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Bilosomes: An Emerging Vesicular Carrier for Enhanced Drug Delivery

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Abstract

Bilosomes have emerged as an innovative vesicular drug delivery system that leverages the unique properties of bile salts integrated into lipid bilayers. This incorporation imparts increased structural integrity, improved stability in the gastrointestinal environment, and enhanced permeability across biological membranes. Unlike traditional vesicular systems such as liposomes and niosomes, bilosomes exhibit exceptional resilience against enzymatic and bile salt degradation, making them especially suitable for oral and transdermal administration.

This review aims to provide a comprehensive overview of bilosomes, detailing their structural composition, preparation methods, and underlying mechanisms that contribute to their superior drug delivery capabilities. The presence of bile salts not only enhances vesicle deformability and mucosal adhesion but also facilitates paracellular transport by modulating tight junctions. These characteristics significantly improve the bioavailability of encapsulated drugs, particularly macromolecules like peptides, proteins, and vaccines that are typically vulnerable to degradation in conventional delivery systems.

Moreover, bilosomes have demonstrated versatility in delivering a wide range of therapeutic agents, including anticancer drugs, anti-inflammatory compounds, and immunomodulators. Recent advancements in nanotechnology have further refined bilosome formulations through surface modifications, polymeric coatings, and targeted ligand attachment, opening avenues for site-specific and combination therapies.

Despite their promise, challenges such as large-scale production, long-term stability, and regulatory hurdles remain. This review also discusses these limitations and outlines future directions for overcoming them. With continued research and development, bilosomes hold significant potential for transforming modern drug delivery strategies and enhancing therapeutic outcomes across various clinical applications.

Keywords: Bilosomes, Vesicular Drug Delivery, Bile Salts, Oral Delivery, Transdermal Delivery, Nanocarriers, Bioavailability, Controlled Release

1. INTRODUCTION

The development of effective drug delivery systems is a critical component in the advancement of modern therapeutics. Conventional drug administration routes, particularly oral and transdermal, are

often associated with multiple challenges, including enzymatic degradation in the gastrointestinal (GI) tract, first-pass hepatic metabolism, erratic absorption profiles, and poor patient adherence due to dosing frequency or side effects. These limitations contribute to reduced therapeutic efficacy and necessitate the development of advanced delivery systems that can protect the active pharmaceutical ingredient (API), control its release, and enhance its absorption.[1]

To address these issues, a variety of vesicular carriers such as liposomes, niosomes, and ethosomes have been explored for their potential to encapsulate drugs and improve pharmacokinetics and biodistribution. While liposomes and niosomes have demonstrated success in delivering both hydrophilic and lipophilic drugs, they are often susceptible to destabilization by bile salts, digestive enzymes, and pH fluctuations in the GI tract. This restricts their efficacy, particularly for oral drug delivery.[2]

In this context, bilosomes have emerged as a novel and promising vesicular system. These vesicles are similar in architecture to liposomes but are uniquely characterized by the incorporation of bile salts such as sodium deoxycholate or sodium taurocholate within the phospholipid bilayer. The inclusion of bile salts offers several functional advantages: it enhances the structural stability of the vesicles against GI degradation, improves membrane fluidity, and facilitates the transport of encapsulated drugs across epithelial barriers by transiently opening tight junctions and promoting both paracellular and transcellular absorption. [3]

Bilosomes also exhibit excellent mucoadhesive properties, prolonging their retention at mucosal sites such as the intestinal epithelium, which further aids in sustained drug release and increased bioavailability. These characteristics make bilosomes particularly attractive for delivering sensitive macromolecules such as vaccines, peptides, and proteins that are otherwise poorly absorbed and rapidly degraded in the GI environment.[4]

Moreover, the biocompatibility and adaptability of bilosomes extend their application beyond oral delivery. They have shown potential in transdermal, ocular, nasal, and pulmonary delivery systems, enabling non-invasive administration of a variety of therapeutics. Recent advancements have also introduced nano-sized bilosomes (nano-bilosomes) and surface-modified bilosomes aimed at targeted delivery and improved cellular uptake. [5]

Despite these promising features, challenges related to scale-up production, long-term stability, and regulatory pathways must be addressed before bilosomes can be widely adopted in clinical settings. Nonetheless, the growing body of preclinical and clinical evidence suggests that bilosomes hold substantial promise as a next-generation vesicular carrier capable of revolutionizing drug delivery.

2. COMPOSITION OF BILOSOMES

Bilosomes are specialized vesicular carriers structurally similar to liposomes but uniquely modified by the inclusion of bile salts in their bilayer membrane. This modification offers significant advantages in terms of stability, permeability, and bioavailability of encapsulated therapeutics, especially when administered via oral or mucosal routes.

