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Formulation and evaluation of Azithromycin Dihydrate Niosomes for the Effective Treatment of Bacterial Infection

Neha Srivastava* [1], Seema Thakur [2], Kamaljeet Kaur[2]

[1]School of Pharmaceutical Sciences, RIMT University, Mandi Gobindgarh, Punjab.
[2]PCTE Group of Institutes, Faculty of Pharmaceutical Sciences, Baddowal Cantt, Ludhiana, Punjab

Corresponding Author: nehasrvstva@gmail.com

Abstract

Azithromycin dihydrate is a semi-synthetic macrolide antibiotic of the azalide class for the treatment of bacterial infection. Niosomes are microscopic lamellar structures of non-ionic surfactant bilayer vesicular structures that allow the medications to be encapsulated within, allowing for regulated delivery of the drugs over an extended period. The niosomes were designed which serve the combined advantages of loading the suitable amount of drug required for treatment, modulating its release at the target site, and controlling the bacterial resistance. It also helps to reduce the dose and frequency of the drugs leads to reduction in side effects. In the present work, niosomes of Azithromycin dihydrate were prepared and evaluated for the treatment of bacterial infection. Different formulations of niosomes were designed and prepared by hand shaking method using different ratios of cholesterol, non-ionic surfactant, sodium deoxycholate, chloroform, and methanol. The prepared niosomes were evaluated for different parameters such as entrapment efficiency, vesicle size and shape, and *in vitro* release study. The drug-excipient interaction was evaluated with the help of FTIR spectra and all the peaks in physical mixtures as well. The efficiency of entrapment was found to be highest for F6 (92.4%). The particle size of the optimized formulation was shown 226.2 nm. SEM analysis indicated that the niosomes are spherical and the size of the niosomes vesicle was in range. *In vitro* drug release studies of the optimized formulation F6 showed maximum drug release of 88.4% after 24 h. The release profiles were subjected to different mathematical models and the best-suited model was found to be the Korsemeyer Peppas model with the highest regression value (0.9225) via the fickian diffusion mechanism.

Keywords Azithromycin, niosomes, vesicular system, drug release kinetics

Introduction

The vesicular drug delivery system is a novel system designed with multiple concentric lipid layers of amphiphilic surfactants and water. Vesicular delivery systems have proven beneficial over conventional delivery systems due to prolonging systemic circulation, improving bioavailability, surpassing metabolic pathways, and reducing drug-related toxicity (1). This system is also highly beneficial as it can hold hydrophilic as well as lipophilic drugs in the multiple-layer concentric vesicular structure. Along with the several advantages the vesicular delivery system also possesses several disadvantages such as lower drug loading capacity, and leakage of drugs from the vesicles during storage and transportation. The efficacy of vesicles highly depends on the size of the vesicle, type, loading capacity, lamellarity, and construction of the vesicle. The vesicular drug delivery system plays a promising role in the target specificity of the drug, can deliver the drug in a controlled

and sustained fashion, and can be administered through multiple routes. Drug delivery systems including nanovesicles are broadly classified as liposomes, niosomes, proniosomes, ethosomes, polymersomes, phytosomes, aquazomes, and many more (2).

Niosomes are microscopic lamellar structures of non-ionic surfactant vesicles that are formed by combining cholesterol with an alkyl or dialkyl polyglycerol ether class non-ionic surfactant, then hydrating the combination in aqueous medium. These lipid vesicular structures resemble liposomes and can transport both lipophilic and amphiphilic medications (3). Figure 1 represents the structure of niosomes. Niosomes are lamellar particles that range in size from 10 to 1000 nm and belong to the nano-size range (4). Niosomes are made of cholesterol and surfactant, which is followed by lipid film hydration. The main component of niosomes, which gives them greater stability than liposomes, is a non-ionic surfactant. These niosomes are less expensive, less prone to oxidation, and less vulnerable to the material's quality affecting their size and shape (5). Numerous non-ionic surfactant types, such as Brij, Tweens, Spans, polyglycerol alkyl ethers, crown ethers, ester-linked surfactants, glucosyl dialkyl ethers, and polyoxymethylene alkyl ethers, have the potential to generate niosomes (6). These carriers show great promise for topical medication delivery and may enhance therapeutic efficacy while mitigating adverse effects (7).

