

**CUBOSOMES: A REVIEW ON THEIR DESIGN, FUNCTION & APPLICATIONS IN
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ABSTRACT

The effect of nanotechnology on the exponential development of a few investigate ranges, especially nanomedicine, is undeniable. One of the nanocarriers created which has drawn researchers' consideration are cubosomes. Cubosomes are nanoparticles but instep of the strong particles, cubosomes are self-assembled fluid crystalline particles of certain surfactant with legitimate proportion of water with a microstructure that gives special properties of viable intrigued. The disclosure of cubosomes is a one of a kind story and ranges the field of nourishment science, differential geometry, natural films and stomach related forms. One of the most common surfactants utilized to make cubosomes is the monoglyceride glycerol monoolein. Bicontinuous cubic fluid crystalline stage is an optically clear, exceptionally gooey fabric that has a one of a kind structure at the nano meter scale. The word bicontinuous alludes to the division of the two ceaseless but non-intersecting watery locales by a lipid bilayer that is bound into a space-filling structure. Hydrating a surfactant or polar lipid that shapes cubic stage and at that point scattering the solid-like stage into littler particles more often than not frame Cubosomes. They are broadly utilized for different sedate conveyance applications such as verbal, visual, transdermal and chemotherapy medicate conveyance. In this audit, the germane writing of cubosomes with accentuation on speculations of self-assembling, the composition of cubosomes, strategies of preparation, approaches for typifying material, interaction of natural fabric with cubosomes, characterization of cubosomes and medicate conveyance applications will be basically surveyed.

KEYWORDS: Cubosomes, Nanotechnology, Drug Delivery Systems, Design, Preparation, Applications.**INTRODUCTION TO CUBOSOMES**

A drug delivery system (DDS) may be depicted as a gadget that discharges a pharmacological substance at an indicated location in the body at a modified rate to set up a viable concentration at the drug's location of activity. Supported discharge over an amplified period may reduce the require for different measurements, coming about in lower costs and way better persistent compliance. Drug delivery systems are expecting to improve the pharmacokinetic parameters and organic dissemination of the operators to which they are connected, to act as stores for the sedate substances (i.e., sustained-release frameworks), or to do both. Nanocarriers delivered utilizing naturally consistent materials appear differential take-up into the focused on cells/tissues, rendering them valuable for different medicate focusing on strategies. Over the past few a long time we have seen an exponential development in investigate for the advancement of modern medicine-delivery technologies. Numerous solutions, both conventional pharmaceutical items and unused atomic substances, can be conveyed in ways that increment both

their security and effectiveness. Through hereditary designing, more one of a kind and more advanced drugs, such as proteins, have ended up accessible; the transport of these drugs is ordinarily more challenging than that of most conventional drugs, requiring the advancement of novel conveyance frameworks.^[1]

Nanotechnology-based drug-delivery stages have been a consistent subject of investigate for the past a few decades due to their flexible and novel applications in the helpful space. Different nanocarriers based on natural or inorganic materials, such as organic-based liposomes, polymeric micelles, strong lipid nanoparticles, dendrimers, and inorganic derived carriers such as carbon nanotubes, exosomes, mesoporous silica particles, cubosomes, hexosomes, and so on, have been created for pharmaceutical applications. These novel medicate carriers are colloidal scatterings in which the restorative moiety is either adsorbed onto or conjugated with the carrier frameworks. Site-specific, supported, or naturally responsive sedate discharge, moved forward steadiness, improved bioavailability, and decreased

harmfulness can be gotten through the tweak of their morphological and surface characteristics. Diverse sorts of atoms, such as hydrophilic, hydrophobic, or amphiphilic compounds, can be stacked into these frameworks based on the sort of substrate utilized for their generation. Among these, amphiphilic lipidic materials are utilized to define a vesicular framework that can in this way typify either hydrophobic or aqueous-soluble medicate substances. From them, the nonlamellar turned around lyotropic fluid gems scattered in a fluid medium such, as micellar spasmodic cubic stages, hexagonal and bicontinuous mesophases, have been escalation explored. Nanostructured lipid-based carriers, such as hexosomes and cubosomes, are shaped by means of the stabilization handle due to electrostatic intelligent or to steric components amid the self-assembly arrange of amphiphilic lipids, shaping non-lamellar stages in the nearness of overabundance water and a stabilizing agent. Nanosized fluid lyotropic crystalline particles empower the stacking of differing hydrophilic and hydrophobic substances effectively; be that as it may, they can moreover be created into nano-engineered conveyance frameworks with controlled-release properties to target organ frameworks and tissues and make strides cell saturation specifically.^[2]

Structurally, cubosomes are liquid-crystalline nanostructures composed of different amphiphilic lipid components in particular extents. These cubic structures contain banded bicontinuous lipid bilayered layers in a honeycomb-like 3D organize that are isolated into double internal fluid pathways utilized by various bioactive components, such as pharmacological operators, amino acids, and proteins. Subsequently, they are considered to be energizing vehicles for different organization courses due to their unconventional qualities, counting thermodynamically steady structures, bio adhesiveness, the capacity to typify hydrophilic, lipophilic, or amphiphilic compounds, as well as their potential for controlled medicate discharge by means of fabrication. Cubosomes are unmistakable, nanovesicles of bicontinuous cubic structures which are defined by scattering of fluid crystalline cubic totals in fluid media and they are characterized by tall surface range and indistinguishable microstructure to its parent cubic totals. They are defined by certain amphiphilic lipids as glycerol monooleate (GMO) and phytantriol (PHYT), which have the capacity to self-assemble in water to frame cubosomes. They include a structure comparative to honeycomb (cavernous) structures with an estimate run of 100 nm to 500nm.^[3]

Cubosomes are picking up uncommon intrigued as a special sedate conveyance framework and as of late they have been utilized in visual, dermatological, verbal and cancer treatment. Really, there is an auxiliary similitude between cubosomes and polymeric micelles and both are commonly utilized in different sedate conveyance applications. In any case, polymeric micelles are shaped by an amphiphilic polymer which self-assembled in

water into core-shell structure when its concentration is over the basic micelle concentration. When a hydrophobic medicate is included, it can be joined inside the hydrophobic center of micelle whereas hydrophilic bioactive particle is consolidated in the external hydrophilic shell of the micelles.^[4]

Advantages^[5]

- They are able to typify distinctive medicate particles with hydrophilic, hydrophobic and amphiphilic properties.
- They have provided a hopeful approach for bioavailability upgrade for poorly-water solvent drugs.
- They can be arranged by straightforward techniques.
- They are composed of biodegradable lipid.
- They can protect the consolidated medicate from physical and chemical degradation.
- The nanovesicles shaping lipid (GMO) give a saturation enhancer amid cubosomes infiltration through corneal and skin layers.
- They have tall sedate loads due to their tall inner surface.
- They render the bioactive medicate particles with focused on and controlled discharge.

Disadvantages^[6]

- Challenging in huge scale generation due to the tall thickness of cubic stage.
- They have moo capture proficiency for water-soluble medicate particles due to their tall water substance interior their structure.

Their little sizes, extending from 1 μ m to 10 μ m, empower them to effectively enter tissue frameworks and convey drugs straightforwardly into cells at a focused on location of action. Lipid-derived cubosomes have more inner divisions than liposome vehicles, which is a basic edge that empowers way better epitome success. Hydrophilic macromolecules such as ovalbumin, brain-derived neurotrophic factor (BDNF), and protein antigens have been appeared to have tall entanglement efficiencies. Anticancer (paclitaxel), antiviral (efavirenz), and anti-inflammatory (flurbiprofen) helpful specialists, along with cancer prevention agents like curcumin, have essentially expanded bioavailability that is inferable to the conservation and conveyance of the facilitating cubic fluid crystalline phases. Entrapment proficiency of curcumin in PHYT-based cubosomes was more noteworthy than that in liposomes, which may be due to the more profound penetration of the sedate inside the bilayer's hydrophobic parcel. Besides, MO-based cubosomes illustrated the most noteworthy enhancement in cytotoxicity in B16F1 cell lines, implying their potential in anticancer therapy. Cubosomes were way better than hexosomes in conveying phenytoin in vivo over 60 min to the brain in rats, relating to the brain-to-plasma proportion and brain

concentrations, in spite of the fact that the cytotoxicity was comparable for both.^[7]

Lipidic System

A sedate conveyance system's fundamental reason is to securely transport a compound in the body until it comes to the focused on natural location. At the location it ought to evoke a viable restorative reaction. To accomplish this reason, the frameworks ought to be outlined in arrange to guarantee the dynamic particle bioavailability and pharmacological behaviour, with diminished side effects. When creating a sedate conveyance framework, it is vital to take in thought the organization course (verbal, nasal, transdermal,

intravenous, intramuscular, etc.). A nanocarrier can have particular structures. It can have a polymeric nanoparticle, micelles, liposomes, and cubosomes, the final two being lipid-based systems. Lipid-based frameworks are commonly utilized as mediate conveyance frameworks, particularly due to its capacity to progress solvency and bioavailability of destitute water- soluble compounds. The lipid-base frameworks are shaped by lipid molecules. Lipid atoms have two diverse spaces: a hydrophilic headgroup, and a hydrophobic tail composed of one or more hydrocarbon chains. Atoms with such properties are named amphiphilic, and it is this characteristic that leads to self-assembled structures.^[8]

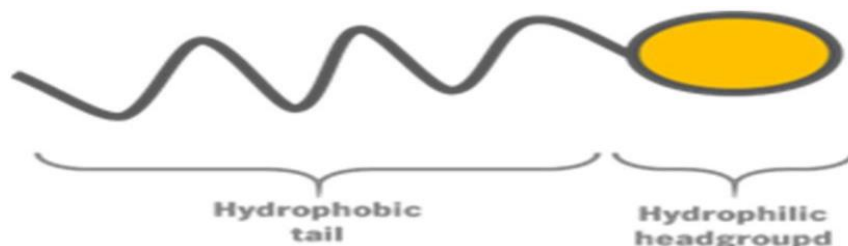


Figure 1: Schematic Representation of Polar Lipid Molecule which Posses Two domain a hydrophilic headgroup (polar) and a hydrophobic tail (apolar).^[8]