2.1 Composition of Bilosomes

The fundamental components of bilosomes include:

Non-ionic surfactants: These are the primary building blocks of the bilayer structure and determine the vesicle's integrity, stability, and biocompatibility. Commonly used surfactants include:[6]

Span series (e.g., Span 60, Span 80): These have a high phase transition temperature, contributing to the formation of rigid and stable vesicles.

Tween series (e.g., Tween 20, Tween 80): Often used to adjust the hydrophilic-lipophilic balance (HLB) and improve dispersion characteristics.

Cholesterol: Incorporated into the bilayer to provide membrane rigidity and mechanical strength. Cholesterol helps to stabilize the vesicles by reducing membrane fluidity and permeability, thus preventing premature leakage of the encapsulated drug.[7]

Bile salts: The defining component of bilosomes, bile salts such as sodium deoxycholate, sodium taurocholate, and sodium glycocholate are amphipathic molecules that mimic physiological surfactants present in the gastrointestinal tract. Their inclusion provides the bilosomes with:[8]

- Resistance to degradation by endogenous bile salts and enzymes in the gut.
- Improved membrane flexibility and fusion with epithelial cells.
- Enhanced penetration and absorption of drugs via mucosal tissues.

The ratio and type of each component significantly affect vesicle properties such as particle size, zeta potential, entrapment efficiency, and drug release kinetics.

2.2 Preparation Methods of Bilosomes

The method of preparation greatly influences the physicochemical characteristics of bilosomes, such as size distribution, lamellarity, surface charge, and encapsulation efficiency. Several techniques have been developed and optimized:

a. **Thin-Film Hydration Method:**
This is one of the most widely used and straightforward methods. The surfactants, cholesterol, and bile salts are dissolved in an organic solvent (e.g., chloroform, methanol), which is then evaporated under reduced pressure to form a thin lipid film on the walls of a rotary flask. This film is subsequently hydrated with an aqueous phase containing the drug of interest. The hydration leads to the spontaneous formation of multilamellar vesicles, which can be downsized using sonication or extrusion.[9]

b. **Reverse-Phase Evaporation Method:**
In this technique, a water-in-oil emulsion is created by dissolving lipid components in an organic phase and then emulsifying it with an aqueous phase containing the drug. The removal of the organic solvent under vacuum results in the formation of unilamellar vesicles. This method is particularly useful for encapsulating hydrophilic drugs due to the large aqueous core volume.[10]

c. **Microfluidization:**
A high-shear technique in which lipid components and aqueous drug solution are forced through a microchannel under high pressure, promoting uniform size reduction and vesicle formation. This method allows for better scalability and reproducibility and is suitable for the industrial production of bilosome formulations.[11]

d. **Hot or Cold Homogenization:**
This involves homogenizing the lipid phase with the aqueous drug solution at elevated or reduced temperatures. The application of high-pressure homogenization cycles leads to the production of nanometer-sized bilosomes.[12]

1. MECHANISM OF ACTION

The therapeutic efficacy of bilosomes is largely attributed to their unique ability to enhance the delivery of drugs across biological membranes. This is accomplished through a combination of physicochemical and biological mechanisms that promote drug stability, adhesion, transport, and absorption.[13]

3.1 Membrane Permeation and Absorption Enhancement

Bilosomes are designed to facilitate drug transport through both **paracellular** and **transcellular** pathways:

- **Paracellular Transport:**

One of the hallmark features of bilosomes is the presence of bile salts, which can transiently modulate the tight junctions between epithelial cells. Bile salts such as sodium deoxycholate and sodium taurocholate act as absorption enhancers by loosening the tight junctions, allowing larger and more hydrophilic molecules to pass through the paracellular route. This mechanism is particularly valuable in the gastrointestinal tract, where tight junctions often limit the passage of macromolecules and hydrophilic drugs.[14]

- **Transcellular Transport:**

Due to their amphiphilic nature, bilosomes can also undergo endocytosis and facilitate drug entry via the transcellular route. The lipid bilayer of the bilosomes merges with the cell membrane, enabling the encapsulated drug to be released directly into the cytoplasm. This process enhances the cellular uptake of poorly permeable drugs, especially peptides, proteins, and nucleic acids.[15]

3.2 Protection from Enzymatic Degradation

Another critical aspect of bilosome action is their ability to protect encapsulated drugs from degradation in hostile environments. In the gastrointestinal tract, enzymes such as proteases, lipases, and nucleases rapidly degrade therapeutic molecules like peptides and proteins. The bilosomal vesicle acts as a physical barrier, shielding the drug until it reaches the site of absorption. Additionally, bile salts stabilize the bilosome membrane, making it more resistant to enzymatic attack. [16]

3.3 Mucoadhesion and Prolonged Residence Time

Bilosomes exhibit mucoadhesive properties due to their surface charge and composition. The bile salts and surfactants present in the bilosome structure can interact with mucin and glycoproteins in the mucosal lining. This interaction enhances the retention time of bilosomes at the site of absorption, whether in the gastrointestinal tract (for oral delivery), on the skin (for transdermal delivery), or on ocular and nasal mucosa.[17]