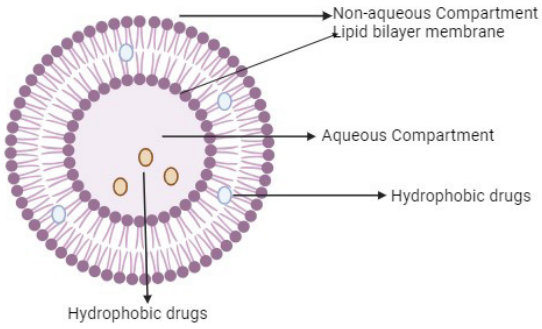

Figure 1: Structure of niosomes

Niosomes are made of bilayer structures that allow the medications to be encapsulated within, allowing for regulated delivery of the drugs over an extended period. This is a new method of medication targeting by surface modification by decreasing the amount that must be given to produce the intended result (8). By encapsulating the medication, lowering its rate of clearance, and directing it to a particular location, the therapeutic efficacy of the medications is increased. Medication targeting aids in drug localization and generates therapeutic benefits at low dose concentrations with fewer adverse effects (9). These could serve as the reservoirs, which release the drug in a controlled manner targeting the affected organs or cells (10).

Although niosomes are less stable physically, they show good chemical stability. The stability of niosomes is caused by a variety of forces, such as van der Waals forces between surfactant molecules, entropic repulsive forces of the head groups of surfactants, short-acting repulsive forces, etc. These are the forces that support the preservation of the noisome's vesicular structure (11).

Azithromycin is one member of the azalide class of semi-synthetic macrolides. By attaching itself to the 50S ribosomal subunit of the bacterial 70S ribosome, it prevents the synthesis of proteins by bacteria. Depending on the organism, it may have bacteriostatic or bactericidal effects (12). Azithromycin undergoes phagocytotic uptake by phagocytic cells and fibroblasts which take the drug and deliver it to the site of infection. On arrival at the site of infection, azithromycin is released to the surface of the cell membrane of the bacteria cell where it binds with ribosomes. The binding of the drug with ribosomes leads to cell death or inhibits the growth of bacteria (13). The current study aims to develop niosomes loaded with the required amount of Azithromycin for the treatment of bacterial infection. The niosomes were designed which serve the combined advantages of loading the suitable amount of drug required for treatment and modulating its release at the

target site. It also improves the penetrability of the drug and helps to deliver the drug to the target site. The controlled delivery system also presents an additional advantage toward bacterial resistance as the dose of the drug in the system remains controlled which reduces the bacterial resistance.

Material and Method

The drug Azithromycin was a gift sample from Biomedica International, Ludhiana. Sodium deoxycholate was obtained from Molychem., Badlapur, Dist., Thane. Cholesterol was obtained from Avarice Laboratories Pvt. Ltd., Ghaziabad. Sorbitan ester 20, Sorbitan ester 40, Sorbitan ester 60, and Sorbitan ester 80 were obtained from Central Drug House (P) Ltd., New Delhi. All the other reagents and solvents were of AR grade.

Methods

Drug excipient interaction

Drug-excipient interaction was done by using FTIR spectrophotometry. KBr pellet technique was used for FTIR studies. The spectrum for the drug Azithromycin in pure form and its physical mixture with other excipients were obtained and studied for any interactions between the drug and excipients (14).

Preparation of Azithromycin dehydrate niosomes

The niosomes of Azithromycin dihydrate with different excipients in different ratios were prepared by the handshaking method. 10 mL of organic solvent (2:1 ratio of methanol and chloroform) was taken in the round bottom flask. To this cholesterol, non-ionic surfactants and drugs were dissolved.

Different ingredients used in the formation of niosomes such as surfactant, cholesterol, and drug are dissolved in 10 ml of organic solvents (2:1 ratio of methanol and chloroform) in a round bottom flask. The organic solvent was allowed to evaporate at room temperature by rotating the flask, which led to the deposition of the thin film on the wall of the flask. The dried film of surfactant was reconstituted with 10 ml of Phosphate buffer pH 7.4 for 1 hour at 60 °C with gentle agitation. The prepared noisomes were kept overnight at 4 °C and then stored in a refrigerator for further use (15, 16). Formulations were prepared using changing ratios of cholesterol, surfactant, and drug are listed in Table 1.

Table 1: Compositions (W/W) Of Niosomes of Azithromycin Dehydrate

Formulation code	Ratios	Span 60	Span 80	Span 20	Cholesterol	Azithromycin dihydrate	SDC
F1	1:1	1	-	-	1	-	-
F2	1:1:1	1	-	-	1	1	1
F3	2:1:1	2	-	-	1	-	1
F4	2:1	2	-	-	1	1	-
F5	1:1:1	-	1	-	1	1	1
F6	1:1:1	1	-	-	1	1	1
F7	1:1:0.5	1	-	-	0.5	1	1
F8	1:1:1	-	-	1	1	1	1

Evaluation of Azithromycin dehydrate niosomes

Particle size analysis and Zeta potential analysis

The size of the particle of the prepared niosomes was determined by Malvern Zeta sizer (Zeta sizer 3, Malvern, UK) at 25 °C using a dynamic light scattering technique (17, 18). The sample of particle size was prepared in demineralized water. The niosome's stability depends upon the zeta potential of niosomal preparation. For nanosized molecules, the zeta potential indicates the strength of repulsion between neighboring particles having similar charges. A higher value of zeta potential indicates stability, or the capacity of the solution or dispersion to withstand aggregation. Zeta potential was determined using a Malvern zeta sizer.

