EUROPEAN JOURNAL OF BIOMEDICAL AND PHARMACEUTICAL SCIENCES
http://www.ejbps.com

ISSN 2349-8870
Volume: 8
Issue: 6
529-536
Year: 2021

PRONIOSOMES AS POTENTIAL DRUG CARRIER

Dr. P. Nagaveni*, G. Charan Kumar Reddy and Dr. CH. Appa Rao

?

***Corresponding Author: Dr. P. Nagaveni**

.

Article Received on 21/04/2021 **Article Revised on 11/05/2021** **Article Accepted on 31/05/2021**

1. ABSTRACT

Nanotechnology has brought a revolution in the field of science, which has subsequently lead to development of novel dosage forms such as niosomes, liposomes and proniosomes. Proniosomes overcome the disadvantages involved with niosomal and liposomal drug delivery systems. Proniosomes are liquid crystalline compact niosome hybrids which upon hydration form niosomes. They help in reducing physical stability problems involved with niosomes such as leaking, fusion, aggregation and provide convenience in dosing, distribution, transportation and storage showing improved results than conventional niosomes. This review focuses on different aspects of proniosome such as preparation, characterization, drug release, applications, merits, demerits, present scenario in market and future trends.

2. KEYWORDS: Niosomes, Liposomes, Proniosomes, Nanotechnology.

3. INTRODUCTION

In modern days vesicular drug delivery system has gained significance because of their distinct characteristics such as specific size, shape and act as drug reservoirs for numerous drug molecules. The alterations in the composition and surface molecules can alter the drug targeting and release characteristics of the drug molecule. The vesicular drug delivery system generates a sustain release of the drug molecule and prevents the toxicity. Hence, the present investigation focuses on the development of proniosomal formulation of Bimatoprost that can act as a carrier for enhancing the pharmacokinetic and pharmacodynamic characteristics of Bimatoprost. However, there is availability of numerous dosage forms such as drops, ointments; gels etc… for topical application, a majority of them suffer with drawbacks such as poor bioavailability, local irritation and dosage adjustment.[4-7] In addition, the above mentioned drawbacks might be the result of specific mechanisms such as reflux blinking, tear turnover, lacrimation, and drainage of tear which results in rapid clearance of drug from the surface of eye. Apart from these, the patient is advised to administer frequently for maintaining the steady state concentrations which generates inconvenience for the patient. The structural configuration of niosomes signifies the existence of an internal cavity for entrapment of hydrophilic molecules and provision at external surface for the attachment of lipophilic molecules. The lipid bilayer system provides the facility for entrapment of drug molecules at either core or at the surface depending on its affinity towards hydrophilic phase or towards the lipophilic phase.[8-10] The bilayer system encourages the drug penetration and enhances the availability of drug concentrations at the targeted site and furthering it prevents the drug degradation in the presence of lacrimal enzymes and serves as a promising carrier for ocular drug delivery.[11-12]

In connection to the above, pronisomes can be administered as drops with enhanced patient compliance and superior drug load. Bimatoprost is a synthetic prostamide analog with ocular hypotensive activity. It is recommended to administer once daily in the evening and more frequent administration causes a rapid decline in the intraocular pressure lowering. The therapeutic activity starts after 4 hrs of the administration and reaches to a peak level within 8 to 12 hours. The pharmacokinetic parameters revel that Bimatoprost reaches the peak blood concentrations within 10minutes of administration and nearly 12% of bimatoprost remains unbound in the plasma which means that nearly 88% of the drug is plasma bounded.[13-15] Hence there arises a necessity to develop a suitable formulation that can effectively target the required site with minimum concentrations and generates a sustained effect. Therefore, the present investigation aims at developing a proniosomal formulation of bimatoprost for fulfilling the predetermined safety and therapeutic attributes.[16] The present investigation incorporates span 60 as surfactant possessing elevated HLB value and leads to formation of bilayer vesicles. The composition also highlights cholesterol, a steroidal metabolite of cell membranes for imparting the rigidity and orientation of bi layers in

niosomes. When cholesterol is incorporated with non-ionic surfactants, it reduces the agglomeration and enhances the stability of niosomes. Cholesterol also prevents the gel to liquid phase transition of bi layers which reduces the drug leakage through vesicles and enhances the entrapment efficacy of drug molecules.[12-15]

Apart from the above, the current exploration use Bimatoprost as a carrier that play a significant role in deciding the flexibility and optimization of formulation. The objective of current investigation is to formulate and optimize a stable, biocompatible, biodegradable and non-toxic niosomal formulation and evaluate its various parameters in terms of efficacy and predictability.[16-17] Therefore, the crucial parameters such as drug content, entrapment efficacy, and vesicle size are to be optimized which in turn depend on concentrations of cholesterol and span 60. In order to fulfill the desired criteria, the investigation adopts three square factorial design for optimization of cholesterol and span 60 at three different levels i.e. low, medium, and high using design expert® software trial version and the corresponding formulations are analyzed. In continuation to the above, niosomes possesses enhanced chemical stability and low material cost in comparison to other vesicular drug delivery and proved to be useful for commercial production. Hence, the future aspectsof niosomes lie in encapsulation of various drug moleculesthat serves as a promisingcarrierin achieving desired bioavailability and drug targeting characteristics with decreased toxicity and side effects.