Self-Assembly of Lipidic Structures

The self-assembly of lipidic nanoparticles is a result of a prepare called hydrophobic impact. In the nearness of water, the amphiphilicity of the lipidic atoms leads to the arrangement of two inverse powers: one favouring the accumulation of the lipid and the other repulsing it. The accumulation constrain is due to a distinction in the extremity between the hydrocarbon chain and water. When these particles are set in contact with water, the water atoms encompassing the tail of the lipid shape organized systems, diminishing entropy and subsequently expanding the framework free vitality. To recapture the misplaced entropy, the adsorbed water atoms require to be discharged. There are two ways to accomplish this objective: either by the adsorption of

lipidic atoms to an interface, or by the affiliation of hydrocarbon chains of distinctive particles, coming about in structures with moo generally vitality framework. These structures are characterized by a hydrophobic center (hydrocarbon chains) and a hydrophilic surface (polar headgroups). When the tails of diverse lipidic atoms relate, due to the hydrophobic impact, the hydrophilic headgroups get closer. As a result, these polar bunches begin repulsing each other (the repulsing force). Several parameters impact the self-assembly of lipidic structures, specifically the adjust between two inverse powers, the chemical structure of the surfactant, the environment conditions (temperature, pH, weight, etc.), the add up to bulk-phase composition, among others.^[8]

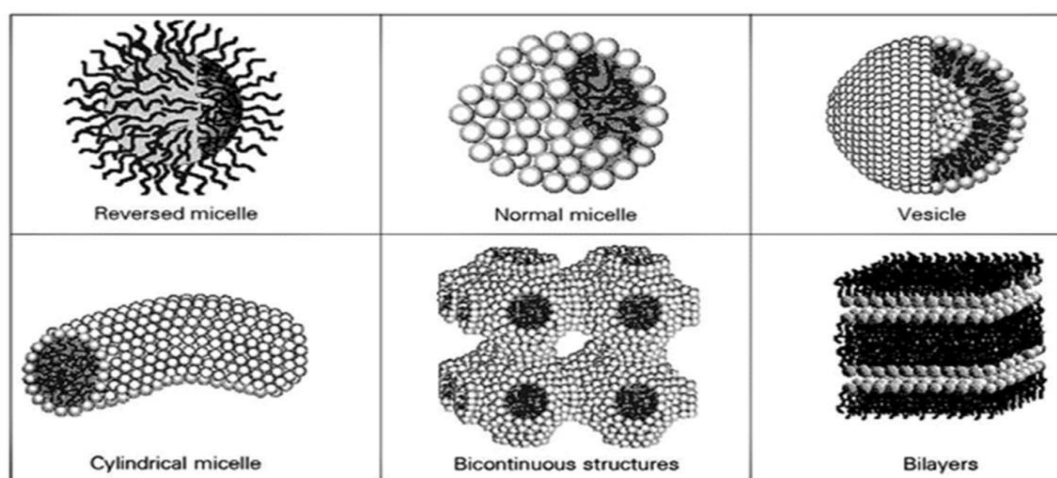


Figure 2: Most common self-assembly structures of amphiphilic molecules, including micelle, vesicle, bilayer, and bicontinuous phases.^[8]

Theories involved in self-assembling of amphilic lipid

There are two main theories which explain the self-assembling.

The principle of opposing forces: Amphiphilic particles when uncovered to a polar dissolvable they are organized in such a way to minimize their free vitality, where the polar dissolvable enters through the amphiphilic particles uncovering the hydrophilic parcels to the watery environment whereas the hydrophobic parcels of the amphiphiles are protected from the dissolvable. In this way restricting powers were created as a result of hydrophobic intelligent happened between the hydrophobic hydrocarbon tails and the hydrophilic head gather of the amphiphilic particles, these intelligent compete together at the interfacial zone. The to begin with drives the affiliation of particles whereas the last mentioned actuates the inverse. This is commonly alluded to as the hydrophobic impact marvel. The hydrophobic holding really varies from van der Waals interaction powers, as they result from diverse intuitive. The hydrophobic holding as already specified happens between the hydrophobic hydrocarbon tails which initiate atomic affiliation together to stow away from water. It is more grounded than the powerless intermolecular van der Waals drive (as hydrogen bond) which happened between iotas as a result of the oppositely polarized electron clouds.^[9]

The principle of packing parameter: The lipid aggregates which may be preferentially formed for a given lipid is best predicted by the principle of packing parameter. It relates the molecular shape and properties of certain lipid to its favoured curvature at the lipid water interface.^[9]

$$P = v/al$$

Where, v = volume of hydrophobic chain, l = Length of hydrophobic chain,

a = Optimal surface area of polar head group,

- The normal micelle, when the critical packing parameter (P) $< 1/2$, in which oil is domain in water (oil in water) with the resulting curvature occurs towards the chain region. According to this value of critical packing, parameter the amphiphiles occupy a cone shape space and hence this conical geometry leads to formation of spheres.^[9]
- Closed lipid bilayer, when the critical packing parameter $1/2 < (P) < 1$, this property derives the lipid molecules to assemble forming closed lipid bilayer structure.^[9]
- Open lipid bilayer (lamellar curvature), when the critical packing parameter (P) = 1 and characterized by zero curvature with identical cross-sections of the polar heads and the lipophilic tails. This phase with no interfacial curvature as the amphiphilic lipids under the critical packing parameter occupy an apparently cylindrical space.^[9]
- Inverted micelles, when the critical packing

parameter (P) > 1 , it is water in oil version and the resultant curvature occurs towards the water region. According to the (P) value, the amphiphiles occupy an inverse cone shape space. This geometry leads to formation of inversed spheres.^[9]

Lipid collecting into a closed lipid bilayer framework comes about in arrangement of distinctive lipid-based nanovesicles which are broadly utilized in pharmaceutical applications this get together depends primarily on the (P) value of the collecting lipid this counting lipid gathering into liposomes which are composed of one or two concentric phospholipid bilayers that are isolated by water compartments, niosomes which are defined from non-ionic surfactant comprising of one or more surfactant bilayers encasing fluid spaces in nearness of film stabilizer as cholesterol and too, lipid might be amassed into cubosomes which confined to certain amphiphilic lipids as glyceryl monooleate and phytantriol that are able to self-assemble into bulk cubic crystallographic structures which are more distant scattered in water in the nearness of reasonable stabilizer coming about in arrangement of our curiously structure "cubosomes".^[9]

Preparation of cubosomes

By and large, there are two fundamental approaches for cubosomes planning, the top-down and bottom-up approaches, both of them require the utilization of appropriate stabilizer such as F127 to avoid cubosomes scattering conglomeration. In any case, steadiness, biocompatibility and ideal medicate discharge stay the primary target in the choice of ideal arrangement method.^[10, 11, 12]

Top Down Approach: The top down-method is the most broadly utilized strategy for cubosomes planning, it includes two fundamental steps. Firstly, blending the cubosomes shaping lipid with an appropriate stabilizer to frame the bulk thick cubic totals. Furthermore, scattering of the created thick cubic totals in watery media by the application of tall vitality as high pressure homogenizer or sonication at last coming about in the arrangement of cubosomes. Fortunately, cubosomes arranged by the top-down strategy are found to be steady against conglomeration up to a year. In any case, this strategy with downsides in expansive scale generation as the arrangement of thick cubic totals require tall vitality input to be scattered into cubosomes, shockingly, these may be a issue when joining of temperature-sensitive bioactive specialists, particularly peptides and protein are required. When the blending temperature is raised to around 80 degree Celsius, the interaction of the hexagonal and the reverse micellar stages takes put, bringing down the net execution of the definition since the coexisting stages affect the estimate conveyance of the particles, its polydispersity list, the stacking of drugs, and other components.^[10,11,12]

Bottom-up approach: This approach is commonly referred to as solvent dilution method, it includes scattering of blend containing cubosomes shaping lipid, the stabilizer and a hydrotrope in overabundance of water with the application of negligible vitality input. Hydrotrope is the key calculate in the bottom-up approach as it is included to break up water-insoluble lipids to shape lipid antecedents and anticipate the arrangement of fluid precious stones at tall concentration. Hydrotrope is an atom able to solubilize particle that able to solubilize ineffectively dissolvable operators in fluid media by hydrotropic solubilization which implies improvement of dissolvability of one solute by expansion of another solute. Urea, sodium alginate, ethanol, PEG, Propylene Glycol and sodium benzoate are among the most commonly utilized hydrotropes. The solubilizing instrument of hydrotrope includes complex arrangement between the hydrotrope and the hydrophobic agent. In this method, the cubic scatterings are made by crystallizing the forerunner solution.^[10, 11, 12]

Two partitioned arrangements are made.^[12]

- (1) A lipid arrangement in a fitting hydrotrope; and
- (2) A watery arrangement of stabilizing agents.

The dropwise expansion of the to begin with arrangement into the moment is carried out beneath steady blending at 1000 rpm.^[12]

For scaling up to mechanical fabricating, the bottom-up methodology is best due to all of the certain advantages.^[13]

- It is suitable for temperature-sensitive materials

since it may be carried out at moo temperatures.

- It has prepared the cubosomes and needs small energy.
- Long-term steadiness of the cubosomes is appeared since of the uniform scattering of stabilizer(s) on the cubosomes layer.
- It has used a hydrotrope to make the cubosomes streamlines the strategy and comes about in an item that is predominant to or comparable to those created utilizing a top- down method.

However, there are certain disadvantages^[14]

- Use of hydrotropes.
- The hazard of unfavourably susceptible reaction and inflammation.
- Furthermore, all through the planning stage, the arrangement of vesicular frameworks is unavoidable.