Surface morphology

The surface morphology of the prepared niosomes was studied using a scanning electron microscope (SEM) (JSM 6100, Jeol, Japan), and the noisome were examined. The sample was directly mounted onto the SEM sample stub using the stick tape and coated with a 200 nm thick gold layer at a lowered pressure of 0.001 mmHg. An appropriate magnification was used to take photographs (19, 20).

Entrapment efficiency

The difference between the total amount of drug and the unentrapped amounts was used to calculate the entrapment efficiency. Each formulation was centrifuged using a Remi centrifuge (REMI LJ01*, Mumbai)* for 30 minutes at room temperature (25°C) at 5000 rpm to separate the drug in supernatant liquid which was entrapped in the niosomal structure. The sample was analyzed using a UV spectrophotometer (Shimadzu, Japan), and the amount of drug was determined at 214 nm. % entrapment efficiency was calculated (21). The below mention equation was used to study the entrapment efficiency:

Entrapment efficiency = (Amount entrapped / total amount) x 100

In-vitro drug release study

In vitro, the release of Azithromycin dihydrate from the niosomal structure was carried out using a slightly modified version of Hu's technique. Drug release experiments were conducted on each formulation. The glass cylinder used to construct the diffusion cell was hollow and made of Borosil glass, measuring 14.5 cm in length and 2.50 cm in diameter on inner wall of the tube. Himedia dialysis membrane (molecular weight cut-off: 12000–14000), which had been soaked in warm water beforehand, was placed over one end of the cylinder. The 250 ml beaker that was used as the receptor compartment held the manufactured diffusion cell. The magnetic bead inside the receptor cell rotated at a steady speed. With the use of a thermostat, the temperature inside the manufactured diffusion cell and receptor cells was kept at 37°C. The receiving cell was filled with phosphate buffer saline (100 ml) with a pH of 7.4. Each formulation's 1 ml sample was added to the dissolution cell. The receiving compartment was sampled (5 mL) at predetermined intervals of 1, 2, 3, 5, 6, 7, 8, 9, 10, 11, 12, 16, and 24 hours—every time the sample was replaced with phosphate buffer saline (pH 7.4). The Azithromycin dihydrate content of the samples was determined using a UV spectrophotometer (Shimadzu UV-160, Japan) set to 214 nm as the λmax (22, 23). The temperature of the system was maintained at 37ºC.

Drug release kinetics

The selected formulations were subjected to kinetics equations for drug release such as first-order kinetics, zero-order kinetics, Korsymeyer-Peppas model, Higuchi model, and Hixon Crowell model. The best-fitted model is selected based on regression value, the model with the highest regression is the governing release pattern for the prepared formulations. The drug release kinetics indicated the release

mechanism pattern from the niosomes. The equation of the different models is as follows:

$$\text{Zero order model}: \ M_t = kt + M_0$$

$$\text{First order model}: \ M_t = M_0^{e^{kt}}$$

$$\text{Higuchi model}: \ \frac{M_t}{M_\infty} = kt^{1/2}$$

$$\text{Korsmeyer} - \text{Peppas model}: \ \frac{M_t}{M_\infty} = kt^n$$

Where M_t stands for the drug amount released from the conjugated system at time t, and M_0 is the total amount of the drug in the conjugated system. M_t/M_∞ is the fraction of the released drug, n is the diffusion exponent and k is the rate constant.

The value of n in the Korsmeyer Peppas model indicated the release mechanism from the formulated structures. If the value ranges between 0.45 to 0.89, indicating the release mechanism. Suppose the value is less than 0.45 the release was considered to follow Fick's law and termed as fickian diffusion from a non-swellable matrix type. If the value of n is between 0.45 to 0.89, the release is non-fickian and considered to be released both by diffusion and erosion. If the value of n is more than 0.89, it is case II transport and if the value is more than 1 it is super case II transport (24).