Controlled release dosage forms are commonly used now a days. They have a prolonged action formulations which gives continues release of their active ingredients at a predeter- mined rate and for a predetermined time.

The essential objective for the development of controlled release dosage forms are used to prolong the increase the duration of action and thus gives assurance for high patient compliance. There are four types of controlled drug delivery systems;

1. Rate Pre-programmed DDS
2. Activation modulated DDS
3. Feedback regulated DDS
4. Site targeted DDS[2]

Today, number of approaches has developed for various routes of administration, to achieve either controlled or targeted delivery. Vesicular drug delivery is one of the method which encapsulate the drug eg. Liposomes, niosomes, transferosomes, pharmacosomes, and provesicles like proliposomes and proniosomes. Vesicular system like Liposomes or Niosomes has specific importance while avoiding demerits associated with conventional dosage forms because the particulate carriers can act as drug reservoirs, but these particulate carriers has disadvantages rather than advantages.[18]

To overcome these disadvantages vesicular system of proniosomes are introduced. Proniosomes are water soluble particles that are coated with surfactant and can be hydrated to form niosomal dispersion instantly before use on brief agitation with hot aqueous substance. These proniosomes has more convenience of the transport, distribution, storage and designing would be dry niosomes a promising industrial product.

Hence, dry niosomes can be manufactured which are often called as proniosomes and it avoids many problems related with niosomes like physical stability. Proniosomes can be hydrated instantaneously before use to give niosomal dispersion.

Proniosomes are dry, free flowing granular materials which upon hydration gives multi lamellar niosomal dispersion.[19] This Proniosomal drug delivery has attracted towards transdermal drug delivery because surfactants themselves act as permeation enhancers and are biodegradable, non-toxic, amphiphillic, possess property of encapsulation and they can entrap both hydrophilic and lipophilic drugs as shown in fig 1.

Structure of proniosomes

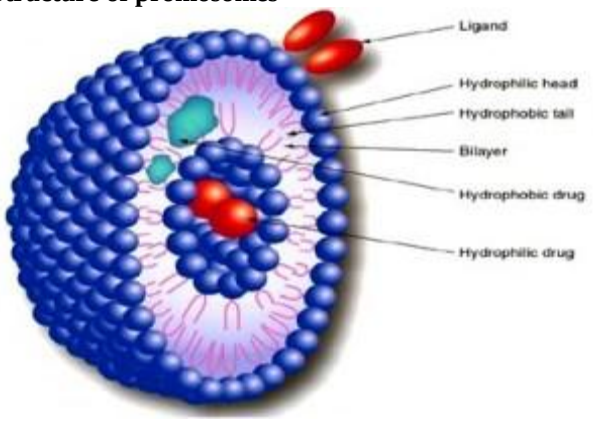

Figure 1: Structure of proniosomes.

Proniosomes are in transparent, translucent or semisolid gel structure because of limited solvent presence and these are mixture of liquid crystals like lamellar, hexagonal, and cubic as shown in (fig 2). Here lamellar phase shows sheets of surfactants arranged in bilayer, hexagonal phase show cylindrical compact structure arranged in hexagonal fashion where as cubic phase contains of curved continuous lipid bilayer extending to three dimentions. While formulating this gel, in the beginning, less viscous composition is developed in some cases but addition of water leads to interaction between water and polar group of surfactant results in swelling of bilayer. If amount of solvent is increased further, then a spherical structure is formed i.e., mult-ilamellar, multi-vesicular. This leads to complete hydration thereby development of Niosomes.

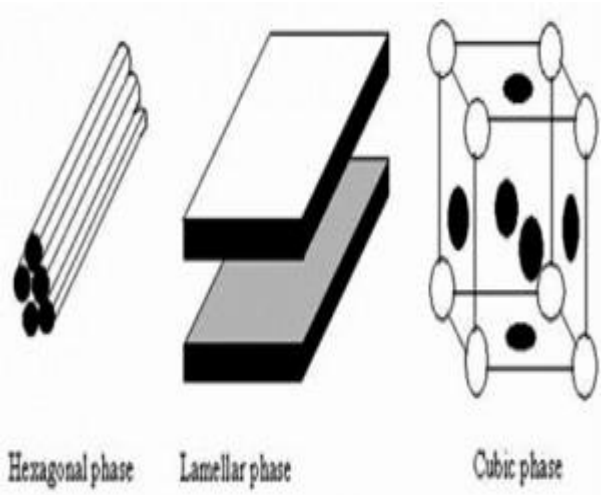

Figure 2: Schematic representation of various liquid crystalline phases.