To delude the limitations related to the bottom-up technique, which employments a phosphate-buffered saline arrangement in a bilayered lipid framework comprising phytantriol and dodecyl dimethyl ammonium bromide (DDAB) as a cationic lipophilic stage, different novel ways have been formulated. The extra PBS reestablishes the bicontinuous bulk-phase cubic structure by creating the charged watch on DDAB, hence changing the bilayer system. A contrarily charged greasy corrosive, 1, 2- dipalmitoyl phosphatidylserine (DPPS), combined with phytantriol, produces a comparable impact.^[15]

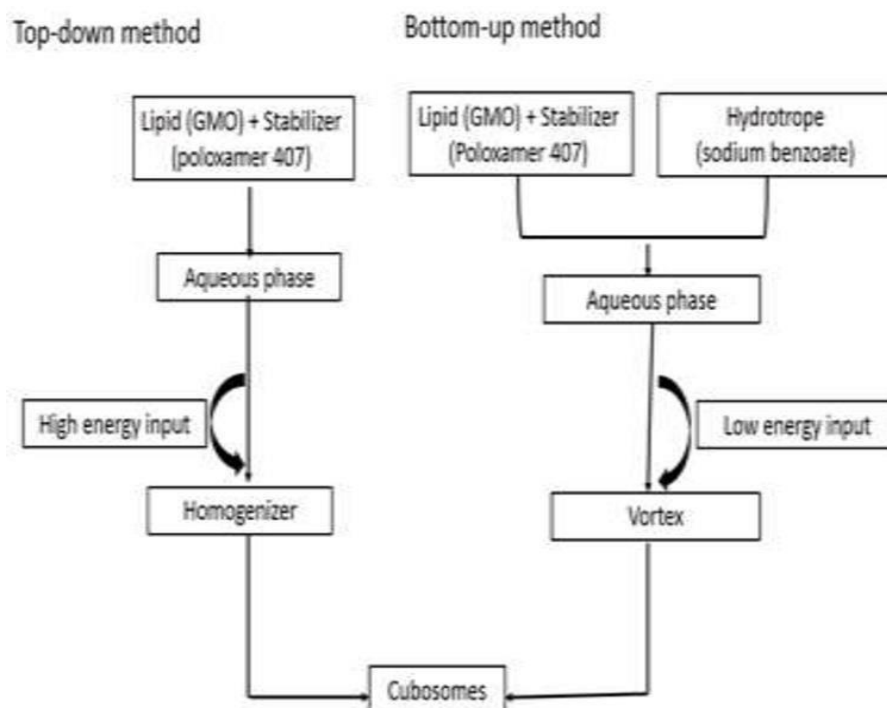


Figure 3: Diagrammatic illustration of cubosomes preparation approaches.^[16]

Materials used in Cubosomes Formulation

Cubosomes can be shaped from the bulk cubic structure when scattered in water into submicron particles with indistinguishable inner structures to the parent cubic structure. Cubosomes are organized in three measurements as "honeycomb" structure and they are primarily composed of amphiphilic lipids which scattered in water in the nearness of reasonable stabilizers. Bicontinuous cubic stages are found in normal lipids, cationic and non-ionic surfactants, and polymer frameworks, in spite of the fact that the lipid most broadly utilized to build bicontinuous cubic stages is the monoglyceride monoolein, monoglycerides suddenly shape bicontinuous cubic stages upon the expansion of water, are moderately insoluble (permitting the arrangement of colloidal scatterings of cubosomes), and are safe to changes in temperature.^[17]

Amphiphilic lipids

Glycerol monooleate (GMO): The primary forerunner of cubosomes arrangement is monoolein. Monoolein or glyceryl monooleate is a blend of the glycerides of oleic acid and other fatty acids, comprising basically of the monooleate. The acyl chain is connected to the glycerol spine by an ester bond. The two remaining carbons of the glycerol have dynamic hydroxyl bunches, giving polar characteristics to this parcel of the atom. The glycerol

moiety may frame hydrogen bonds with water in a watery environment and is commonly alluded to as the head bunch. The hydrocarbon chain gives hydrophobic characteristics to monoolein and is frequently named the tail. Commercially accessible monoolein may be gotten in two shapes, a blended glyceride frame or as refined monoolein; the refined monoolein is favoured for pharmaceutical applications since of its tall immaculateness. Monoolein happens as a waxy yellow paste with a characteristic odour. It swells in water, giving rise to a few lyotropic fluid crystalline structures. From a pharmaceutical angle the stage behaviour of the framework appears a number of curiously properties. GMO is a polar unsaturated monoglyceride, with a dissolving point: 35 – 37°C, capacity temperature –20°C, having HLB esteem 3. Figure speaks to the chemical structure of GMO, it appears that GMO with hydrophilic and hydrophobic characters at the same time (amphiphilic particle) this is property to the nearness of hydroxyl bunches in the head parcel which mindful for arrangement of H- bonds with water and the nearness of hydrocarbon chains in the tail. It is commonly utilized as an emulsifier in nourishment industry, characterized by being secure, nontoxic, biodegradable and biocompatible fabric and it was to begin with suggested as a biocompatible typifying fabric in 1984.^[17]

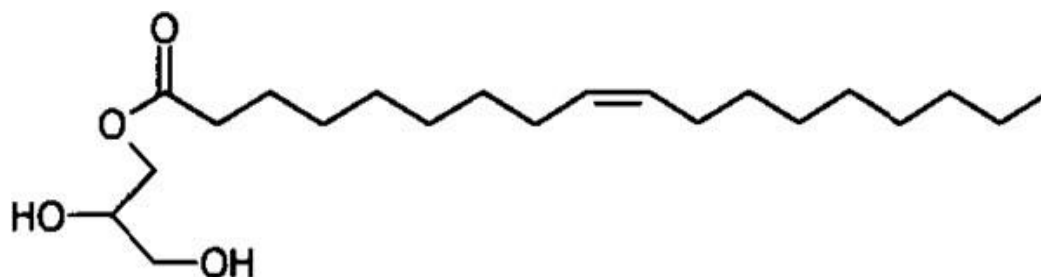


Figure 4: Structure of Glycerol Monooleate.^[17]

Phytantriol (PHYT): Phytantriol which is a common constituent in corrective items, is considered as a brilliant elective to GMO in cubosomes preparation. Phytantriol (3, 7, 11, 15-tetramethyl-1, 2, 3-hexadecantriol) has the capacity to frame a bicontinuous cubic structure in watery media beneath physiological condition and temperature. As of late it picked up more intrigued, compared to monoglycerides, in biomedical field due to its tall chemical soundness compared to monoglycerides as a result of nonattendance of ester gather, its improved

skin entrance properties, progressed dampness maintenance and it is commercially accessible with tall virtue (95 %), whereas monoglycerides are with distinctive purities as they are created from different sources. PHYT-based fluid crystalline frameworks were found to be able to support the discharge of distinctive medicate particles particularly those having hydrophilic properties and, in this way, it is considered as a momentous maintained sedate conveyance framework.^[18]

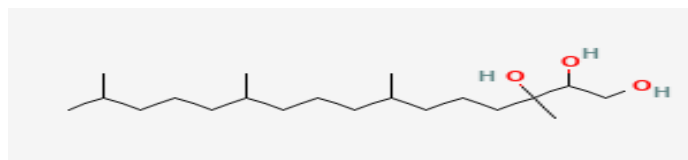


Figure 5: Structure of Phytantriol.^[18]

Stabilizers: In spite of the bulk cubic totals are thermodynamically steady, in any case, when they are scattered in fluid media, the scattered particles are not

dynamically steady as they tend to total as a result of presentation of hydrophobic parcels to the outside hydrophilic fluid media, in this way utilizing stabilizing

specialists ended up a pivotal step in cubosomes planning to avoid re-coalescence of the scattered particles into the parent bulk cubic structure when scattered in water. The fundamental work of the stabilizer is to give an electrostatic obstruction between particles to avoid near molecule contact and, in this way, keeping the scattered particles in a steady frame. This impact is created through the interest of the utilized stabilizer in the lipid water gathering without disturbing the cubic fluid crystallinity, in this way the choice of a fitting stabilizer is a basic step. The most commonly utilized stabilizing operators are Pluronics, particularly F127 (Poloxamer 407) which considered to be the "Gold standard". Pluronics are water soluble self-assembled triblock copolymers which composed of polyethylene oxide (PEO) and polypropylene (PPO) orchestrated in PEO-PPO-PEO arrangement where PPO and PEO parcels are dependable for hydrophobic and hydrophilic properties individually. In case of cubosomes, the stabilizing activity of F127 is thought to be a result of adsorption of the hydrophobic (PPO) parcel onto the surface of the particles, whereas the hydrophilic (PEO) parcel expands out into the fluid media giving steric protecting. Stabilizer, depending on the scattered particles, is as a rule utilized with a concentration up to 20 % w/w whereas GMO- polymer blend is as a rule utilized with a concentration between 2.5 % (w/w) and 10 % depending on the add up to weight of the scattering. As of late, Chong et al considered the capacity to utilize a verity of non-ionic atoms as stabilizers for cubosomes arrangement with GMO and PHYT lipids and compared their impact with F127. Interests, they uncovered that poly stearate (ethylene oxide) stabilizers were found to be more successful in stabilization of PHYT- cubosomes than the gold standard F127. Be that as it may the reason for this made strides solidness still hazy.^[19]

Loading and release of drug from cubosomes

The dispersion of therapeutics and peptides for overseeing an infection is essential for cubosomes stages. Lipid structures have long been utilized for dispersion, be that as it may, most regularly in the shape of filled vesicles. The natural fluid capacity and surface morphology region of vesicles and cubosomes shift altogether. The hydrophobic volume extent of a monoolein-based cubosomes with a width of 100 nm has been calculated to be more than three times that of a additionally measured solitary bilayer vesicle. Another advantage is that the bulk of the bilayer layer is interior the cubosomes or maybe than on the surface as in vesicles, protecting the payload from the environment. The structure and basic system of the bilayer film, i.e., their connections with the visitor atoms, can be altered, for occurrence, by including lipids with different acyl bunches to alter the charges characterizing the watery systems. The productivity of development can be controlled by changing the estimate of the have cubic phase's water channels, with included adaptability given by changing the concentrated of the electrostatic

fascination including the visitor components and the channel surfaces. The distance across of the watery channels can be balanced utilizing added substances; for occasion, including the octyl glucoside surfactant to GMO-derived LCPs was illustrated to increment the breadth of the hydrophilic channels.