Result and Discussion

Different formulations were prepared using Azithromycin dehydrate niosomes for ocular delivery along with other ingredients. Azithromycin is a macrolide antibiotic that is semi-synthetic and efficient against a broad range of bacteria. Its main application is the treatment of bacterial illnesses linked to weakened immune systems. It was observed that Azithromycin dehydrate has poor solubility and low bioavailability which makes it a suitable candidate for drug delivery (19). Noisomes are prepared using non-ionic surfactants and cholesterol. Typically, cholesterol is added to non-ionic surfactants to give the niosomal bilayer stiffness and orientational order. It increases stability, lessens aggregation, and permits vesicle formation. It is also known that cholesterol prevents the niosomal systems' gel-to-liquid phase transition, making the niosomes less prone to leakage (25). Span 60 is a non-ionic surfactant. It has a higher alkyl chain length and phase transition temperature. Span 60 has shown higher entrapment efficiency due to their higher alkyl chain length (26). Span 80 has an unsaturated alkyl chain and a lower phase transition temperature. Span 80 has shown lower entrapment efficiency due to the unsaturated alkyl chain in their structure. Sodium deoxycholate is a negative charge inducer (27). It was observed that formulations F1, F2, F3, and F4 vesicle were not formed due to lower content of cholesterol and surfactants in the formulations while from F5 to F8 vesicles were formed due to high concentration. The results of different formulations are tabulated in Table 2.

Drug excipient interaction

FTIR spectra of pure Azithromycin dihydrate and the polymers individually as well as the combination of the drug with all the other polymers are shown in Figures 2 and 3 and tabulated in Tables 3 and 4. The spectra indicated the presence of prominent peaks at 1720 cm^{-1} corresponding to carboxylic acid; another peat of C-O stretching was present at 1378.19 cm^{-1}. The stretching of the OH group is observed at 1188.48 cm^{-1}. The physical mixture exhibits similar peaks at 1720.77 for carboxylic acid, 1378.36 cm^{-1} for C-O stretching, and 1188.48 cm^{-1} for OH stretching. No significant changes in intensity of the FTIR bands of Azithromycin dihydrate were observed with polymers indicating the absence of interaction. The peak wavelength is shown in Table 2**.** Figures 2 and 3 represent the FTIR spectrum of the pure drug azithromycin dihydrate and the physical mixture of azithromycin dihydrate-excipient

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Table 2: FTIR peak frequency and their corresponding functional groups of physical Mixture of drug and excipients

S . No	(KBr disc) peaks at	Indications
1.	1720.77	C=O group stretching is present (1500 - 2000)
2.	1378.36	C-O stretching is present (1400 - 1310)
3.	1188.86	O-H group stretching is present (1100-1200)
4.	955.73	C-H group bending is present (700-950)

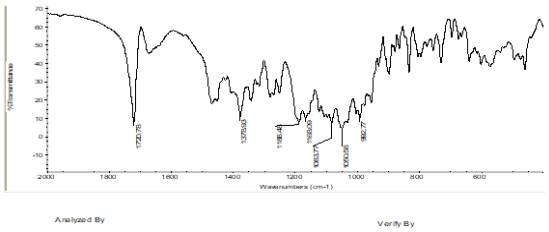


Figure 2: FTIR spectrum of pure drug azithromycin dihydrate

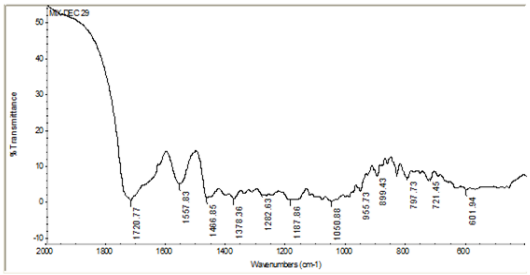


Figure 3: FTIR spectra of the physical mixture of azithromycin dihydrate-excipient

Entrapment efficiency

The drug content analysis of different formulations was done according to the procedure given in the methodology section. The results were tabulated in Table 3. It was observed that the entrapment efficiency of the vesicle of span 60 was found to be higher due to higher alkyl chain length. Very low cholesterol content in formulation also causes low entrapment efficiency which might lead to leakage of the vesicle. The entrapment efficiency was enhanced by increasing the cholesterol along with span 60 due to higher phase transition temperature (28).

The highest entrapment efficiency for the formulation F6 was found to be 92.4% and F5 was found to be 88.2%. The highest entrapment was due to the optimum concentration of span 60 and cholesterol. This leads to higher entrapment of the drug Azithromycin dihydrate.

The head groups of Span 60 and Span 80 are similar, the difference is in the chain length and branching of the chain. The Alkyl chain of span 80 is unsaturated. The permeability of liposomes is markedly enhanced upon the introduction of double bonds into paraffin chains, which may account for the Span 80 systems' lower entrapment efficiency (29). The entrapment efficiency was lower in the formulations F7 (81.8%) and F8 (79.3%) due to the unsaturated alkyl chain length of their surfactant and lower transition temperature.