Advantages of proniosomes over the niosomes

- Avoiding problem of physical stability like aggregation, fusion and leaking
- Avoiding hydrolysis of encapsulated drugs which limiting the shelf life of the dispersion[20]

4. Mechanism of drug permeation of vesicles through skin

- Absorption and fusion of vesicles onto skin surface leads to increase in thermodynamic activity gradient of the drug at interface, which act as driving force for absorption of lipophilic drugs across stratum corneum.
- Alteration in the structure of stratum corneum is also type of interaction involved in the ultra-structural changes in the intracellular lipid region of the skin and its deeper layers which is revealed by freeze fracture electron microscopy and small angle x-ray scattering.
- Bilayer present in niosomes act as rate limiting barrier for drugs.
- Proniosomes consists both non-ionic surfactants and phospholipids, both can act as permeation enhancers and useful in increasing penetrability of many drugs.
- The permeation enhancers effect of vesicles leads to decrease stratum corneum barrier properties.

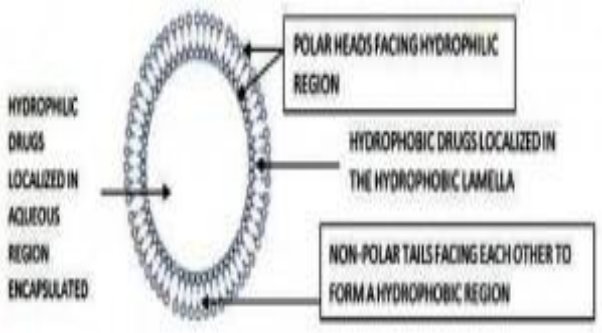

Figure 3: Regions of drug penetration in proniosomes.

Factors affecting penetration of vesicles

- Nature of drug
- Size and composition of vesicles
- Bio physical factors

Proniosomes as drug carriers: The proniosomes are capable drug carriers, because they possess greater chemical stability and lack of many disadvantages associated with liposomes. It has extra merits with niosomes are low toxicity due to non-ionic nature, nor requirement of special safety measures and conditions for formulation and preparation. Niosomes has shown advantages as drug carriers, such as low cost and chemical stability as compared to liposomes but they are connected with problems related to physical stability like fusion, aggregation, sedimentation and outflow and storage.[21]

Proniosomes are dry formulations of surfactant coated carrier vesicles which can be measured out as essential and rehydrated by short agitation in hot water the resulting niosomes are very similar to conventional niosomes and more consistent size.[14] These proniosomes are minimize the problems using dry, free flowing product which is more even during storage and sterilization and it has extra merits of easy of transfer, distribution, measuring and storage make proniosomes a pronouncing versatile delivery system.

Suitability of drug to the proniosomes: Various categories of drugs selections for proniosomes formation based upon the below mentioned points,

- Low Aqueous solubility drugs
- Increased dosage frequency drugs
- Low half-life
- Controlled drug delivery suitable drugs
- Higher adverse drug reactions drugs

5. Preparation of proniosomes: Proniosomes are manufactured by two methods

5.1. Slurry method

5.2 Coacervation Phase Separation Method

5.3 Spray coated method

1. **Slurry method:** The slurry method is developed to create proniosomes using maltodextrin as carrier. The time required to produce proniosomes. This method is independent ratio of surfactant solution to carrier substance. The whole volume of surfactant solution is added to maltodextrin powder is a rotary evaporator and vacuum applied until the powder appears to be dry and free flowing.[22]

2. **Coacervation phase separation method:** This process is widely adopted to prepare Proniosomal gel. Specifically weighed amounts of surfactant, lipid and drug are taken in a clean and dry wide mouthed glass vial of 5.0 ml capacity & alcohol (0.5 ml) is added to it. After warming, all the ingredients are mixed well with a glass rod; the open end of the glass bottle is enclosed with a lid to avoid the loss of

solvent from it and warmed over water bath at 60-70°C for about 5 min until the surfactant mixture is dissolved completely. Then the aqueous phase (0.1% glycerol solution) is added and warmed on a water bath till a clear solution was formed which is then transformed into Proniosomal gel on cooling.