Prolonged residence at the absorption site allows for:

- Sustained drug release
- Increased drug concentration gradient across the membrane
- Improved bioavailability of the therapeutic agent

3.4 Immune System Interaction

When used for vaccine delivery, bilosomes can stimulate both systemic and mucosal immune **responses**. The bile salts facilitate uptake by M-cells in the Peyer's patches of the intestine, promoting antigen presentation and immune activation. This makes bilosomes an attractive platform for oral immunization, as they can deliver antigens directly to gut-associated lymphoid tissue (GALT). [18]

3.5 Enhanced Skin Permeation (Transdermal Delivery)

For transdermal applications, bilosomes improve drug penetration through the stratum corneum. The deformable nature of bilosomes—attributable to bile salts and surfactantsallows them to squeeze through intercellular lipid pathways in the skin, carrying the drug into deeper dermal layers. This improves both local and systemic delivery without the need for invasive methods. [19]

2. ADVANTAGES OF BILOSOMES OVER CONVENTIONAL VESICULAR SYSTEMS

Bilosomes offer a series of unique and compelling advantages over traditional vesicular systems such as liposomes, niosomes, and transfersomes. Their enhanced performance, particularly for oral and mucosal delivery, arises from the structural integration of bile salts into the vesicle membrane. The key advantages include:

4.1 Enhanced Stability in Gastrointestinal Fluids

Traditional vesicular carriers often disintegrate in the harsh conditions of the gastrointestinal tract due to the presence of bile salts, enzymes, and varying pH levels. Bilosomes, by contrast, are inherently stabilized by the incorporation of bile salts into their bilayer.[20] This inclusion:

- Increases resistance to bile salt-induced solubilization,
- Minimizes vesicle aggregation or fusion,
- Maintains vesicle integrity in acidic (stomach) and alkaline (intestinal) environments.

4.2 Improved Bioavailability

Bilosomes significantly improve the oral bioavailability of encapsulated drugs. Bile salts enhance membrane permeability by:

- Increasing membrane fluidity,
- Disrupting tight junctions to allow paracellular transport,
- Facilitating fusion with epithelial cells for transcellular drug delivery.

This is especially beneficial for macromolecular drugs like peptides, proteins, and nucleic acids that typically have poor oral absorption.[21]

4.3 Versatile Routes of Administration

Bilosomes have demonstrated utility across multiple administration routes:

- **Oral**: Protection against enzymatic degradation and enhanced intestinal uptake.
- **Transdermal**: Enhanced skin permeation due to deformable vesicle structure.
- **Ocular**: Prolonged residence time and increased corneal permeability.
- **Nasal and Pulmonary**: Rapid absorption and systemic delivery via the respiratory tract.
- **Rectal and Vaginal**: Localized and systemic drug delivery with enhanced mucosal adherence.[22]

4.4 Biocompatibility and Safety

Bilosomes are generally composed of materials recognized as safe (GRAS), including:

- Non-ionic surfactants (e.g., Span and Tween),
- Cholesterol,
- Naturally occurring bile salts.
  These components are non-toxic, non-immunogenic, and biodegradable, making bilosomes suitable for chronic administration and use in sensitive populations. [23]

4.5 High Encapsulation Efficiency

Bilosomes can encapsulate a wide variety of drug molecules, including:

- Hydrophilic drugs (e.g., insulin, vaccines),
- Lipophilic drugs (e.g., curcumin, cyclosporine),
- Biologics (e.g., DNA, proteins).[24]

4.6 Mucoadhesive Properties

The presence of bile salts and surfactants enhances the mucoadhesive nature of bilosomes. This prolongs their retention time on mucosal surfaces such as:

- Intestinal epithelium (oral),
- Nasal mucosa,
- Ocular surfaces,
  Extended residence time improves the drug's interaction with the absorption site and boosts bioavailability. [25]

4.7 Potential for Immune Stimulation

Bilosomes are particularly promising for vaccine delivery. Their ability to target M-cells in Peyer's patches and trigger both mucosal and systemic immune responses make them ideal carriers for oral vaccines against infectious diseases. [26]

4.8 Flexibility in Formulation Design

Formulators can modify bilosomes to suit specific therapeutic needs by:

- Altering surfactant types and concentrations,
- Adjusting bile salt ratios,
- Adding polymers or ligands for targeting or controlled release.