Table 3: Entrapment efficiency of F5-F8 formulations

Sr. No.	Formulation Code	Entrapment Efficiency (%)
1	F5	88.2±0.56
2	F6	92.4±0.12
3	F7	81.8±0.43
4	F8	79.3±0.14

Particle size and polydispersibility

By using the Malvern Zetasizer (Zets sizer, 3, Malvern, UK) at 25 °C based on dynamic light scattering, the noisome size distribution was determined. Niosome particle size is depicted in Figures 4 and 5. When a drug is encapsulated in nonionic surfactant vesicles, the particle size typically rises. This is likely because of the interaction of the drug with the surfactant head groups. This leads to a rise in the charges and results in mutual repulsion of the bilayer of surfactant (30).

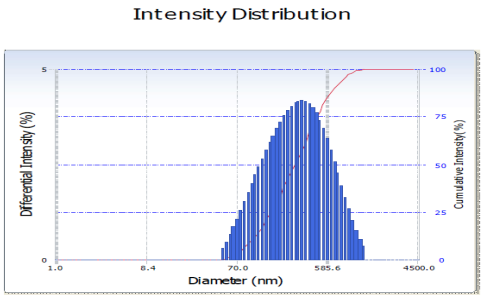


Figure 4: Particle Size of Niosomes

Cumulants Results			
Diameter	(d)	: 226.2	(nm)
Polydispersity Index	(P.I.)	: 0.304	
Diffusion Const.	(D)	: 2.175e-008	(cm²/sec)
Measurement Condition			
Temperature		: 25.0	(°C)
Diluent Name		: Buffer	
Refractive Index		: 1.3318	
Viscosity		: 1.0788	(cP)
Scattering Intensity		: 9840	(cps)

Figure 5: Polydispersity index of Niosomes

The polydispersity index was calculated as a polydispersity index of 1 indicating that globule size is varying on the larger scale. The observed value of 0.3 indicated that size variation is narrow and acceptable. Figure 5 shows the polydispersity index of niosomes.

Scanning electron microscopy

SEM images shown in Figure 6 confirm the preparation of niosomes. By field emission SEM image of the best formulation confirms that there is no aggregation the photograph reveals a smooth surface and the average length of the structure was in the nm range (31). Figure 6 represents the SEM images of prepared niosomes.

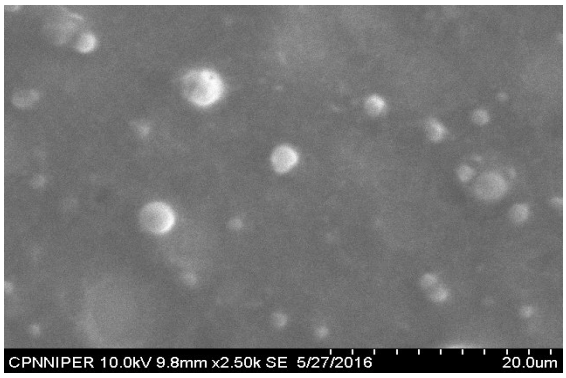


Figure 6: SEM image of prepared niosomes

The *in vitro* permeation of Azithromycin dihydrate was carried out by cellophane membrane. Vesicle suspensions were placed over the membrane. The donor compartment was clamped over it with the help of springs. Samples of 3 ml were withdrawn at predetermined (1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 16, 20 and 24 hrs) time intervals from the receptor compartment. Graphs plotted between the percent cumulative drug releases versus time are shown in Figure 7**.** It was observed that a 1:1:1 ratio of Span: Cholesterol: Azithromycin shows maximum drug release and is considered as optimum formulation (32). Figure 7 represents the in vitro drug release profile of different formulations.