Symmetrical lipid-based LLC frameworks were created that can alter in reaction to changes in pH, empowering controlled medicate conveyance.^[20]

Loading mechanism and characterization methods

Cubosomes examples can be stacked in three ways: interior the lipid layer, tied down to the lipid bilayer, or localized interior the cubic phase's fluid stages. Cubosomes can be consolidated either some time recently or after scattering by co-lyophilizing the dynamic compounds with the bilayer movies, by presenting the payload to liquid lipids, or upon scattering by stacking the cubosomes after brooding. The lion's share of considers on pressed cubosomes so distant have been for proteins or minor atoms coordinates interior the lipid bilayer, with single or double lipid combinations, basically of monoolein or phytantriol being the most common. Cubosomes frameworks have moreover been detailed to be loaded with natural atoms such as cancer drugs (EE 71–103%), headache medicine (EE 61.9–71.6%), and antibacterial peptides (with a variable EE from 7 to 60%), and to serve as potentiating operators for the organization of immunomodulators (EE 89.5 3.51%). Evaluation of the stacking and its auxiliary suggestions is a basic component of cubosomes. Small-angle X-ray scrambling (SAXS) can be utilized to decide that the insides lipid films of cubosomes have been stacked or maybe than the surfaces. When particles are stacked, particularly ionic atoms, they influence the characteristics of the bilayer by causing stage moves or changing the estimate of the cubic cross section constants, causing a move in the perceptible diffraction crests. To comprehend the soundness, official with cells, and the utilize of minor substances and peptides, it is fundamental to get it the conformational changes created by stacking them. One think about looked into the energy of consolidating neurotrophin BDNF into monoolein and eicosapentaenoic acid-based cubosomes. Inside the to begin with moment, it illustrated a typifying effectiveness of up to 82%, with stage moves and peptide stacking showing up in milliseconds. SAXS is a profoundly specialized method that is not perfect for deciding how much fabric is kept or ousted from the cubosomes. After the basic steadiness has been affirmed, emptied sedate substances can be expelled by means of weight ultrafiltration went with by reverse-phase HPLC analysis went with by UV/Vis spectroscopy, or dialysis went with by fluid chromatography–mass spectrometry to decide the stack capacity and energy of sedate discharge. Sephadex columns have essentially been created to isolate cubosomes from free fluorescent atoms, empowering the stacking execution to be calculated utilizing fluorescence. Isothermal titration

calorimetry is frequently utilized to illustrate the integration of palmitoyl polypeptide into the lipidic cubic stages when treated with cubosomes for anti-aging purposes. A Zetasizer as a rule makes a difference in deciding the molecule estimate, PDI, and zeta potential of the arranged cubosomes. To examine the plainly visible viewpoints of cubosomes, a polarizing light magnifying instrument and SEM are utilized broadly.^[21, 22, 23]

Approaches for encapsulating various drugs

At display, the epitome of drugs in cubosomes can be classified into three unmistakable stacking methods.

Pre-loading: In pre-loading, the sedate is joined into a liquid-crystalline gel at first, taken after by its scattering into Nanoparticles. The consolidated medicate can conceivably combine into the LC system, in this way advertising a higher embodiment proficiency and extra worth for pre-loaded cubosomes.^[24]

Post-loading: In post-loading, the medicate is adsorbed onto the pre-modelled cubosomes. This strategy makes a sterile cubosomes scattering as the post-incorporated particles involvement a pre-heat-processing cycle some time recently the sedate is joined into the cubosomes. The extreme item created by means of sonication too as a rule shows a diminished division of vesicles.^[24]

Hydrotrope-loading: In hydrotrope-loading, the sedate is stacked whereas cubosomes are suddenly created in a blend of ethanol/GMO. The previously mentioned strategy does not require the utilize of high-shear blending device, from there on decreasing the taken a toll of fabricating.^[24]

Interactions of biological molecules with cubosomes

The stage moves and intelligent of cubosomes with organic cell films are basic for understanding their potential drug-delivery applications. Jablonowska et al. examined the intelligent of phytantriol and GMO-based cubosomes with natural and lipid film models of unbiased monolayers of 1, 2-dipalmitoyl-sn-glycero-3-phosphocholine (DPPC) and anionic monolayers of 1, 2-dimyristoyl-sn-glycero-3-phospho-L-serine (DMPS) and their combination arranged utilizing the Langmuir procedure. It was watched that after joining of PHYT- and GMO-cubosomes into the monolayers, a move from strong monolayer to fluid condensed and from strong layer to more fluid layers was seen for DMPS and DPPC monolayers, separately, with a higher joining of PHYT cubosomes into DMPS than for GMO cubosomes. This was accomplished due to the solid hydrophobic intuitive between the PHYT chains and the lipid tail bunches. In spite of the fact that PHYT- and GMO-cubosomes have a comparable negative charge, their improved consolidation into the negligibly hydrated contrarily charged DMPS may depend on the degree of polar head intelligent. In solid DPPC, the introductory destabilizing activity was taken after by the higher

surface weight due to the consolidation of the cubosomes. Additionally, the impact of PHYT- and GMO cubosomes on HeLa cells uncovered that in PHYT-derived cubosomes, rhodamine color entered the cytoplasm and atomic locales of the cells, whereas the GMO cubosomes appeared as it were the color in the cytoplasmic locale. This underpins the thought that PHYT-based cubosomes can discharge their cargo into diverse cellular structures since of their hydrophilic surfaces, while GMO can shape as it were hydrophobic bonds, which limits the take-up of cubosomes by the cells. The correct carrier-cell interaction for PHYT cubosomes still needs clarification, but the above-mentioned considers proposed that PHYT-derived cubosomes cause a higher film unsettling influence than the GMO-derived cubosomes and can be utilized to discharge cancer therapeutics productively. For the most part, PHYT based cubosomes shown a higher haemolytic potential than GMO- based ones, indeed at moo dosages of 2 – 12.5 mg/ml. This can be decreased through surface alterations utilizing poly(ethylene glycol)methyl ether acrylate [P (PEGA)], a branched brush copolymer. The creators prescribed that the lipid exchange and layer disturbance can be controlled through adjusting the inner stabilizer polymer joining and the outside surface introduction. Phytantriol-based cubosomes moreover evoke 200 fold and 23 fold improved pro-inflammatory reactions by intergalactic (IFN)- α and inducible nitric oxide synthase (iNOS), individually. GMO based cubosomes, in turn, moved forward IFN- α and iNOS levels as it were 15 fold and 4 fold, separately. An amazing audit by Tan et al. summarized the intuitive of lyotropic fluid gems with different biomolecules. Briefly, cubosomes have not been broadly considered with respect to their steadiness in a cell culture medium. In Dulbecco's adjusted Eagle's medium, they kept up their cubic stage with minor cross section varieties for up to 24 hours, but on long-term capacity, they experienced creaming. The component of cubosomes take-up inside cell layers may influence their transfection adequacy. A ponder by Dyett et al. watched that a hydrophobic color typified in cubosomes (regularly 200 to 350 nm) was conveyed by means of combination of the whole cubosomes with the cell surface compared with liposomes (for the most part 80 to 150 nm), which were quickly exchanged to the lysosomes for debasement, which may clarify the less transfection effectiveness. Hence, cubosomes may combine with different cells by layer intuitive between the outside lipid cell film and the lipid bilayers of the cubic structures.^[25, 26]

Characterization of Cubosomes

Cubosomes comprise of nanosized lipid-based bicontinuous cubic stages scattered in overabundance water and regularly stabilized by an amphiphilic polymer. Their physicochemical properties are fundamental for sedate or quality conveyance, such as molecule measure and structure. There are a few procedures to characterize the cubosomes. These strategies have been assembled into two categories,

coordinate strategies which give stage distinguishing proof, and roundabout procedures where estimations lead to stages of characterization.^[27]

Direct Techniques^[27]

- Electron Microscopy
- Small Angle X-ray Scattering (SAXS)
- **Electron Microscopy:** Cryogenic transmission electron microscopy (Cryo-TEM) permits the characterization of complex liquid structures, giving a coordinate visualization of the estimate and morphology of the nano systems (in this case, cubosomes). It is too conceivable to analyze the grid symmetry and rehearsed distances. The characteristic “honeycomb” structure of cubosomes has been carefully imaged by Cryo-TEM in a tremendous number of thinks about, utilizing different lipid definitions for different applications which incorporate, for case, sedate conveyance system. The cubosomes were composed of dioleoyl phosphatidyl ethanolamine (DOPE) supplemented with little sums of PEGylated GMO. In this specific framework, DOPE/PEG(660)-GMO/water, DOPE was utilized, as it is more appropriate for parenteral pharmaceutical applications when compared to unsaturated monoglycerides (uMGs)-based cubic stage materials, which display harmfulness when infused in vivo at tall concentration. Cytryniak et al. detailed the utilize of cubosomes for the to begin with time as a dual-modality medicate conveyance framework for inner radiotherapy combined with chemotherapy. Cryo-TEM uncovered a well-ordered structure interior these monoolein-based cubosomes stacked with doxorubicin and radionuclide. Angelov et al. distinguished extra-large nanochannels in cationic particles stabilized by PEGylation (Monoolein/dioctadecyl dimethyl ammonium bromide (DOMA)/DOPE- PEG2000). Advance considers by Angelov et al. too empowered the perception of little cubosomes particles with well-defined water channels, forerunners of bigger cubosomes, which affirms the nanochannel-network development in diamond-type cubic lipid particles. Cryo-Field Outflow Filtering Electron Microscopy (Cryo-FESEM) was utilized to get 3D auxiliary data concerning cubosomes and hexosomes. In differentiate to Cryo-TEM, the Cryo-FE-SEM did not give any data with respect to the inside structure, but given surface and in general morphology bits of knowledge. This strategy permitted the portrayal that the bulk cubic stage and cubosomes arranged from phytantriol and pluronic F127 displayed a convoluted structure and bicontinuous nature with a non-intersecting organize of water channels. the control of Cryo-Electron Tomography (CET), which was utilized to reveal the surface structure between the cubic stage molecule and the encompassing water. The coming about translation is that the insides constituted a idealize bicontinuous cubic stage, whereas the exterior appears interlamellar connections, which speak to a move state between the fluid crystalline insides stage and the exterior vesicular

structure.^[27]

