle colloidosomes.
Characterization of Colloidosomes

The characterization of colloidosomes including particle size measurement, zeta potential measurement, gravitational separation, small droplet concentration, optical microscopy, and surface area are as follows (Saraf et al., 2011):

**Particle size analysis**

The particle size distribution of the emulsions is measured using a laser diffraction particle size analyzer. This instrument measures the angular dependence of the intensity of light scattered from a stirred dilute emulsion. To avoid multiple scattering effects the emulsions were diluted with buffer prior to making the light-scattering measurements. The emulsions were stirred continuously throughout the measurements to ensure the samples were homogenous. Dilution and stirring are likely to disrupt any flocculated droplets or break up any free oil into droplets, hence particle size data on highly flocculated or coalesced samples should be treated with caution. A refractive index ratio of 1.08 is used by the instrument to calculate the particle size distributions. The size of the latex particles was measured by dynamic light scattering and by differential scanning calorimetry (Gu et al., 2007).

**Zeta potential measurements**

Emulsions are diluted using a buffer solution prior to analysis to avoid multiple scattering effects. Diluted emulsions are injected directly into the measurement chamber of a particle electrophoresis instrument. The zeta (ζ) potential is then determined by measuring the direction and velocity of droplet movement in a well-defined electric field.

The zeta potential measurements are reported as the mean and standard deviation of two separate injections,
was then placed on a microscope slide, covered by a
ensure that they were homogeneous. A drop of emulsion
gently agitated in a glass test tube before analysis to
determined using optical microscopy. Emulsions were
adsorbing to the surfaces of the large droplets is
aggregation in an emulsion (Gu et al., 2007).

Gravitational separation

Emulsions are poured into glass tubes (100 mm height, 16 mm internal diameter), covered and stored at room temperature for 24 h before the heights of any separated layers were measured manually using a ruler. Different kinds of layers are observed depending on emulsion type, e.g., a clear oil layer at the top, an opaque emulsion layer in the middle and/or a transparent serum layer at the bottom. The serum layer is to be the sum of the turbid and transparent layers. The total height of the emulsion (HE) and the height of the serum layer (HS) are measured. The extent of creaming is characterized by a creaming index =100X (HS/HE). The creaming index provided indirect information about the extent of droplet aggregation in an emulsion (Gu et al., 2007).

Free small droplet concentration

The concentration of small droplets that is not adsorbing to the surfaces of the large droplets is determined by using a gravitational separation and turbidity method. The composite emulsions are allowed to stand at room temperature for 24 h, which meant that any large droplets creamed to the surface while the small droplets remained in the lower layer. A sample of the lower phase containing the free small droplets is then collected and the turbidity is measured (Gu et al., 2007).

Optical microscopy

The microstructure of selected emulsions is determined using optical microscopy. Emulsions were gently agitated in a glass test tube before analysis to ensure that they were homogeneous. A drop of emulsion was then placed on a microscope slide, covered by a cover-slip and observed at a magnification of 40X using an optical microscope (Gu et al., 2007).

Applications of colloidosomes

Colloidosomes have been widely used as encapsulating agent with endless application in biomedical, pharmaceutical and cosmetic industries with strong interest in encapsulation and controlled release in the field of drug delivery, cosmetic delivery, food delivery, LCD display devices, polymer blends, paints, catalytic material or even living cells (Saraf et al., 2011).

The functional biopolymers (colloidal particles) can further act as a targeting agent, wherein the functional biopolymer acts to direct the colloidosomes to a desired location. Accordingly, the resulting colloidosomes can provide an integrated mechanism for targeted delivery of the colloidosomes and potentiate controlled release of the encapsulated material, and biocompatibility with the subject.

It exhibited 80% decrease in volume when actuated; thus, they can be of immense potential in applications that require targeted pulsed-release of active materials (Lee et al., 2008).

Colloidosomes may be used for the following range of therapeutic and pharmaceutical applications (Vyas et al 2002):

- Colloidosomes as drug/protein delivery carrier
- Colloidosomes for controlled and sustained drug release
- Colloidosomes for enhanced drug solubilization
- Colloidosomes for altered pharmacokinetics and biodistribution
- Colloidosomes in tumor therapy
- Colloidosomes in antimicrobial, antifungal, antiviral therapy
- Colloidosomes in ocular drug delivery
- Colloidosomes in brain delivery
- Colloidosomes in DNA delivery

Conclusions

Colloidosomes that consist of large droplets surrounded by a layer of small droplets can be produced by mixing coarse and fine emulsions containing oppositely charged droplets together. The capsules are fabricated by the self-assembly of colloidal particles onto the interface of emulsion droplets. After the particles are locked together to form elastic to elastic shells, the emulsion droplets are transferred to a fresh continuous phase fluid that is the same as that inside the droplets. The resultant structures called colloidosomes are hollow, elastic shells whose permeability and elasticity can be precisely controlled. They have a wide range of applications in products that are consumed by humans, ex-beverages, food, pharmaceuticals, flavors, fragrances, and cosmetics industries. It may be possible to reduce their susceptibility to gravitational separation to control their stability to environmental stresses, to develop novel controlled or triggered release system, compartmentalize active agents or control certain chemical reactions. The choice of different colloidal particles allows for additional flexibility. A variety of release strategies may be feasible, either through control of their permeability for slow but sustained release, or through control of their rupture stress for shear-induced breakup. This flexibility will allow a wide range of potential applications to be explored.

References


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