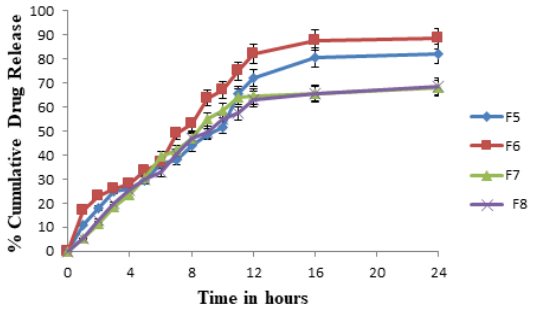


Figure 7: *In vitro* release profile of niosomes of Azithromycin dehydrate

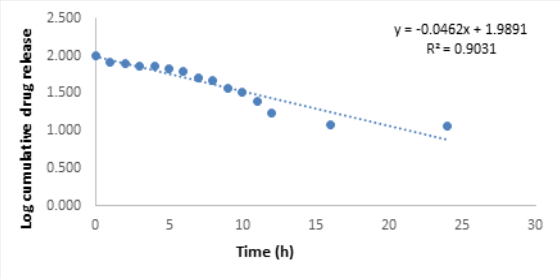
Drug release kinetics

The drug release profile of Azithromycin dehydrates was subjected to release kinetics such as first order, zero order, Korsemeyer Peppas, Higuchi, and Hixon-Crowell methods. The best-fitted model was selected based on the R^2 value of the different models. The Regression value of different formulations using different release kinetic models is tabulated in Table 4.

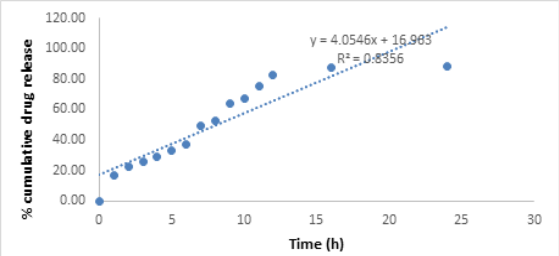
Table 4: Drug release kinetics of formulation F5-F8

S.No	Formulation Code	Release kinetics					
		First-order	Zero-order	Korsmey-er-Peppas	n value	Higuchi	Hixon
1	F5	0.9201`	0.8798	0.8841	0.96821	0.9201	0.9154
2	F6	0.9031	0.8356	0.8538	0.9462	0.9225	0.891
3	F7	0.9197	0.7668	0.8247	0.8248	0.9032	0.8075
4	F8	0.9218	0.8088	0.8573	0.8256	0.9183	0.8455

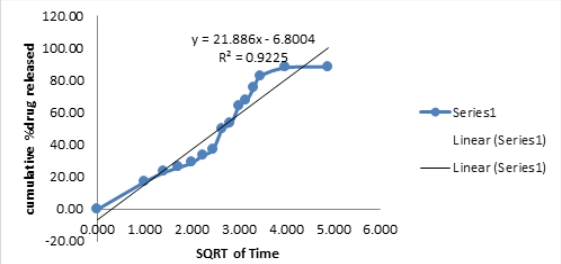
All the formulations are subjected to different release kinetics models and found that all the formulation follows the Korsmeyer-Peppas model. The best-fitted model was selected as the R^2 was found to be the highest for Korsmeyer-Peppas. F6 is the optimized formulation that follows the Korsmeyer-Peppas model with the highest R^2 value (0.9225). Korsmeyer-Peppas model governs the drug release through the polymeric matrix system. This model helps to understand the release mechanism through the diffusion of water into the matrix, swelling of the matrix, and dissolution of the matrix. n value was found to be 0.0462 indicating that the release is through fickian diffusion. Figure 8 represents (a) First order kinetics of optimized formulation F6 (b) Zero order kinetics of optimized formulation F6 (c) Higuchi release kinetics of optimized formulation F6 (d) Korsmeyer-Peppas kinetics of optimized formulation F6 (e) Hixon Crowell kinetics of optimized formulation



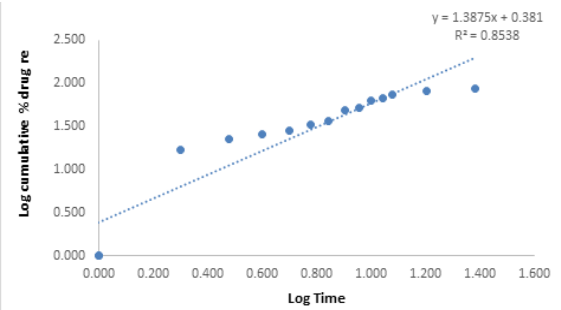
(a) First-order release kinetics of optimized formulations F6



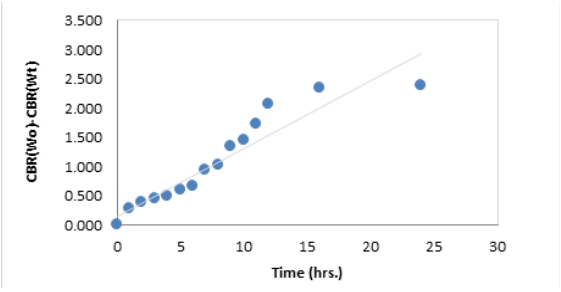
(b) Zero order release kinetics of optimized formulations F6