- **Small Angle X-ray Scattering (SAXS):** Diffusing procedures have been endlessly utilized in surface and colloidal science to explore estimate, shape, and molecule intuitive in bulk arrangement, and too at interfacing. Diffusing methods are non- invasive, which is an advantage, and a few executions permit their utilize in situ to characterize progressing forms. These strategies do not depend on the expansion of tests or other extra particles. The result speaks to a normal of the physical properties of the sample. SAXS is a capable procedure to recognize the crystallographic structure of fluid crystalline stages. In a SAXS explore, X-rays connected with the electrons in the particles and are scattered flexibly. SAXS distinguishes occasional spatial courses of action of diverse bunches in any given direction. It is worth highlighting that SAXS is commonly utilized to complement Cryo-TEM and other expository strategies for fluid crystalline structure confirmation. In this way, SAXS gives a worldwide see of the requested structure of the test, whereas CryoTEM advises a neighbourhood see on the cubosomes structure at the person molecule level. Concurring to Angelov et al., the basic SAXS information was utilized to affirm the arrangement of extra-large watery channel on for cubosomes shaped by Monoolein/DOMA/DOPE-PEG2000. Rizwan et al. complemented Cryo-FESEM micrographs on cubosomes arranged with phytantriol and pluronic F127 with SAXS, affirming a bicontinuous cubic fluid crystalline stage with Pn3m geometry. Structural SAXS information is too essential to characterize unused cubosomes details for different applications. Mathews et al. created a cubosomes for in- vivo medicate conveyance applications. The framework SAXS design uncovered a cross breed cubosomal lipid nanocarriers with a pH-sensitive shell made by biopolymer complexes. Johnsson et al. detailed that the Bragg crests distinguished in the X-ray diffractograms for a DOPE/PEG(660)-GMO/water framework might be related with an Ia3D cubic stage (45% w/w H₂O) and the Pn3M cubic stage (50% w/w H₂O). Most displayed a Im3m or Pn3m geometry, be that as it may the gyroid was too perceptible. SAXS tests have too been utilized to explore cubosomes stage behaviour in reaction to stacked atoms. A few cases incorporate indomethacin, curcumin, capsaicin, rapamycin, Fe₃O₄ magnetic particles, cisplatin, and paclitaxel. In expansion, the impact of outside parameters can too be analyzed utilizing SAXS. Yang et al. utilized SAXS to appear that the prepare of homogenization connected on a phytantriol-based cubosomes scattering did not lead to changes in the inner nanostructure. Although SAXS is respected as one of the most solid structure assurance strategies, it is not void of restrictions. Issues related with SAXS incorporate frail reflections when procured utilizing a lab source. This shortcoming is indeed more noteworthy when managing with scattered fluid crystalline frameworks due to their little measure and possibly non-uniform crystallographic

microstructure. Besides, a few frameworks may display two or a few co-existing mesophases, making the task of SAXS reflection crests to particular bunches a complex task.^[27]

Indirect Techniques^[27]

- Dynamic Light Scattering (DLS) and Zeta(ζ)-Potential
- Nuclear Magnetic Resonance (NMR)
- Rheology
- Polarized Light Microscopy
- Differential Scanning Calorimetry (DSC)
- Entrapment Efficiency
- Stability Studies

• **Dynamic Light Scattering (DLS) and Zeta(ζ)-Potential:** DLS is one of the most omnipresent strategies to ponder molecule estimate and flow in colloidal frameworks. It is a moderately straightforward non-invasive strategy to characterize particles in suspension. When a molecule arrangement is lighted with a monochromatic coherent light, the bar is scattered in all bearings, making a grainy dot design. Particles in arrangement are not inactive. They experience Brownian movement, and their nearby concentration vacillates in time. In a DLS try, the changes of the light escalated (I) of these dots, due to the dissemination of particles with time (t), are recorded and deciphered into an autocorrelation function, $g_2(t)$.^[27]

$$g_2(t) = I(t) I(t + \tau) / I(t)^2$$

Where, I (t) is the intensity of the scattered light at a time (t) and I(t + τ) the light scattered at time (t + τ), respectively.^[27]

This procedure is regularly utilized to degree the estimate of the arranged cubosomes. The major disadvantage of DLS estimations is the reality that heavier and bigger particles contribute more unequivocally to the generally cruel rot rate of a polydisperse arrangement, which frequently lead to an overestimation of such bigger particles. In spite of this, this profitable data relative to translational dissemination of the particles, and in this manner molecule measure, runs hand in hand with characterizing cubosomes. The sizes of “honeycombed” organized cubosomes have been measured for an expansive assortment of diverse frameworks predetermined for different applications, extending from 10 – 500 nm in diameter. Malheiros et al. utilized the DLS strategy to analyze the normal hydrodynamic distance across and polydispersity of phytantriol cubosomes arranged in full hydrated conditions in the nearness of expanding hexa decyl phosphocholine (HePC) concentrations. They illustrated that there was an increment of 2-fold in the polydispersity record at both 10 and 14% mol, uncovering the impact of the HePC in the colloidal framework at these higher concentrations. In turn, Victorelli et al. created mucoadhesive cubosomes for the

conveyance of curcumin for cervical cancer treatment and utilized DLS to characterize the framework. The curcumin-loaded cubosomes displayed a homogeneous estimate conveyance with a polydispersity file lower than 0.4. Another imperative parameter and regularly gotten to for fluid crystalline frameworks is the assurance of ζ -potential. ζ -potential estimations can as it were be performed by implication, where tests are outlined to degree the speed of a charged molecule that moves beneath the impact of a connected electric field (electrophoretic portability). For pharmaceutical applications, ζ -potential is vital to decide the presence of cationic or anionic particles in arrangement. For occurrence, Victorelli et al. was able to affirm the nearness of positive charges on curcumin-loaded cubosomes through ζ -potential estimations. Patil et al. created an inhalable bedaquiline- loaded cubosomes nanocarriers for non-small cell lung cancer treatment. The ζ -potential estimations appeared that the nanocarriers were cationic and steady with respect to molecule measure for periods of eight weeks at storage conditions.^[27]

• **Nuclear Magnetic Resonance (NMR):** NMR is a multifaceted procedure which can be utilized to decide physical and chemical properties of molecules and atoms, giving nitty gritty data in terms of structure, flow, response state, and chemical environment of the atoms. It is worth highlighting the spearheading work of Pieter Cullis when examining the utilize of NMR to characterize organic and manufactured lipidic frameworks. Rajesh et al. investigated lipidic poly(2-methyl-2-oxazoline) (PMeOx) as an elective to F-127 to stabilize cubosomes. Two lipopolymers, PMeOx40-OA and PMeOx80-OA, with distinctive degrees of polymerization, were effectively synthesized and analyzed for end- group effectiveness by ¹H NMR. The ¹H-NMR design appeared that polymers had a exceptionally tall degree of end-group functionalization ($\geq 95\%$), hence reasonable for the arrangement of PMeOx stabilized cubosomes.^[27]

• **Rheology:** Rheology bargains with how a framework reacts to mechanical irritation in terms of flexible distortions and of gooeey stream. Shear rheology can give both auxiliary and dynamical bits of knowledge of fluid crystalline mesophases. Viscoelastic materials are more often than not autonomous of strain up to a basic strain level. The viscoelastic behaviour of tests is examined by measuring the strain sufficiency reliance of the capacity and misfortune moduli (G' and G'' , respectively). The common behaviour for cubic stages of G' and G'' as a work of recurrence is G'' prevails at lower frequencies, whereas G' is prevailing at higher frequencies. $G'' > G'$ shows that the fabric is profoundly organized and carries on solid-like. Expanding the strain over the basic strain disturbs the organize structure and the fabric gets to be continuously more fluid-like ($G' < G''$). The recurrence at which the hybrid, $G' > G''$, happens depends on the composition of the test. Among

the fluid crystalline stages, bicontinuous cubic stages (Ia3d, Pn3m, and Im3m) are respected as the most unbending structures taken after by the turn around hexagonal stage, which have halfway viscoelastic properties, and the lamellar stage, which is characterized by plastic liquid properties. Mezzenga et al. detailed that each particular fluid crystalline stage has a particular rheological signature. The estimations of G' and G'' as a work of temperature and composition can moreover uncover basic moves between fluid crystalline structures. Based on these contrasts in viscoelastic behaviour, auxiliary moves between the mesophases have been recognized in a few exploratory frameworks, counting cubic-to-hexagonal, cubic-to-cubic, and hexagonal-to-isotropic liquid. It was moreover appeared that cubic stages have diverse characteristic unwinding times. The unwinding time of the frameworks is gotten from the converse of the recurrence at which the hybrid of G' and G'' takes put. It can be seen as the time scale for the water-lipid interface to unwind to or get balance arrangement, after having been annoyed by shear distortions. Villalva et al. immobilized cubosomes in oxi-HA/ADH hydrogels and performed rheological thinks about illustrating that the nearness of both anionic and cationic cubosomes somewhat expanded the G' values for hydrogels of moo and tall degrees of cross-linking with regard to their individual immaculate hydrogels. Hence, the cubosomes charge does not influence the mechanical properties of the hydrogel. These rheological properties affirmed that the in situ applicability of the hydrogel is not affected. It is worth specifying here that, in later a long time, Fluorescence Recuperation After Photobleaching (FRAP) has gotten to be a common device to assess lipid self-diffusion. FRAP is a fluorescence-based procedure which assesses dissemination of a fluorescent fabric into a locale, where a tall escalated laser has already delivered photobleaching.^[27]