This versatility allows customization for targeted drug delivery, improved patient compliance, and site-specific release. [27]

3. APPLICATIONS OF BILOSOMES

Bilosomes have emerged as a highly versatile drug delivery platform with wide-ranging applications across multiple therapeutic areas. Their ability to improve drug stability, absorption, and bioavailability—particularly through oral and mucosal routes has positioned them as a promising alternative to traditional vesicular systems. The incorporation of bile salts not only enhances gastrointestinal stability but also promotes transport across biological barriers, allowing bilosomes to effectively deliver both small and large molecules.[28]

5.1 Vaccine Delivery

One of the most promising applications of bilosomes is in oral vaccine delivery. Traditional oral vaccines face challenges such as enzymatic degradation and poor mucosal uptake. Bilosomes protect antigens from degradation and facilitate their uptake via M cells in Peyer's patches, leading to the activation of both mucosal (IgA) and systemic (IgG) immune responses.[29]

Examples:

Oral bilosomal formulations of tetanus toxoid, hepatitis B, and cholera antigens have demonstrated strong immunogenicity in preclinical and clinical models.

Bilosomes have also been explored as adjuvant systems, enhancing antigen presentation and immune stimulation without the need for additional adjuvants.

5.2 Delivery of Peptides and Proteins

Peptides and proteins are notoriously unstable in the gastrointestinal tract, suffering from enzymatic degradation and low permeability. Bilosomes offer a protective encapsulation system, allowing these macromolecules to reach the absorption site intact.[30]

Examples:

Insulin, calcitonin, and interferon-α have been successfully encapsulated in bilosomes, showing improved oral bioavailability and prolonged pharmacological effects.

They also hold potential in oral delivery of biologics such as monoclonal antibodies and enzyme replacement therapies.

5.3 Anticancer Drug Delivery

Bilosomes can be engineered to provide site-specific delivery of anticancer agents, minimizing systemic toxicity and enhancing local drug concentration at tumor sites. Their flexible surface modification allows for ligand-based targeting strategies, such as folate or antibody conjugation. [31]

Examples:

Bilosomal formulations of curcumin, paclitaxel, and 5-fluorouracil have demonstrated increased cytotoxicity against cancer cells and reduced off-target effects.

Surface-modified bilosomes have been investigated for targeted colon and breast cancer therapy.

5.4 Anti-inflammatory and Analgesic Drugs

Bilosomes offer controlled and sustained release of anti-inflammatory drugs, improving therapeutic outcomes while minimizing gastrointestinal irritation and systemic side effects commonly associated with NSAIDs.[32]

Examples:

Encapsulation of ibuprofen, naproxen, and diclofenac in bilosomes has shown prolonged anti-inflammatory activity and reduced ulcerogenic potential.

Transdermal bilosomal gels have been developed for localized delivery in arthritic and neuropathic conditions.

5.5 Antimicrobial and Antiviral Applications

The rising resistance to conventional antibiotics and antivirals has led to the exploration of novel delivery systems. Bilosomes improve the therapeutic index of antimicrobial agents by enhancing penetration into infected tissues and ensuring sustained release.[33]

Examples:

Delivery of amphotericin B, ciprofloxacin, and acyclovir via bilosomes has improved pharmacokinetics and reduced toxicity.

Bilosomes have also been tested in topical antifungal and oral antiretroviral formulations with promising results.

5.6 Ocular Drug Delivery

The eye presents several barriers to drug absorption, such as tear turnover and limited permeability of the corneal epithelium. Bilosomes improve drug retention and corneal penetration, making them suitable for ophthalmic use. [34]

Examples:

Bilosomal formulations of timolol, pilocarpine, and dexamethasone have been shown to enhance intraocular delivery and therapeutic efficacy in glaucoma and inflammatory eye conditions.

5.7 Dermatological and Transdermal Delivery

The deformability and small size of bilosomes allow them to traverse the stratum corneum and reach deeper skin layers. This makes them ideal for topical delivery of corticosteroids, antifungals, and cosmeceuticals. [35]

Examples:

Hydrocortisone, ketoconazole, and retinoids have been successfully incorporated into bilosomal gels and creams for improved local action.

5.8 Neurological Applications

Although still in early stages, bilosomes are being investigated for nose-to-brain delivery of neurotherapeutics, bypassing the blood-brain barrier. This approach has potential for treating Parkinson's, Alzheimer's, and other neurodegenerative diseases. [36]

Examples:

Intranasal bilosomal formulations of levodopa, rivastigmine, and resveratrol are being explored for direct CNS targeting with reduced peripheral side effects.