(c) Higuchi release kinetics of optimized formulations F6



(d) Korsmeyer-Peppas release kinetics of optimized formulations F6



(e) Hixon-Crowel release kinetics of optimized formulations F6

Figure 8: (a) First order kinetics of optimized formulation F6 (b) Zero order kinetics of optimized formulation F6 (c) Higuchi release kinetics of optimized formulation F6 (d) Korsmeyer-Peppas kinetics of optimized formulation F6 (e) Hixon Crowell kinetics of optimized formulation

Conclusion

The study finding reveals that the prepared niosomes help to prolong the release of the drug for 24 hr. The formulation with a 1:1:1 ratio of span 60:cholesterol: drug showed good results with 92.4% entrapment efficiency and 88.4% drug release. The SEM results show the spherical and smooth vesicles without aggregations. From the study, it can be concluded that vesicle delivery can be used to improve the rate of release of drugs as well as improve bioavailability.

Release Kinetics

Conflict of interest

No conflict of interest.

References

1. Myneni, G. S., Radha, G., & Soujanya, G. V. R. L. (2021). Novel vesicular drug delivery systems: a review. Journal of Pharmaceutical Research, 11, 1650-1664.
2. Chacko, I. A., Ghate, V. M., Dsouza, L., & Lewis, S. A. (2020). Lipid vesicles: A versatile drug delivery platform for dermal and transdermal applications. Colloids and Surfaces B: Biointerfaces, 195, 111262.
3. Jain, A. K., & Mishra, K. (Eds.). (2022). Nanoparticles and Nanocarriers Based Pharmaceutical Formulations. Bentham Science Publishers, pp.333.
4. Mishra, D. K., Shandilya, R., & Mishra, P. K. (2018). Lipid based nanocarriers: a translational perspective. Nanomedicine: Nanotechnology, Biology and Medicine, 14(7), 2023-2050.
5. Chen, S., Hanning, S., Falconer, J., Locke, M., & Wen, J. (2019). Recent advances in non-ionic surfactant vesicles (niosomes): Fabrication, characterization, pharmaceutical and cosmetic applications. European journal of pharmaceutics and biopharmaceutics, 144, 18-39.
6. Rashwan, A. K., Karim, N., Xu, Y., Xie, J., Cui, H., Mozafari, M. R., & Chen, W. (2023). Potential micro-/nano-encapsulation systems for improving stability and bioavailability of anthocyanins: An updated review. Critical Reviews in Food Science and Nutrition, 63(19), 3362-3385.
7. Biswasroy, P., Pradhan, D., Kar, B., Ghosh, G., & Rath, G. (2021). Recent advancement in topical nanocarriers for the treatment of psoriasis. AAPS Pharmscitech, 22(5), 164.
8. Gharbavi, M., Amani, J., Kheiri-Manjili, H., Danafar, H., & Sharafi, A. (2018). Niosome: a promising nanocarrier for natural drug delivery through blood-brain barrier. Advances in Pharmacological and Pharmaceutical Sciences, 2018.
9. Chenthamara, D., Subramaniam, S., Ramakrishnan, S. G., Krishnaswamy, S., Essa, M. M., Lin, F. H., & Qoronfleh, M. W. (2019). Therapeutic efficacy of nanoparticles and routes of administration. Biomaterials Research, 23(1), 1-29.
10. Mehrarya, M., Gharehchelou, B., Haghighi Poodeh, S., Jamshidifar, E., Karimifard, S., Farasati Far, B., ... & Seifalian, A. (2022). Niosomal formulation for antibac-

terial applications. Journal of Drug Targeting, 30(5), 476-493.