- **Polarized Light Microscopy:** The expansion of cross polarizers can empower refinements between stages that, to the exposed eye, may appear indistinguishable. This implies that polarized light microscopy can uncover the morphology of the fluid crystalline based on the optical birefringence marvels, recognizing between anisotropic and isotropic courses of action. For case, hexagonal fluid crystalline due to the anisotropic atomic course of action will show up to be shinning when watched through crossed polarized light due to birefringence. For cubic fluid crystalline, the isotropic atomic course of action is non-birefringence and hence will show a dim field in the polarizing picture. Among all the strategies, polarized light microscopy gives the least demanding way to subjectively recognize between phases. Tian et al. created folate- modified cubosomes containing etoposide in arrange to accomplish way better focusing on properties and helpful impacts compared with the conventional cubosomes. The arranged cubosomes were characterized by polarized light microscopy, which affirmed their cubic morphology

. Also, Fan et al. created a fluid taste-masking framework based on lyotropic fluid crystalline nanoparticles (LLCNs) for paediatric medication. The creators too typified cefpodoxime proxetil (CFP) into the LLCNs (CFP- LLCNs) to make strides their taste and in this way encourage the organization of drugs to children. The mesophase investigation of the arranged nanoparticles was performed by polarized light microscopy and SAXS, uncovering the cubic stage of CFP- LLCNs and clear LLCNs, in this manner affirming that CFP stacking did not impact the stage of LLCNs.^[27]

- **Differential Scanning Calorimetry (DSC):** DSC is greatly important to ponder the thermotropic behaviour of different frameworks due to the extractable data in terms of move temperatures and enthalpies of moves. Fluid crystalline is a thermodynamic harmony framework. A stage move is as a rule went with by endothermic or exothermic vitality changes, which can be decided by DSC. Mansour et al. arranged dexamethasone- loaded cubosomes that were subjected to DSC considers in arrange to distinguish the impact of excipients and prepare parameters on the physical soundness of the shaped particles. These ponders appeared that the commonplace endothermic top of dexamethasone vanished in the sedated lyophilized cubosomes, proposing its entanglement into the shaped cubosomes in a shapeless frame. Moreover, Zhang et al. stacked anticancer drugs, to be specific, cisplatin and paclitaxel in cubosomes coated with a layer of poly- ϵ -lysine. The arranged cubosomes were subjected to DSC considers, which shown that the drugs were not show in the cubosomes in crystalline frame but scattered all through the cubosomes.^[27]

- **Entrapment Efficiency:** Cubosomes have perpetual potential as sedate conveyance frameworks as these frameworks can hold satisfactory sums of little drugs, peptides, biologic, or bioactive particles. There are various approaches for stacking the sedate into cubosomes, with a common denominator of victory detailed as the entanglement effectiveness. Entanglement effectiveness and medicate stacking of cubosomes can be gotten to turning to chromatography procedures, analysis, small-angle X-ray diffusing, or ultra-filtration methods. The sum of untrapped sedate can be encourage analyzed utilizing a UV spectrophotometer, fluorescence, HPLC examination, Fluorescence Relationship Spectroscopy, or radioactivity for the reason of, for case, getting a sedate discharge profile. Alharbi et al. created ciprofloxacin-cubosomal (Cf-Cub) in situ gel with the point of moving forward eye saturation, dragging out the visual maintenance time, and improving its antimicrobial movement. In this consider the capture productivity was decided, appearing its reliance on calculate levels in definitions. Besides, Elakkad et al. created transcutaneous tenoxicam stacked hyal-cubosomes prepared by including sodium hyaluronate to cubosomes components to control osteoarthritis without common side impacts. The capture

productivity rate of the created vesicles was decided by roundabout procedure. This consider uncovered that the capture proficiency of the tenoxicam-loaded hyal-cubosomes is marginally higher, at around 3%, when compared to tenoxicam-loaded cubosomes. These comes about may be due to the inflexibly organized vesicle shaped by hyaluronate, which favoured the consideration of tenoxicam interior the vesicles.^[27]

• **Stability Studies:** The physical steadiness of cubosomes is examined by the examination of organoleptic and morphological angles as a work of time. Basically, all strategies portrayed over can be utilized in a way to assess these perspectives. The confirmation of the well-defined internal structure characteristic of cubosomes can be performed at distinctive time-points. Particles estimate conveyance and medicate substance can be surveyed over time and be utilized to assess conceivable variations. As already specified, Elakkad et al. arranged tenoxicam-loaded hyal-cubosomes and tenoxicam-loaded cubosomes. These tests were moreover subjected to a steadiness ponder, which appeared the molecule estimate, zeta-potential, and entanglement productivity of cubosomes detailing and hyal-cubosomes detailing some time recently and after 3 months at $4^{\circ}\text{C} \pm 1$. This consider permitted us to conclude that capacity of the arranged vesicles in firmly closed golden glass holders at temperature 4°C did not antagonistically influence either the molecule measure and PDI, whereas zeta potential and entanglement effectiveness rate were influenced. In expansion, Bessone et al. created Latanoprost-loaded phytantriol cubosomes for the treatment of glaucoma. The arranged tests were put away in firmly closed golden glass vials in a controlled stickiness thermostatic cabin (25°C) for a period of 30 days. In this way, the soundness ponders illustrated that the concentration of latanoprost in the cubosomes and the physicochemical properties did not alter over one month, evidencing great stability.^[27]

Medical Applications of Cubosomes

The characteristics of cubosomes make them promising arrangements to a few biomedical applications. The capacity to consolidate hydrophobic, hydrophilic, and amphiphilic compounds, the capacity to ensure and secure convey atoms, and the convoluted inside arrangement that permits a maintained discharge are a few cases of highlights that make cubosomes particles with such awesome potential. From therapeutics to imaging, or indeed a combination of both, endless applications have been proposed with respect to the utilize of cubosomes in the biomedical field, such as in cancer, contamination, or indeed cerebrovascular disorders.^[28]

Cancer Therapy: Cancer is a major worldwide wellbeing challenge, with millions of unused cases each year, and speaks to the moment driving cause of passing agreeing to World Wellbeing Organization. The logical advancement permits the disclosure of unused approaches towards the treatment of distinctive sorts of cancer. In the advancement of unused methodologies, and particularly modern drugs, a few of the challenges are the organization, the transport, and steadiness of the compound interior the life form and the capacity to stir the needed impact. Nanocarriers play an imperative part in overcoming these deterrents, as their fundamental reason is to provide the atoms of intrigued securely and effectively to the craved area. A great sedate conveyance framework permits the lessening of the sedate dose without compromising the therapeutics, and along these lines reduces the side impacts. A few ponders have been distributed with respect to the application of cubosomes as a nanocarrier for cancer therapeutics. Doxorubicin, cisplatin, paclitaxel, curcumin, and quercetin are a few cases of atoms that have been effectively typified in cubosomes, either alone or in a few kind of synchronous encapsulation.^[28]

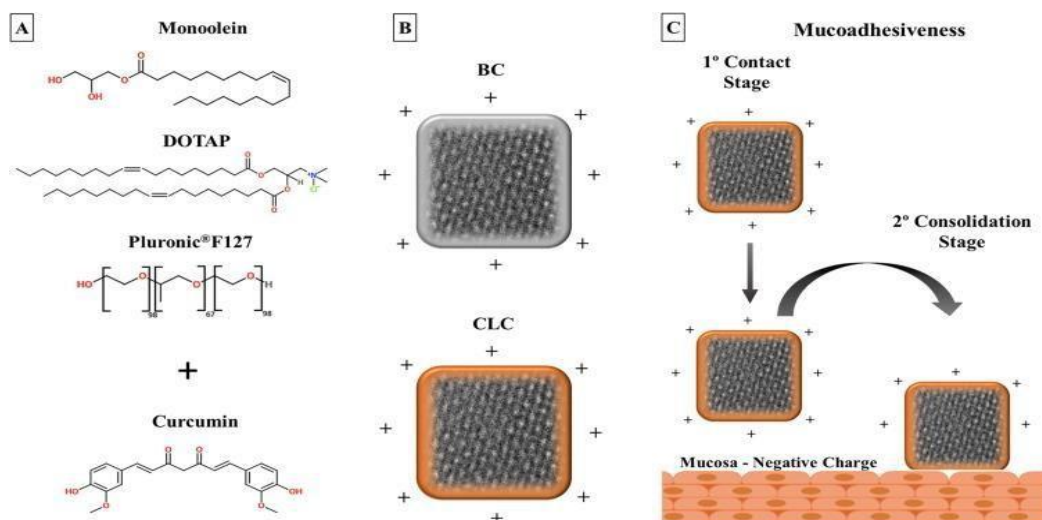


Figure 6: (A) Components of the cubosomes, (B) blank cubosomes (BC) and curcumin- loaded cubosomes (CLC), and (C) the interaction of the cubosomes with the negatively- charged mucosa.^[28]

Zhai et al. examined the adequacy of monoolein cubosomes stacked with paclitaxel in the treatment of ovarian cancer. The in-vitro comes about in Hello cells (HX-62) appeared that the clear cubosomes did not display cytotoxicity, and that stacked cubosomes were viable in the tumor restraint, having a more effective activity than free paclitaxel. In vivo tests demonstrated the past comes about and permitted us to conclude that the stacked cubosomes expanded the survival of the tumor bearing mice. In another work, L. Zhang et al. endeavor to at the same time typify two distinctive compounds into the cubosomes. Monoolein cubosomes were stacked with either cisplatin, paclitaxel, or a combination of both, and coated with poly- ϵ -lysine. Cytotoxicity and activity against cancer cells were assessed by MTT measure on human hepatoma Hep G2 cells and fluorescence on HeLa cells, separately. The gotten comes about appeared that the coated cubosomes had a higher embodiment proficiency, altogether decreased the starting burst discharge, and permitted for a controlled and moderate discharge of the atoms. Besides, the coating permitted for an expanded reasonability of the Hep G2 cells and a passing increment on HeLa cells. At last, the cubosomes with a combination of paclitaxel and cisplatin appeared a higher effectiveness against HeLa cells when compared to single-loaded cubosomes. Such comes about prove that cubosomes have gigantic potential as medicate carriers and may be an arrangement to diminish the sedate dose without compromising the therapy. Bazylinska et al. created a photodynamic approach towards the treatment of skin threatening melanoma. Monoolein cubosomes were stacked with Chlorin e6 (Ce6) or meso-Tetraphenyl porphine-Mn (III) chloride (TPP-Mn), two photosensitizing colours. The cubosomal arrangement appeared particles with a normal measure of 130 nm and more polydispersity (0.13). In vivo tests with respect to cellular take-up, cytocompatibility, and photodynamic action were conducted in two human harmful melanoma cell lines (Me45 and MeWo). In both cases, the take-up of the typified photosensitizers was more noteworthy when compared to the free shape of these compounds.