6. Recent Advances in Bilosome Technology

Recent innovations in bilosome-based drug delivery have significantly expanded their potential in pharmaceutical applications. One of the key developments is the creation of nano-bilosomes, which are nanoscale vesicles that offer improved tissue penetration, enhanced cellular uptake, and reduced clearance by the reticuloendothelial system. These nanocarriers have shown promise in cancer therapy, transdermal systems, and ocular applications. Another significant advancement is the surface modification of bilosomes with targeting ligands such as folic acid, transferrin, lectins, and antibodies. These ligand-functionalized bilosomes enable site-specific drug delivery, particularly to tumors, the brain, and mucosal tissues, enhancing therapeutic efficacy while minimizing off-target effects.[37]

In addition to surface engineering, the incorporation of polymers such as chitosan, Carbopol, and PEG has been widely explored to enhance bilosome stability, mucoadhesiveness, and controlled drug release. Chitosan-coated bilosomes, for example, exhibit improved residence time and absorption across mucosal surfaces, making them suitable for nasal, buccal, and vaginal delivery. Moreover, co-encapsulation strategies have gained momentum, enabling the delivery of multiple drugs or a combination of therapeutic agents (e.g., drug plus adjuvant or drug pairs) within a single bilosome. This approach offers synergistic effects, improved treatment outcomes, and simplified dosing regimens. [38,39]

Recent studies have also introduced stimuli-responsive bilosomes that release drugs in response to physiological triggers such as pH, temperature, or specific enzymes. These "smart" bilosomes provide precise, on-demand release profiles, particularly beneficial in targeting tumors or inflammatory sites. Furthermore, hybrid systems integrating bilosomes with hydrogels, micelles, or liposomes are being developed to enhance formulation versatility, patient compliance, and drug stability. On the

technological front, advances in scalable manufacturing processes, including microfluidization and lyophilization, are paving the way for clinical translation. Collectively, these advances underscore the growing importance of bilosomes as a dynamic and adaptable platform for modern drug delivery challenges. [40,41]

7. CHALLENGES AND FUTURE PERSPECTIVES

Despite the promising potential of bilosomes as advanced drug delivery systems, several critical challenges remain before their widespread clinical adoption. One major hurdle is the scalability of production; while laboratory-scale methods such as thin-film hydration and microfluidization provide precise control over vesicle characteristics, translating these techniques into reproducible, cost-effective, and high-throughput manufacturing processes remains complex. Ensuring batch-to-batch consistency and maintaining vesicle integrity at large scales are essential for regulatory approval and commercial viability. Another significant challenge lies in the long-term physical and chemical stability of bilosomal formulations. Although bile salts confer enhanced gastrointestinal stability, bilosomes are still susceptible to aggregation, drug leakage, and degradation during storage, which could compromise their therapeutic efficacy. Developing optimized lyophilization techniques, stabilizing excipients, and packaging solutions are active areas of research to address these concerns. [42,43]

Additionally, the regulatory landscape for bilosome-based products is currently underdeveloped. Due to their hybrid nature—combining liposomal structures with bile salts—there is no clear, standardized pathway for approval, leading to uncertainties in preclinical and clinical evaluation requirements. This regulatory ambiguity necessitates early engagement with authorities and comprehensive safety profiling, including immunogenicity, toxicity, and pharmacokinetic assessments. Furthermore, while preclinical studies have demonstrated encouraging results, there is a pressing need for robust clinical trials to establish the safety, efficacy, and therapeutic superiority of bilosomes in humans across various indications. [44]

Looking ahead, future research should emphasize the development of scalable, GMP-compliant manufacturing processes, improved formulation stability, and detailed mechanistic understanding of bilosome interactions with biological systems. Innovations such as stimuli-responsive bilosomes, targeted delivery, and combination therapies are likely to enhance clinical outcomes but will require rigorous validation. Collaboration between academia, industry, and regulatory bodies will be crucial to overcoming these challenges and unlocking the full therapeutic potential of bilosomes in modern medicine. [45,46]

8. CONCLUSION

Bilosomes represent a novel and versatile vesicular drug delivery system that addresses many limitations associated with conventional carriers such as liposomes and niosomes. By incorporating bile salts into their structure, bilosomes exhibit enhanced stability, improved permeability, and increased resistance to the harsh gastrointestinal environment, making them especially suitable for oral and mucosal delivery of diverse therapeutic agents. Their ability to protect labile molecules, promote targeted and controlled release, and facilitate mucosal immune responses highlights their potential across a broad spectrum of applications—from vaccines and peptides to anticancer and anti-inflammatory drugs. Recent

technological advancements, including nano-sizing, surface modification, and stimuli-responsive designs, have further expanded their utility and therapeutic efficacy.

However, the journey from bench to bedside requires overcoming challenges related to large-scale manufacturing, formulation stability, and regulatory approval. Addressing these obstacles through multidisciplinary research and well-designed clinical studies will be key to realizing the full clinical potential of bilosomes. As the field continues to evolve, bilosomes are poised to become an integral component of next-generation drug delivery platforms, offering safer, more effective, and patient-friendly therapeutic solutions. Ultimately, the convergence of innovative formulation strategies and a deeper mechanistic understanding will accelerate the translation of bilosome technology into impactful clinical applications, advancing personalized medicine and improving patient outcomes worldwide.