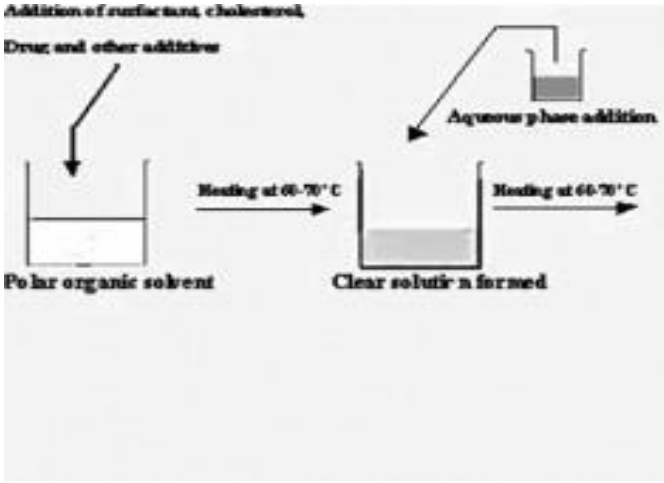

Figure 4: Diagrammatic representation for preparation of proniosomal gel by coaservation phase separation technique.

3. **Spray coated method:** Proniosomes are generally prepared by spraying surfactant in organic solvent into sorbitol powder and evaporating the solvent. It is essential to repeat the process until the required surfactant load has been achieved. The surfactant coating on the carrier formed and a very thin layer and hydration of the coating allows multi lamellar vesicles to form. By adding drug to the surfactant mixture earlier to spraying the solution on to the sorbitol or aqueous solution.[23-26]

Separation of free un entrapped drug: The encapsulation efficiency of proniosomes is determined after separation of the un entrapped drug using these techniques:

1. Dialysis
2. Gel filtration.
3. Centrifugation

1. **Dialysis:** The aqueous niosomal dispersion is dialysed tubing against suitable dissolution medium at room temperature. Then samples are withdrawn from the medium at suitable time period centrifuged and analysed for drug content using UV spectroscopy.
2. **Gel filtration:** The free drug is isolated by gel filtration of niosomal dispersion through a sephadex G50 column and removed with suitable mobile phase and analysed with suitable analytical techniques.
3. **Centrifugation:** The niosomal suspension is centrifuged and the surfactant is removed. The pellet is washed and then re suspended to obtain a niosomal suspension free from un entrapped drug.

In-vitro drug release from proniosomal vesicles: In vitro drug release and skin permeation studies for proniosomes were determined by different techniques like

1. Franz diffusion cell
2. Dialysis tubing
3. Reverse dialysis

1. **Franz diffusion cell:** Franz diffusion cell has a donor chamber fitted with a cellophane membrane. The proniosomes are positioned in it and dialysed against a suitable dissolution medium at room temperature. The drug substance is analysed using suitable method (UV Spectroscopy, HPLC) maintenance of sink conditions is essential.
2. **Dialysis tubing[23]:** This apparatus has prewashed dialysis tubing which can be hermetically sealed. The proniosomes are placed in it and then dialysed against a proper dissolution medium at a room temperature. The samples are withdrawn from the medium at suitable time intervals. Centrifuged and analysed for drug content using suitable method (UV Spectroscopy, HPLC).
3. **Reverse dialysis:** In this apparatus a number of small dialysis tubes containing 1 ml of dissolution medium are placed. Then proniosomes then displaced into the dissolution medium. The direct dilution of the proniosomes is possible with this method. But the rapid release cannot be quantified using this technique.[27-30]

Characterisation of proniosomes: Proniosomes are characterized for vesicle size, size distribution, shape and surface morphological studies.

1. **Measurement of angle of repose:** The angle of repose of dry proniosomes powder was measured by a funnel method. The proniosomes powder was poured into a funnel which was fixed at a position so that the 13mm outlet orifice of the funnel is 5cm above a level black surface. The powder flows down from the funnel to form a cone on the surface and the angle of repose was then calculated by measuring the height of the cone and the diameter of its base.
2. **Scanning electron microscopy:** Particle size of proniosomes is very important characteristic. The surface morphology (roundness, smoothness and formation of aggregates) and the size circulation of proniosomes were studied by Scanning Electron Microscopy (SEM). Proniosomes were sprinkled on to the double-sided tape that was affixedon aluminum stubs. The aluminum stub was placed in the vacuum chamber of a scanning electron. The samples were observed for morphological characterization using a gaseous secondary electron detector (working pressure: 0.8torr, acceleration voltage: 30.00 KV) XL 30, (Philips, Netherlands).
3. **Optical microscopy:** The niosomes were mounted on glass slides and viewed under a microscope with a magnification of 1200X for morphological observation after suitable dilution. The

photomicrograph of the preparation also obtained from the microscope by using a digital SL camera