Besides, Ce6-loaded cubosomes shown moo cytotoxicity earlier to illumination, and tall cytotoxicity after the enthusiastic input, in this manner being biocompatible therapeutic-effective particles. In the case of TPP-Mn, a moo photocytotoxicity was gotten. The creators conclude that in spite of this drawback, TPP-Mn's tall cellular take-up might be utilized for cancer cells imaging. The one of a kind characteristics of cubosomes makes them curiously to utilize in modern cancer treatment approaches. An inventive work examined the utilize of cubosomes as synchronous carriers of doxorubicin, a chemotherapeutic, and Lutetium-177, a radionuclide, in this way permitting for a combined chemo- and radiotherapy. In the explore, the combination come about in steady cubosomes, but with immaterial cancer therapeutics when compared to other arrangements. The creators conclude that the doxorubicin displayed a much higher cytotoxicity than Lutetium, subsequently encourage ponders are required, and with a distinctive sort of radionuclides that ought to be able to store more vitality in shorter periods. Additionally, it is worth taking note the improvement of inhalable bedaquiline-loaded cubosomes (BQLC) for non-small cell lung cancer treatment (NSCLC). These nanoparticles were arranged by means of a dissolvable vanishing procedure, appearing great characteristics such as molecule estimate (150 nm), zeta potential (35 mV), and aerosolization behaviour. This consider prove the proficiency of the nanoencapsulation of bedaquiline, an anti-tuberculosis sedate with extraordinary anti-cancer adequacy, permitting to overcome its destitute water solvency that limits its conveyance through the lungs. Without a doubt, the effective internalization of the BQLC in lung cancer cells (A549 cells) has been uncovered, which come about in the diminish of the half-maximal inhibitory concentration (IC₅₀) (3-fold), as well as in the critical restraint of tumor development compared to free bedaquiline. The great aerosolization execution and made strides anti-cancer movement illustrated by the gotten comes about highlight the potential of BQLC as NSCLC treatment.^[28, 29]

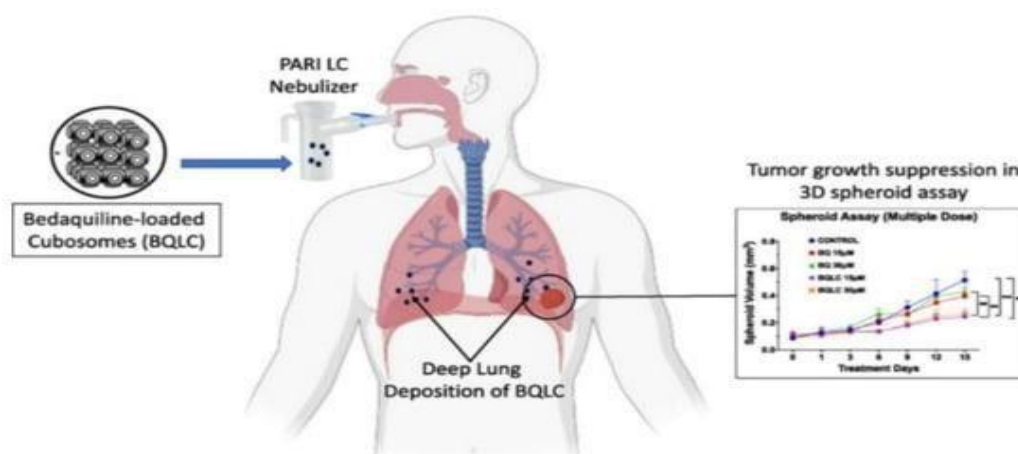


Figure 7: Conceptual representation of the work by Patil et al., 2021. Bedaquiline- loaded cubosomes were designed for inhalatory administration in the context of cancer therapy.^[29]

Transfection: Gene treatment comprises of the utilize of nucleic acids to accomplish a hereditary control of particular cells. It can be utilized, for case, in the treatment of monogenic infections by substituting the lost or transformed quality with a typical allele. In expansion, quality treatment can be utilized in the treatment of cancer, Helps, Parkinson, among others. In spite of its potential, quality treatment utilize has been restricted due to the need of effective carriers. This trouble is related with finding carriers that satisfy the prerequisites required, for illustration: dodge the debasement of the nucleic acids, encourage cellular take-up, and advance the discharge of the hereditary particle in the cell. In the case of viral vectors, these carriers have appeared immunogenicity and are troublesome to handle and to deliver on a large-scale. In terms of physical quality transfection frameworks, a few of its drawbacks are the troublesome large-scale generation and the moo transfection productivity. Chemical carriers have been made with the point of overcoming the downsides of viral and physical frameworks. Lipids have the capacity to connected with plasmid DNA and thus pass the cell layer, subsequently being one of the materials utilized in the advancement of such frameworks. Cubosomes are lipidic nanoparticles, thus they can be a great arrangement for the conveyance of the hereditary molecules. To investigate the contrasts between the utilize of cubosomes and liposome-based frameworks as carriers of siRNA, Kim et al. created

“PEGylated cuboplexes”: monoolein and DOTAP cubosomes coated with PEG and stacked with siRNA. In their work, HeLa cells were pre-treated to express firefly luciferase. A while later, cubosomes focusing on the FF mRNA for sequence-specific corruption were included to the cells. As a result, the cuboplexes effectively conveyed siRNA to the cells and quieted luciferase. Besides, the created particles appeared a superior productivity when compared to the classical liposomal options. In another work, Kim et al. created cuboplexes utilizing microfluidics. This strategy permitted to accomplish a way better control over the particles estimate and polydispersity without compromising the capacity of the particles to carry siRNA, thus advancing the craved gene-silencing. Another case of a fruitful transfection utilizing cubosomes was detailed by Sarkar et al. Diverse combinations of monoolein doped with cationic lipids were utilized in the get together of cubosomes stacked with antisense green fluorescent protein (GFP). GFP communicating Chinese Hamster Ovary cells were treated with the cubosomes. The comes about permitted us to conclude that the knockdown effectiveness is related to the cationic lipid utilized and that, for a period of up to 48 h, a few of the combinations display a quality knockdown effectiveness comparative to lipofectamine (liposomal-based detailing that is commercially accessible), and a higher effectiveness after 72 hours.^[30]

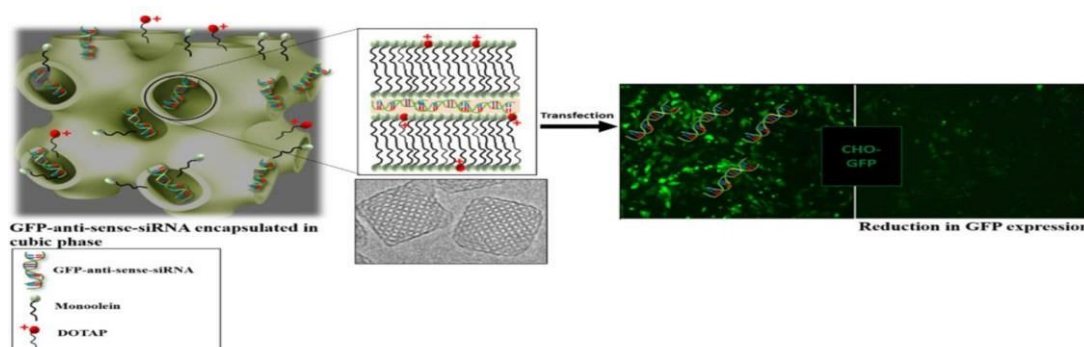


Figure 8: Conceptual representation of the work by Sarkar et al., 2021. Cubosomes loaded with GFP-antisense siRNA are shown to reduce the expression of GFP in vitro using Chinese Hamster Ovary cells.^[30]

Gajda et al. made a curiously restorative combination where monoolein cubosomes were stacked with miR-7-5p (miRNA) and doxorubicin (DOX-chemotherapeutic) for a glioblastoma treatment. Drug-sensitive (A172 and TPC-1) and drug-resistant (HeLa and T98G) cell lines were utilized to test the adequacy of the combined treatment with the cubosomes stacked with miRNA and DOX. The comes about appeared that stacked cubosomes with DOX/miR-7-5p were more viable in actuating cellular apoptosis than cubosomes stacked as it were with DOX. The creators conclude that miR-7-5p permits the decrease of cells' drug-resistance, in this manner upgrading the productivity of the drug.^[30]