REFERENCES:

1. T. Rajput and M. K. Chauhan, “Bilosome: a Bile Salt Based Novel Carrier System Gaining Interest in Pharmaceutical Research,” *Journal of Drug Delivery and Therapeutics*, vol. 7, no. 5, 2017, doi: 10.22270/jddt.v7i5.1479.
2. Sankhyan and P. Pawar, “Recent trends in niosome as vesicular drug delivery system,” *J Appl Pharm Sci*, vol. 2, no. 6, pp. 20–32, 2012, doi: 10.7324/JAPS.2012.2625.
3. R. K. Sahoo, N. Biswas, A. Guha, N. Sahoo, and K. Kuotsu, “Nonionic surfactant vesicles in ocular delivery: Innovative approaches and perspectives,” *Biomed Res Int*, vol. 2014, 2014, doi: 10.1155/2014/263604.
4. J. S. Wilkhu, S. E. McNeil, D. E. Anderson, and Y. Perrie, “Characterization and optimization of bilosomes for oral vaccine delivery,” *J Drug Target*, vol. 21, no. 3, pp. 291–299, 2013, doi: 10.3109/1061186X.2012.747528.
5. Shukla, V. Mishra, and P. Kesharwani, “Bilosomes in the context of oral immunization: Development, challenges and opportunities,” *Drug Discov Today*, vol. 21, no. 6, pp. 888–899, 2016, doi: 10.1016/j.drudis.2016.03.013.
6. J. Montanari, M. Vera, E. Mensi, M. Morilla, and E. Romero, “Nanoberries for topical delivery of antioxidants,” *J Cosmet Sci*, vol. 64, no. 6, pp. 469–481, 2013.
7. G. M. M. El Maghraby, A. C. Williams, and B. W. Barry, “Oestradiol skin delivery from ultradeformable liposomes: Refinement of surfactant concentration,” *Int J Pharm*, vol. 196, no. 1, pp. 63–74, 2000, doi: 10.1016/S0378-5173(99)00441-X.
8. Y. Dai, R. Zhou, L. Liu, Y. Lu, J. Qi, and W. Wu, “Liposomes containing bile salts as novel ocular delivery systems for tacrolimus (FK506): In vitro characterization and improved corneal permeation,” *Int J Nanomedicine*, vol. 8, pp. 1921–1933, 2013, doi: 10.2147/IJN.S44487.
9. H. Haritha P and L. Lakshmi PK, “Probilosomes: A Novel Bile Salt Containing Nanocarrier for Enhancing Oral Bioavailability,” *Int J Pharm Investig*, vol. 10, no. 1, pp. 49–53, 2020, doi: 10.5530/ijpi.2020.1.9.
10. Y. S. R. Elnaggar, “Multifaceted applications of bile salts in pharmacy: An emphasis on nanomedicine,” *Int J Nanomedicine*, vol. 10, pp. 3955–3971, 2015, doi: 10.2147/IJN.S82558.
11. Zafar *et al.*, “Bioactive Apigenin loaded oral nano bilosomes: Formulation optimization to preclinical assessment,” *Saudi Pharmaceutical Journal*, vol. 29, no. 3, pp. 269–279, 2021, doi: 10.1016/j.jsps.2021.02.003.