4. **Measurement of vesicle size:** The vesicle dispersions were diluted about 100 times in the same medium used for their preparation.Vesicle size was measured on a particle size analyzer (Laser diffraction particle size analyzer, Sympatec, Germany). The apparatus consists of a He-Ne laser beam of 632.8 nm focused with a minimum power of 5mW using a Fourier lens [R-5] to a point at the centreof multi element detector and a small volume sample holding cell (Su cell). The sample was stirred using a stirrer before determining the vesicle size. Hu C. and Rhodes in 1999 reported that the average particle size of proniosomes derived niosomes is approximately 6µm while that of conventional niosomes is about 14µm.[31-33]
5. **Entrapment Efficiency:** The vesicles obtained after removal of unentrapped drug by dialysis is then resuspended in 30% v/v of PEG 200 and 1 ml of 0.1%v/v triton x-100 solution was added to solubilize vesicles, the resulted clear solution is then filtered and analysed for drug content. The percentage of drug entrapped is calculated by using the following formula.[33]

EE% = total drug-diffused drug ÷ total drug × 100

In-vitro methods for the assessment of Drug Release from Proniosomes: *In vitro* drug release can be done by (Chen DB *et al.*, 2001);

1. Dialysis tubing
2. Reverse dialysis
3. Franz diffusion cell

1. **Dialysis tubing:** Muller et al., in 2002 studied in vitro drug release could be achieved by using dialysis tubing. The proniosomes is placed in washed dialysis tubing which can be hermetically preserved. The dialysis sac is then dialyzed against a suitable dissolution medium at room temperature; the samples are taken from the medium at suitable intervals, centrifuged and analysed for drug content using suitable method (U.V. Spectroscopy, HPLC etc.). The maintenance of sink condition is essential.
2. **Reverse dialysis:** In this technique, a number of small dialysis as containing 1ml of dissolution medium are placed in proniosomes. The proniosomes are then displaced into the dissolution medium. The direct dilution of the proniosomes is possible with this method; however the quick release cannot be quantified using this method.
3. **Franz diffusion cell:** The in vitro diffusion studies can be performed by using Franz diffusion cell. Proniosomes is placed in the donor chamber of a Franz diffusion cell fitted with a cellophane membrane. The proniosomes is then dialyzed against a suitable dissolution medium at room temperature; the samples were withdrawn from the medium at suitable intervals and analysed for drug content using suitable method (U.V Spectroscopy, HPLC, etc). The maintenance of sink condition is essential.

Drug release kinetic data analysis: The release data obtained from various formulations were studied further for their fitness of data in various kinetic models like Zero order, Higuchi's and Peppa's. In order to understand the kinetic and mechanism of drug release, the result of *in-vitro* drug release study of Niosome were fitted with different kinetic equation like zero order (Equation 1) as cumulative percentage release vs time, Higuchi's model (Equation 2) as cumulative percentage drug release vs square root of time. r^2 and k values were calculated for the linear curve obtained by regression analysis of the above plots.

$C = k_0 t$ …..(1)

Where k_0 is the zero order rate constant expressed in units of concentration / time and t is time in hours.

$Q = kHt1/2$ …..(2)

Where kH is higuchi's square root of time kinetic drug release constant.

To understand the release mechanism *in-vitro* data was analyzed by peppa's model **(**Equation 3**)** as log cumulative % drug release vs. log time and the exponent n was calculated through the slope of the straight line.

$Mt / M\infty = btn$ …..(3)

Where Mt is amount of drug release at time t, $M\infty$ is theoverall amount of the drug, b is constant, and n is the release exponent indicative of the drug release mechanism. If the exponent n = 0.5 or near, then the drug release mechanism is Fickian diffusion and if n have value near 1.0 then it is non-Fickian diffusion.

Osmotic shock: The change in the vesicle size can be determined by osmotic studies. Niosomal preparations are incubated with hypotonic, isotonic, hypertonic solutions for 3 hours. Then the changes in the size of vesicles in the formulations are viewed under optical microscopy.

Stability studies: To determine the stability of proniosomes, the optimized batch was stored in airtight preserved vials at different temperatures. Surface characteristics and percentage drug engaged in proniosomes and proniosomes derived niosomes were selected as parameters for evaluation of the stability, since instability of the formulation would reflect in drug leakage and a decrease. In the percentage drug retained. the proniosomes were sample at regular intervals of time (0, 1, 2 and 3 months), observed for color change, surface characteristics and tested for the percentage drug retained after being hydrated to form niosomes and analysed by suitable analytical methods (UV Spectroscopy, HPLC methods etc.)

Zeta potential analysis: Zeta potential analysis is done for determining the colloidal properties of the preparation. The properly diluted proniosomes derived niosome dispersion was determined using zeta potential analyzer based on Electrophoretic Light Scattering and

Laser Doppler Velocimetry method (Zetaplus™, Brookhaven Instrument Corporation, New York, USA). The temperature was set at 25°C. Charge onvesicles and their mean Zeta Potential values with standard deviation of 5 measurements were obtained directly from the measurement.