Topical Drug Delivery and Antimicrobial Therapy: Transdermal sedate conveyance permits for a pain-free

medicate conveyance that can maintain a strategic distance from to begin with pass digestion system, hence permitting a lower measurement. Be that as it may, there are a few challenges in the utilize of this organization course. One case is the stratum corneum, one of a few layers that compose the skin, which acts like an obstruction, making it troublesome for the atoms to enter. Lipid-based colloidal frameworks can offer assistance to fluidize skin lipids, permitting to upgrade the entrance of atoms. Cubosomes can, in this manner, be a conceivable vehicle in the conveyance and transport of drugs to and through the skin. In addition, cubosomes moreover have extraordinary potential to be utilized in antimicrobial treatment, improving the activity of drugs against pathogens. Boge et al. investigated the capacity of cubosomes to topically provide LL-37, an

antimicrobial peptide (AMP). LL-37 was stacked into monoolein cubosomes by three distinctive approaches: pre-loading, post-loading, and hydrotrope-loading, which compare to the embodiment of the compound earlier, after, and amid the cubosomes arrangement, separately. Comes about appeared that the epitome approach had an effect on the cubosomes sizes and structures, with hydrotrope-loading appearing the littlest cubosomes. Not one or the other of the details prove skin-irritation. Beneath physiological conditions, a few AMPs are vulnerable to proteolytic corruption. Once uncovered to *Pseudomonas aeruginosa* elastase or human neutrophil elastase, immaculate LL-37 was totally debased, while the LL-37 stacked into the cubosomes remained intaglio. Besides, the typified LL-37 shown bactericidal impacts against *E.coli* and *S.aureus*, indeed after the protein introduction. Bioadhesion of cubosomes makes them a fitting choice as carriers for the conveyance of particles to outside tissues. Monoolein cubosomes were stacked with norfloxacin, an anti-microbial utilized in the treatment of intense otitis externa, ordinarily caused by microscopic organisms, basically *pseudomonas*. In vivo tests conducted in rabbits prove that the utilize of cubosomes as carriers permitted to move forward the statement of norfloxacin in rabbit ears, in comparison with a non-encapsulated norfloxacin suspension. This affirmed the *ex vivo* ponder comes about, which too appeared a superior entrance of the typified norfloxacin. Histopathological examinations did not uncover any signs of skin bothering. This illustrates the potential utilize of cubosomes as carriers for the upgrade of transdermal conveyance of norfloxacin. These comes about are exceedingly curiously and can upgrade the topical application of other sorts of particles (common and manufactured), modernizing the dermo-cosmetic industry or indeed medicine. Glaucoma is a major worldwide wellbeing issue that can lead to irreversible visual impairment. In a glaucoma crisis, acetazolamide is the sedate of choice. Be that as it may, the as it were conceivable organization is through systemic tablets. Teba *et al.* proposed the definition of a cubosomes based framework for topical conveyance of acetazolamide. In their work, the comes about permitted the conclusion that the optimized definition of acetazolamide-loaded monoolein cubosomes were not an aggravation for the eye and were able to stir a longer enduring and viable therapeutics than other pharmacological arrangements commercially accessible. In expansion, Bessone *et al.* arranged latanoprost-loaded phytantriol cubosomes (CubLnp) utilizing a top-down strategy. These cubosomes appeared a exceptionally moderate discharge of Latanoprost, an anti- glaucoma medicate, evidencing a supported discharge profile. It has too been appeared that a single subconjunctival application of CubLnp actuated more prominent lessening in intraocular weight of around 30% at 24 hours. Besides, moo concentrations of CubLnp uncovered a hypotensive impact, with an intraocular weight diminish of 20%. Cubosomes can moreover be joined in hydrogels in arrange to get a superior

maintained sedate conveyance framework. In this way, Sanjana *et al.* created and characterized a novel cubosomal detailing, counting dexamethasone (DMS), for vitiligo treatment. The cubosomes were arranged through top-down strategy and were joined into hydrogel for delayed conveyance of DMS. The cubogel was shaped by joining an optimized detailing in 1% carbopol 940. The arranged cubogel shows great properties, such as upgraded spreadability, higher medicate substance, perfect pH, and an advantageous maintained discharge design at the conclusion of 12 hours, that makes it appropriate to be a promising topical supported medicate conveyance framework to treat vitiligo. Additionally, Rapalli *et al.* created topical hydrogel comprising of ketoconazole-loaded cubosomes with lower surfactant concentrations. This hydrogel appeared a higher saturation and skin maintenance, as well as an upgraded anti-fungal movement, being a curiously procedure for topical medicate conveyance. Villalva *et al.* effectively immobilized diclofenac-loaded cubosomes in oxi-hyaluronic acid/ADH hydrogels, uncovering its potential as an imperative stage for biomedical applications in sedate conveyance. Recently, Lai *et al.* appeared that polymyxin- loaded cubosomes may disturb the external layer of Gram-negative microbes, coming to a prevalent polytherapy action. Without a doubt, the external film was at first destabilized by the electrostatic intelligent between polymyxin and lipid A, and in this way, the layer was encouraged disturbed through a lipid trade handle caused by a convergence of cubosomes.^[30]

Vaccines: Vaccination is a fundamental way to secure the body from bacterial or viral contaminations and cancer. A compelling antibody ought to propel solid humoral and cellular safe reactions. The victory of antibodies includes antigen and coordinating adjuvants. So distant, an assortment of nano systems have been created into adjuvants, such as liposome, PLGA, and cubosomes. In particular cases of cubosomes, later considers have uncovered their adjuvant movement and potential utility as an antigen conveyance framework. The later examinations found that cubosomes have the capacity to advance more humoral and cellular resistant reactions than antigen alone. Ginseng stem-leaf saponins (GSLs) display amazing immunoregulatory applications, but a moo bioavailability. Qiu *et al.* considered chitosan altered monoolein cubosomes, arranged by dissolvable moving, and stacked with GSLs and ovalbumin (OVA-antigen). The gotten cubosomes did not show cytotoxicity and an increment in the take-up effectiveness of OVA was enrolled. Moreover, the comes about displayed a tall generation of OVA-specific IgG antibodies, and a way better cellular safe reaction compared to the other examined bunches (e.g., cubosomes stacked with OVA, but not with the GSLs). In vivo comes about appeared that the particles might move forward lymphocyte enactment and resistant reaction in splenocytes. Thereby, the cubosomes illustrate extraordinary potential to be utilized as carriers for transport and focused on conveyance of antigens and

antibodies adjuvants.^[31]

Ocular applications: Numerous later thinks about have concerned with the application of cubosomes in visual medicate conveyance. Utilizing their benefits of being biodegradable, able to typify all 3 sorts of medicate particles as hydrophilic, hydrophobic and amphiphilic, and they render bioactive operators with focused on discharge and controlled discharge. They are found to move forward visual bioavailability of the stacked drugs since they have long home time at the corneal surface and characterized by mucoadhesive properties due to the nearness of GMO driving to make strides corneal penetrability and thus progress visual bioavailability of the joined drugs. Curiously comes about were gotten when cubosomes were examined as a topical visual medicate conveyance frameworks. In-vitro penetration think about of cubosomes stacked with dexamethasone through extracted rabbit corneas, comes about appeared that cubosomes detailing was found to increment the clear porousness coefficient. Furthermore, the precorneal home time test and pharmacokinetic consider of fluid humour tests comes about uncovered that cubosomes details cause a noteworthy increment in preocular maintenance time compared to Dex-Na phosphate eye drop and ordinary ophthalmic arrangement, the comes about uncovered no noteworthy contrast in in- vitro corneal penetration characteristics but essentially quicker onset and higher concentrated of mydriatic activity brought about through in-vivo consider for the cubosomes detailing. Few cases of cubosomes stacked drugs for visual application, all comes about appeared incredible benefits of cubosomes for visual sedate conveyance in dragging out the precorneal home time, progressing visual bioavailability of stacked sedate too histopathology considers demonstrated that cubosomes planning are secure and non-irritant for visual employments.^[31]

CONCLUSION

The advancement of nanocarriers driven to vital progresses in a few investigate regions, especially in the therapeutic field. Investigate on nanocarriers has demonstrated to increment the effectiveness of restorative medications, for the most part due to their capacity to target particular cells. The fundamental intrigued behind the biomedical utilize of cubosomes depends on their capacity to consolidate particles with distinctive physicochemical properties, such as polar and apolar molecules. Cubosomes arranged in scatterings have a nano meter scale structure indistinguishable to bulk cubic stage, but the scattering itself has much lower, water like consistency. Compared to liposomes or vesicles, cubosomes have much higher bilayer area-to-particle volume proportions as well as higher gooeey resistance to rupture. Cubosomes are among an extraordinary lesson of lipid-based nanovesicles which characterized by fluid crystalline nature of their nanostructure, arranged from amphiphilic lipid which self-assembled in water and in nearness of stabilizer into

cubosomes. As of late various distributed reports demonstrated their potential employments as a novel sedate conveyance framework. Cubosomes have been endorsed as an successful visual medicate conveyance with upgraded visual home time, bioavailability and no bothering to the eye. Verbal application outlined that cubosomes can be utilized viably to increment assimilation of ineffectively water dissolvable drugs, secure the at risk medicate from enzymatic corruption and in focused on sedate conveyance. They give a promising vehicle for viable transdermal sedate conveyance with upgraded skin penetration and moo bothering potential. Interests, cubosomes were connected for conveyance of anticancer drugs with diminished genuine side impacts of the chemotherapeutic specialists and focused on medicate delivery.

Cubosomes offer promising choices to lipid-based nanostructured lipid carriers for solvency improvement and the wanted conveyance of different restorative particles, counting hydrophobic, hydrophilic, and zwitterionic substances and biomolecules. Surface functionalization with different polymers and ligands assist optimizes their properties for particular drug delivery applications. Solvency improvement of ineffectively water soluble compounds and the maintained or amplified conveyance of stacked dynamic fixings are two noteworthy points of interest of their utilize as conveyance carriers. As the investigate proceeds for modern shrewd lipid nanocarriers, cubosomes will have broader utilize, extending their applications to makeup, immunoassays, and bio macromolecular conveyance, and their harmfulness.

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