12. Faustino, C. Serafim, P. Rijo, and C. P. Reis, "Bile acids and bile acid derivatives: use in drug delivery systems and as therapeutic agents," *Expert Opin Drug Deliv*, vol. 13, no. 8, pp. 1133–1148, 2016, doi: 10.1080/17425247.2016.1178233.
13. E. Aziz, A. A. Abdelbary, and A. I. Elassasy, "Investigating superiority of novel bilosomes over niosomes in the transdermal delivery of diacerein: in vitro characterization, ex vivo permeation and in vivo skin deposition study," *J Liposome Res*, vol. 29, no. 1, pp. 73–85, 2019, doi: 10.1080/08982104.2018.1430831.
14. M. Joseph Naguib, A. Moustafa Kamel, A. Thabet Negmeldin, A. H. Elshafeey, and I. Elsayed, "Molecular docking and statistical optimization of taurocholate-stabilized galactose anchored bilosomes for the enhancement of sofosbuvir absorption and hepatic relative targeting efficiency," *Drug Deliv*, vol. 27, no. 1, pp. 996–1009, 2020, doi: 10.1080/10717544.2020.1787557.
15. Y. S. R. Elnaggar, S. Omran, H. A. Hazzah, and O. Y. Abdallah, "Anionic versus cationic bilosomes as oral nanocarriers for enhanced delivery of the hydrophilic drug risedronate," *Int J Pharm*, vol. 564, pp. 410–425, 2019, doi: 10.1016/j.ijpharm.2019.04.069.
16. P. Palekar-Shanbhag, S. Lande, R. Chandra, and D. Rane, "Bilosomes: Superior Vesicular Carriers," *Curr Drug ther*, vol. 15, no. 4, pp. 312–320, 2019, doi: 10.2174/1574885514666190917145510.
17. A. Matloub, A. H. Salama, H. A. Aglan, M. M. AbouSamra, S. S. M. ElSouda, and H. H. Ahmed, "Exploiting bilosomes for delivering bioactive polysaccharide isolated from Enteromorpha intestinalis for hacking hepatocellular carcinoma," *Drug Dev Ind Pharm*, vol. 44, no. 4, pp. 523–534, 2018, doi: 10.1080/03639045.2017.1402922.
18. P. Parashar, P. Rana, M. Dwivedi, and S. A. Saraf, "Dextrose modified bilosomes for peroral delivery: improved therapeutic potential and stability of silymarin in diethylnitrosamine-induced hepatic carcinoma in rats," *J Liposome Res*, vol. 29, no. 3, pp. 251–263, 2019, doi: 10.1080/08982104.2018.1551408.
19. N. Pavlović *et al.*, "Bile acids and their derivatives as potential modifiers of drug release and pharmacokinetic profiles," *Front Pharmacol*, vol. 9, no. NOV, 2018, doi: 10.3389/fphar.2018.01283.
20. J. Li *et al.*, "A review on phospholipids and their main applications in drug delivery systems," *Asian J Pharm Sci*, vol. 10, no. 2, pp. 81–98, 2015, doi: 10.1016/j.ajps.2014.09.004.
21. J. Ahmad *et al.*, "Bile Salt Stabilized Vesicles (Bilosomes): A Novel Nano-Pharmaceutical Design for Oral Delivery of Proteins and Peptides," *Curr Pharm Des*, vol. 23, no. 11, pp. 1575–1588, 2017, doi: 10.2174/1381612823666170124111142.
22. Samad, Y. Sultana, and M. Aqil, "Liposomal Drug Delivery Systems: An Update Review," *Curr Drug Deliv*, vol. 4, no. 4, pp. 297–305, 2007, doi: 10.2174/156720107782151269.
23. N. Islam *et al.*, "Improvement of solubility and dissolution of ebastine by fabricating phosphatidylcholine/ bile salt bilosomes," *Pak J Pharm Sci*, vol. 33, no. 5, pp. 2301–2306, 2020, doi: 10.36721/PJPS.2020.33.5.SUP.2301-2306.1.
24. G. P. Kumar and P. Rajeshwarrao, "Nonionic surfactant vesicular systems for effective drug delivery—an overview," *Acta Pharm Sin B*, vol. 1, no. 4, pp. 208–219, 2011, doi: 10.1016/j.apsb.2011.09.002.

25. J. Jiao, "Polyoxyethylated nonionic surfactants and their applications in topical ocular drug delivery," *Adv Drug Deliv Rev*, vol. 60, no. 15, pp. 1663–1673, 2008, doi: 10.1016/j.addr.2008.09.002.
26. Z. S. Rizwanullah, M. Rizwanullah, S. R. Mir, and S. Amin, "Bilosomes nanocarriers for improved oral bioavailability of acyclovir: A complete characterization through in vitro, ex-vivo and in vivo assessment," *J Drug Deliv Sci Technol*, vol. 57, 2020, doi: 10.1016/j.jddst.2020.101634.
27. S. Mosallam, N. M. Sheta, A. H. Elshafeey, and A. A. Abdelbary, "Fabrication of Highly Deformable Bilosomes for Enhancing the Topical Delivery of Terconazole: In Vitro Characterization, Microbiological Evaluation, and In Vivo Skin Deposition Study," *AAPS PharmSciTech*, vol. 22, no. 2, 2021, doi: 10.1208/s12249-021-01924-z.
28. Shukla, B. Singh, and O. P. Katare, "Significant systemic and mucosal immune response induced on oral delivery of diphtheria toxoid using nano-bilosomes," *Br J Pharmacol*, vol. 164, no. 2 B, pp. 820–827, 2011, doi: 10.1111/j.1476-5381.2011.01452.x.
29. M. H. Aburahma, "Bile salts-containing vesicles: promising pharmaceutical carriers for oral delivery of poorly water-soluble drugs and peptide/protein-based therapeutics or vaccines," *Drug Deliv*, vol. 23, no. 6, pp. 1847–1867, 2016, doi: 10.3109/10717544.2014.976892.
30. D. Arora, B. Khurana, M. S. Kumar, and S. P. Vyas, "Oral immunization against hepatitis B virus using mannosylated bilesomes," *International Journal of Recent Advances in Pharmaceutical Research*, vol. 1, no. January, pp. 45–51, 2011.
31. H. F. Salem, M. M. Nafady, A. A. Ali, N. M. Khalil, and A. A. Elsisi, "Evaluation of Metformin Hydrochloride Tailoring Bilosomes as an Effective Transdermal Nanocarrier," *Int J Nanomedicine*, vol. 17, pp. 1185–1201, 2022, doi: 10.2147/IJN.S345505.
32. D. A. Seleci, V. Maurer, F. Stahl, T. Scheper, and G. Garnweitner, "Rapid microfluidic preparation of niosomes for targeted drug delivery," *Int J Mol Sci*, vol. 20, no. 19, 2019, doi: 10.3390/ijms20194696.
33. M. A. Obeid, A. Elburi, L. C. Young, A. B. Mullen, R. J. Tate, and V. A. Ferro, "Formulation of Nonionic Surfactant Vesicles (NISV) Prepared by Microfluidics for Therapeutic Delivery of siRNA into Cancer Cells," *Mol Pharm*, vol. 14, no. 7, pp. 2450–2458, 2017, doi: 10.1021/acs.molpharmaceut.7b00352.
34. M. A. Obeid, I. Khadra, A. B. Mullen, R. J. Tate, and V. A. Ferro, "The effects of hydration media on the characteristics of non-ionic surfactant vesicles (NISV) prepared by microfluidics," *Int J Pharm*, vol. 516, no. 1–2, pp. 52–60, 2017, doi: 10.1016/j.ijpharm.2016.11.015.
35. R. V. D'Elia *et al.*, "Exploitation of the bilosome platform technology to formulate antibiotics and enhance efficacy of melioidosis treatments," *Journal of Controlled Release*, vol. 298, pp. 202–212, 2019, doi: 10.1016/j.jconrel.2019.02.002.
36. Ismail, M. Teiama, B. Magdy, and W. Sakran, "Development of a Novel Bilosomal System for Improved Oral Bioavailability of Sertraline Hydrochloride: Formulation Design, In Vitro Characterization, and Ex Vivo and In Vivo Studies," *AAPS PharmSciTech*, vol. 23, no. 6, 2022, doi: 10.1208/s12249-022-02339-0.
37. J. F. S. Mann *et al.*, "Oral delivery of tetanus toxoid using vesicles containing bile salts (bilosomes) induces significant systemic and mucosal immunity," *Methods*, vol. 38, no. 2, pp. 90–95, 2006, doi: 10.1016/j.ymeth.2005.11.002.