11. Kauslya, A., Borawake, P. D., Shinde, J. V., & Chavan, R. S. (2021). Niosomes: a novel carrier drug delivery system. Journal of Drug Delivery and Therapeutics, 11(1), 162-170.
12. Zitouni, K. (2021). Structural exploration and study of the QSAR properties of several series of macrolide antibiotics (Doctoral dissertation, Université de mohamed kheider biskra).
13. Vázquez-Laslop, N., & Mankin, A. S. (2018). How macrolide antibiotics work. Trends in biochemical sciences, 43(9), 668-684.
14. Bhattacharyya, S., Pasha, I., Verma, A., Kothapalli, R., Jafar, F., & Hr, K. (2019). Formulation and evaluation of liquisolid compact of azithromycin dihydrate. Journal of Research in Pharmacy, 23(6), 1022-32.
15. Aarti, H., Jamindar, D., Mahajan, A., & Hardia, A. (2017). Formulation and In-vitro and Skin Permeability Evaluation of Dexamethasone Loaded Niosomal Gel. Asian Journal of Pharmaceutical Research and Development, 5(2), 01-09.
16. Rao, A., Arvapalli, S., Rao, G. S. N., Malothu, N., & Bandaru, N. R. (2021). Design and evaluation of acyclovir niosomes. Research Journal of Pharmacy and Technology, 14(8), 4185-4188.
17. Allam, A., El-Mokhtar, M. A., & Elsabahy, M. (2019). Vancomycin-loaded niosomes integrated within pH-sensitive in-situ forming gel for treatment of ocular infections while minimizing drug irritation. Journal of Pharmacy and Pharmacology, 71(8), 1209-1221.
18. Ramadan, A. A., Eladawy, S. A., El-Enin, A. S. M. A., & Hussein, Z. M. (2020). Development and investigation of timolol maleate niosomal formulations for the treatment of glaucoma. Journal of Pharmaceutical Investigation, 50, 59-70.
19. Mohanty, D., Rani, M. J., Haque, M. A., Bakshi, V., Jahangir, M. A., Imam, S. S., & Gilani, S. J. (2020). Preparation and evaluation of transdermal naproxen niosomes: formulation optimization to preclinical anti-inflammatory assessment on murine model. Journal of liposome research, 30(4), 377-387.
20. Jacob, S., Nair, A. B., & Al-Dhubiab, B. E. (2017). Preparation and evaluation of niosome gel containing acyclovir for enhanced dermal deposition. Journal of liposome research, 27(4), 283-292.
21. Sailaja, A. K., & Shreya, M. (2018). Preparation and characterization of naproxen loaded niosomes by ether injection method. Nano Biomedicine and Engineering, 10(2), 174-180.
22. Abdelbary, A., Salem, H. F., Khallaf, R. A., & Ali, A. M. (2017). Mucoadhesive niosomal in situ gel for ocular tissue targeting: in vitro and in vivo evaluation of lomefloxacin hydrochloride. Pharmaceutical development and technology, 22(3), 409-417.
23. Gugleva, V., Titeva, S., Ermenlieva, N., Tsibranska, S., Tcholakova, S., Rangelov, S., & Momekova, D. (2020). Development and evaluation of doxycycline niosomal thermoresponsive in situ gel for ophthalmic delivery. International Journal of Pharmaceutics, 591, 120010.
24. Siswanto, A., Fudholi, A., Nugroho, A. K., & Martono, S. (2015). In vitro release modeling of aspirin floating tablets using DDSolver. Indonesian Journal of Pharmacy, 26(2), 94.
25. Shirsand, S. B., & Keshavshetti, G. G. (2019). Recent advances in niosomal drug delivery—A review. Research Journal of Life Sciences, Bioinformatics, Pharmaceutical and Chemical Sciences, 3, 514-531.

26. Bnyan, R., Khan, I., Ehtezazi, T., Saleem, I., Gordon, S., O'Neill, F., & Roberts, M. (2018). Surfactant effects on lipid-based vesicles properties. Journal of pharmaceutical sciences, 107(5), 1237-1246.

27. Nava-Arzaluz, M. G., Piñón-Segundo, E., & Ganem-Rondero, A. (2019). Lipid nanocarriers as skin drug delivery systems. In Nanoparticles in Pharmacotherapy . William Andrew Publishing. pp.311-390

28. Yeo, L. K., Olusanya, T. O., Chaw, C. S., & Elkordy, A. A. (2018). Brief effect of a small hydrophobic drug (cinnarizine) on the physicochemical characterisation of niosomes produced by thin-film hydration and microfluidic methods. Pharmaceutics, 10(4), 185.

29. Akbari, V., Abedi, D., Pardakhty, A., & Sadeghi-Aliabadi, H. (2015). Release studies on ciprofloxacin loaded non-ionic surfactant vesicles. Avicenna journal of medical biotechnology, 7(2), 69.

30. Fouda, N. H., Abdelrehim, R. T., Hegazy, D. A., & Habib, B. A. (2018). Sustained ocular delivery of Dorzolamide-HCl via proniosomal gel formulation: in-vitro characterization, statistical optimization, and in-vivo pharmacodynamic evaluation in rabbits. Drug delivery, 25(1), 1340-1349

31. Sahu, A. K., Mishra, J., & Mishra, A. K. (2020). Introducing Tween-curcumin niosomes: Preparation, characterization and microenvironment study. Soft matter, 16(7), 1779-1791.

32. Barani, M., Hajinezhad, M. R., Sargazi, S., Rahdar, A., Shahraki, S., Lohrasbi-Nejad, A., & Baino, F. (2021). In vitro and in vivo anticancer effect of pH-responsive paclitaxel-loaded niosomes. Journal of Materials Science: Materials in Medicine, 32, 1-13.