6. **Applications of proniosomes:** The application of niosomal technology is widely varied and can be used to treat a number of diseases. The following are the few uses of niosomes which are either proven or under research;

1. **Drug targeting:** One of the most useful aspects of niosomes is their capability to target drugs. Niosomes can be used to target drugs to the reticulo-endothelial system. The reticulo-endothelial system (RES) preferentially takes up niosome vesicles. The uptake of niosomes is controlled by circulating serum factors called opsonins. These opsonins mark the niosome for consent. Such localization of drugs is utilized to treat tumors in animals recognized to metastasize to the liver and spleen. This localization of drugs can also be used for treating parasitic infections of the liver. Niosomes can also be utilized for targeting drugs to organs other than the RES. A carrier system (such as antibodies) can be attached to niosomes to target them to specific organs. Many cells also possess the intrinsic ability to identify and bind specific carbohydrate determinants and this can be exploited by niosomes to direct carrier system to particular cells.[34-36]

2. **Anti-neoplastic treatment:** Most anti neoplastic drugs cause high side effects. Niosomes can adjust the metabolism; extend circulation and half-life of the drug, thus decreasing the side effects of the drugs. Niosomal entrapment of Doxorubicin and Methotrexate showed beneficial effects over the un entrapped drugs, such as decreased rate of propagation of the tumour and higher plasma levels accompanied by slower elimination.

3. **Leishmaniasis:** Leishmaniasis is a disease in which a parasite of the genus Leishmania invades the cells of the liver and spleen. Usually prescribed drugs for the treatment are derivatives of antimony, which in higher concentrations can cause cardiac, liver and kidney damage. Use of niosomes in tests conducted to show that it was possible to administer higher levels of the drug without the triggering of the side effects and thus authorize greater efficacy in treatment.[37-38]

4. **Delivery of peptide drugs:** Oral peptide drug delivery has long been faced with a challenge of by passing the enzymes which would breakdown the peptide. Use of niosomes to successfully defend the peptides from gastrointestinal peptide breakdown is being investigated. In an *in vitro* study conducted by *Yoshida et al.*, oral delivery of a vasopressin derivative entrapped in niosomes showed that entrapment of the drug significantly increased the stability of the peptide.

5. **Uses in studying immune response:** Brewer and Alexander in 1992 studied niosomes are used in studying immune response due to their immunological selectivity, low toxicity and higher stability. Niosomes are being used to study the nature of the immune response provoked by antigens.

6. **Niosomes as carriers for haemoglobin:** Moser P. and Marchand Arvier M. in 1989 reported that niosomes can be used as carriers for haemoglobin within the blood. The niosomal vesicle is permeable to oxygen and hence, can act as a carrier for haemoglobin in anaemic patients.

7. **Transdermal drug delivery systems utilizing niosomes:** One of the most useful aspects of niosomes is that they greatly develop the uptake of drugs through the skin. Transdermal drug delivery utilizing niosomal technology is widely used in cosmetics; In fact, it was one of the first uses of the niosomes. Topical use of niosome entrapped antibiotics to treat acne is done. The penetration of the drugs through the skin is greatly increased as compared to un-entrapped drug.

Recently, transdermal vaccines utilizing niosomal technology is also being researched. A study conducted by P.N. Gupta *et al.*, has shown that niosomes can be utilized for topical immunization using tetanus toxoid. However, the current technology in niosomes allows only a weak immune response, and thus more research needs to be done in this field.[39-42]

7. **Other applications**

i. **Sustained release:** Azmin et al., suggested the role of liver as a depot for methotrexate after niosomes are taken up by the liver cells. Sustained release action of niosomes can be applied to drugs with low therapeutic index and low water solubility since those could be maintained in the circulation via niosomal encapsulation.

ii. **Localized drug action:** Drug delivery through noisome is one of the approaches to achieve localized drug action, since their size and low penetrability through epithelium and connective tissue keeps the drug localized at the site of administration. Localized drug action results in improvement of efficacy of potency of the drug and at the same time reduces its systemic toxic effects e.g. Antimonial encapsulated within niosomes are taken up by mononuclear cells resulting in localization of drug, increase in potency and hence, decrease both indose and toxicity. The evolution of niosomal drug delivery technology is still at an infancy stage, but this type of drug delivery system

has shown promise in cancer chemotherapy and anti-leishmanial therapy.[43-46]

8. CONCLUSION

Proniosomes are a novel and efficient approach to drug delivery. Their vesicular membrane is mainly composed of nonionic surfactants and cholesterol, and the enclosed interior usually contains a buffer solution at appropriate pH. Proniosomes may be prepared by various methods, which affect their formations along with the properties of the drug, cholesterol content, and amount, structure, and type of surfactant. They improve the stability of the entrapped drug during delivery. They do not require special conditions for handling, protection, storage, or industrial manufacturing. In addition, they can be prepared with different structural characteristics (composition, fluidity, and size), and can be designed for particular routes of administration. Overall, proniosomes are a very effective tool for drug delivery and targeting of numerous therapeutically active moieties. They have the potential to provide better treatment than conventional drug delivery systems.