38. M. Conacher, J. Alexander, and J. M. Brewer, "Oral immunisation with peptide and protein antigens by formulation in lipid vesicles incorporating bile salts (bilosomes)," *Vaccine*, vol. 19, no. 20–22, pp. 2965–2974, 2001, doi: 10.1016/S0264-410X(00)00537-5.
39. Y. Chen *et al.*, "Enhanced bioavailability of the poorly water-soluble drug fenofibrate by using liposomes containing a bile salt," *Int J Pharm*, vol. 376, no. 1–2, pp. 153–160, 2009, doi: 10.1016/j.ijpharm.2009.04.022.
40. M. Teaima, R. Abdelmonem, Y. A. Adel, M. A. El-Nabarawi, and T. M. El-Nawawy, "Transdermal delivery of telmisartan: Formulation, in vitro, ex vivo, iontophoretic permeation enhancement and comparative pharmacokinetic study in rats," *Drug Des Devel Ther*, vol. 15, pp. 4603–4614, 2021, doi: 10.2147/DDDT.S327860.
41. F. Uchegbu and S. P. Vyas, "'Non–ionic surfactant–based vesicles (niosomes) in drug delivery',".
42. M. Niu *et al.*, "Enhanced oral absorption of insulin-loaded liposomes containing bile salts: A mechanistic study," *Int J Pharm*, vol. 460, no. 1–2, pp. 119–130, 2014, doi: 10.1016/j.ijpharm.2013.11.028.
43. P. Guan *et al.*, "Enhanced oral bioavailability of cyclosporine A by liposomes containing a bile salt.," *Int J Nanomedicine*, vol. 6, pp. 965–974, 2011, doi: 10.2147/ijn.s19259.
44. M. Al-Mahallawi, A. A. Abdelbary, and M. H. Aburahma, "Investigating the potential of employing bilosomes as a novel vesicular carrier for transdermal delivery of tenoxicam," *Int J Pharm*, vol. 485, no. 1–2, pp. 329–340, 2015, doi: 10.1016/j.ijpharm.2015.03.033.
45. R. H. Müller, C. Jacobs, and O. Kayser, "Nanosuspensions as particulate drug formulations in therapy: Rationale for development and what we can expect for the future," *Adv Drug Deliv Rev*, vol. 47, no. 1, pp. 3–19, 2001, doi: 10.1016/S0169-409X(00)00118-6.
46. S. Modi and B. D. Anderson, "Determination of drug release kinetics from nanoparticles: Overcoming pitfalls of the dynamic dialysis method," *Mol Pharm*, vol. 10, no. 8, pp. 3076–3089, 2013, doi: 10.1021/mp400154.