9. REFERENCES

1. Linden C, Nuija E, Alm A. Effects on IOP restoration and blood-aqueous barrier after long term treatment with latanoprost in open angle glaucoma and ocular hypertension. Br J Ophthal, 1997; 81: 370–372.
2. Kwon YH, Kim YI, Pereira ML, Montague PR, Zimmerman MB, Alward WL. Rate of optic disc cup progression in treated primary open-angle glaucoma. J Glaucoma, 2003; 12: 409–416.
3. Nordmann JP, Auzanneau N, Ricard S, Berdeaux G. Vision related quality of life and topical glaucoma treatment side effects. Health Qual Life Outcomes, 2003; 1: 75.
4. Mokhtar M, Sammour OA, Hammad MA, Megrab NA Effect of some formulation parameters on flurbiprofen encapsulation and release rates of niosomes prepared form proniosome. Int J Pharm, 2008; 361: 104-111.
5. Alam MI, Baboota S Pharmacodynamic evaluation of proniosomestransdermal therapeutic gel containing Celecoxib. Sci Asia, 2010; 36: 305-311.
6. Gurrapu A, Jukanti R, Bobbala SR, Swetha K, Jee vana JB Improved oral delivery of valsartan from maltodextrin based proniosome powder. Powder Technol, 2011; 23: 583-590.
7. Abdallah M, Sammour O, EL-Ghamry H, et al., Preparation and In-vitro Evaluation of Diclofenac Sodium Niosomal Formulatuion. IJPSR, 2013; 4(5): 1757-1765.
8. Mahmoud M, Omaima AS, Mohammed AH, Nagia AM Effect of some formulation parameters on flurbiprofen encapsulation and release rates of niosomes prepared from proniosomes. Int J Pharm, 2008; 361: 104-111.
9. Chauhan M, Sharma S, Span 60 niosomal oral suspension of fluconazole: Formulation and in vitro evaluation. JPRHC, 2009; 1: 142-156.
10. Uchegbu I.F, Vyas S.P, Non ionic surfactant based vesicles (niosomes) indrug delivery. Int. J. Pharm, 1998; 172: 33-70.
11. Yoshioka T, Sternberg B, Florence AT. Preparation and properties of vesicles (niosomes) of sorbitan mono-esters (span 20, 40, 60, 80) and a sorbitan trimester (span 85), Int J. Pharma, 1994; 105: Y6.
12. Solankia A, Preparation, characterization, optimization, and stability studies of aceclofenac proniosomes. Iranian Journal of Pharmaceutical Research, 2008; 7(4): 237-246.
13. Reza mozafari M, Bioactive entrapment and targeting using nanocarrier technologies: An introduction Frontiers of nanotherapy. University of Michigan, Springer, 2006; 1-6.
14. Shi B, Fang C, Pei Y., Stealth, PEG-PHDCA niosomes: effects of chain length of PEG and Particle size on niosomes surface properties, in vitro drug release, phagocytic uptake, in vivo pharmacokinetics and antitumor activity, J. Pharm. Sci, 2006; 95: 1873–1887.
15. Azmin MN, Flourence AT., Handjani –Vila RM, Stuart FB, Vanlerberghe G, Whittaker JS, The effect of non-ionic surfactant vescicles (niosomes) entrapped on the absorption and distribution of methotextrate in mice, J. Pharm. Pharmacol, 1985; 37: 237-242.
16. Bhaskaran. S and Panigrahi. L. Formulation and evaluation of niosomes using different nonionic surfactant. Indian Journal of pharmaceutical sciences, 2002; 1: 63-65.
17. Rawat AS, Murugesan S, Mahadevan N: Proniosomal gel: A Novel topical delivery system. Int j Adv PR, 2011; 3: 1-10.
18. Ijeoma F, Uchegbu, Suresh Vyas: Non-ionic surfactant based vesicles (niosomes) in drugdelivery. Int J Pharm, 1998; 172: 33–70.
19. Schreief H, Bouwstra J: Liposomes and niosomes as topical drug carriers, dermal and transdermal drug delivery. J Control Release, 1994; 30: 1-15.
20. Baillie AJ, Florence AT, Hume LR, Muirhea GT and Rogerson A: The preparation and properties of niosomes non-ionic surfactant vesicles. J Pharm Pharmacol, 1985; 37: 863-868.
21. Fang JY, Hong CT, Chiu WT, Wang YY: Effect of liposomes and niosomes on skin permeation of Enoxacin. Int J Pharm, 2001; 21: 61–72.
22. Thejaswi C, Rao K M, Gobinath M: A review in Design and Characterization of Proniosomes as drug carrires. Int J Adv Phar and Nanotech, 2011; 1: 16-19.
23. Sagar GH, Arunagirinanthan MA, Bellar JR: Self-assembled surfactant nanostructures important in drug delivery. Indian j Exp Biol, 2007; 45: 133-144.
24. Comells F, sanchez-leal J, Gonzalez JJ: Influence of ionic surfactant on the formation of liquid crystals in

oleic acid/glycol/water systems. Journal of surfactants and detergents, 2007; 10: 137-144.
25. Hu C. and Rhodes D.G. Proniosomes: a noveldrug carrier preparation. Int J Pharm, 1999; 185: 23-35.
26. Almira I, Blazek-Welsh, Rhodes DG: Maltodextrin-Based Proniosomes. AAPSPharmSciTech, 2001; 3(1): 1-9.
27. Blazek-Walsh AI and Rhodes DG: SEM imaging predicts quality of niosomes from maltodextrin-based proniosomes. Pharm Res, 2001; 18: 656-661.
28. Mahdi, Jufri, Effionora, Anwar, Joshita, Djajadisastra: Preparation of MaltodextrinDE 5-10 based ibuprofen Proniosomes. J Pharm, 2004; 1: 10 – 20.
29. Yadav K, Yadav D, Saroha K, Nanda S, Mathur P: "Proniosomal Gel: A Provesicular approach for transdermal drug delivery. Scholars Reasearch Library, 2010; 2: 189-198.
30. Jain NK: controlled novel drug delivery system, 1: 1-26.
31. Rakesh PP, Hardik HP, Ashok HB: Nanoparticle vesicular systems: A versatile tool for drug delivery. Int J Drug Delivery Technology, 2009; 1(2): 42 – 45.
32. Solanki AB, Parikh JR, Parikh RH: Formulation and Optimization of PiroxicamProniosomes by 3-Factor, 3-Level Box-BehnkenDesign. AAPS PharmSciTech, 2007; 8(4): 86.
33. Vora B, Khopade AJ and Jain NK: Proniosomes based transdermal delivery of levonorgestrel for effective contraception. J Control Rel, 1998; 54: 149-165.
34. Ajay Solanki, Jolly Parikh and RajeshParikh. Preparation, Characterization, optimization, and stability studies of AceclofenacProniosomes. Iranian J Pharm Research, 2008; 7(4); 237-246.
35. Khandare JN, Madhavi G, Tamhankar BM. Niosomesnovel drug delivery system. The Eastern Pharmacist, 1994; 37: 61-64.
36. Gayatri Devi S, Venkatesh P and Udupa N. Niosomalsumatriptan succinate for nasal administration. Int J Pharm Sci, 2000: 62(6): 479-481.
37. Hu C and Rhodes DG: Proniosomes: a novel drug carrier preparation. Int J Pharm, 1999; 185: 23-35.
38. Vyas SP, Khar RK: Niosomes- Targeted and Controlled Drug delivery, 2002; 1: 249 – 279.
39. Muller RH, Radtke M, Wissing SA: Solid lipid nanoparticles (SLN) and nanostructured lipid carriers (NLC) in cosmetic and dermatological preparations. Adv Drug Deliv Rev, 2002; 54: 131–155.
40. Alsarra A, Bosela AA, Ahmed SM and Mahrous GM. Proniosomes as a drug carrier fortransdermal delivery of ketorolac. Eur J Pharm and Biopharm, 2004; 105: 1-6.
41. ChandraprakashK S: pharmacokinetic evaluation of surfactant vesicles containing methotrexate intumor bearing mice. International journal of Pharmacy, 1990; 61: 11.
42. Puglia C, Trombetta D, Venuti V, Saija A, Bonina F: Evaluation of in vivo topicalanti-inflammatory activity of indometacin fromliposomal vesicles. J Pharm Pharmacol, 2004; 56: 1225-1232.
43. Gibaldi M and Perrier D: Pharmacokinetics 2nd edi, New York, Marcel Dekker, Inc, 1982.
44. Biju SS, Talegaonkar S, MisraPR and Khar RK: Vesicular systems: An overview. Indian J Pharm Sci, 2006; 68: 141-153.
45. Malhotra M and Jain NK. Niosomes as Drug Carriers. Indian Drugs, 1994: 31(3): 81-86.
46. Junyaprasert, Veerawat Teeranachaideekul and Tasaneeya: Effect of Charged and Non-ionic Membrane Additives on Physicochemical Properties and Stability of Niosomes. AAPS Pharm Sci Tech, 2008; 9(3): 851-859.