Asian Journal of Pharmaceutical Education and Research
Vol -13, Issue-3, July- September 2024
ISSN:2278 7496

RESEARCH ARTICLE Impact Factor: 7.014

# FORMULATION AND CHARACTERIZATION OF TOPICAL GEL OF ACYCLOVIR

**Ankita Namdeo Shende*, C.R. Doijad, Sachin B Dudhe**

**Maharashtra Institute of Pharmacy, Betala, Bramhapuri, Maharashtra**

*Corresponding Author's E mail: snankita1997@gmail.com
Received 15 June. 2024; Revised 19 June. 2024; Accepted 25 June. 2024, Available online 10 July. 2024

Cite this article as: Shende NA, Doijad CR and Dudhe SB. Formulation and Characterization of Topical Gel of Acyclovir. Asian Journal of Pharmaceutical Education and Research. 2024; 13(3): 349-358.

https://dx.doi.org/10.38164/AJPER/13.3.2024.349-358

## ABSTRACT

This study comprehensively characterizes Acyclovir and its formulations, elucidating their physical and chemical properties, solubility behavior, formulation attributes, and drug release kinetics. Acyclovir, a white crystalline powder, exhibits odorlessness and a bitter taste. It shows varying solubility in different solvents, is slightly soluble in water, soluble in hydrochloric acid and sodium hydroxide solutions, and sparingly soluble in ethanol and methanol. It demonstrates a consistent melting point (254-256°C), with FT-IR spectra revealing characteristic peaks corresponding to functional groups. Moisture content is low (0.109%), and linear regression analysis shows a strong relationship between concentration and absorbance. Characterization of Acyclovir-loaded invasomes formulations indicates variability in entrapment efficiency and vesicle size. The optimized invasomes formulation (F5) shows a vesicle size of 210.32 nm, an entrapment efficiency of 75.65%, and a zeta potential of -37.45 mV. Further characterization of invasomes gel formulations (IG-1, IG-2, IG-3) includes viscosity, pH, drug content, extrudability, and spreadability assessments. The in vitro release study of the optimized gel formulation (IG-2) demonstrates sustained release characteristics, with regression analysis supporting various kinetic models. Overall, this study provides valuable insights into Acyclovir's pharmaceutical development, highlighting its potential in treating various conditions.

**Keywords:** Acyclovir, physical properties, chemical properties, solubility, formulation, drug release kinetics, invasomes, gel formulation, pharmaceutical development.

## INTRODUCTION

Herpes simplex virus (HSV) infections, including oral and genital herpes, remain prevalent worldwide, necessitating effective antiviral therapies for symptom management and viral suppression. Acyclovir, a nucleoside analog, is widely used for its potent antiviral activity against HSV by inhibiting viral DNA replication [1]. While oral and intravenous formulations of acyclovir

are available, topical administration offers advantages such as targeted delivery to affected areas, enhanced efficacy, and reduced systemic side effects [2].

In recent years, invasomes have emerged as promising carriers for enhancing the topical delivery of drugs like acyclovir. Invasomes are vesicular systems composed of phospholipids and edge activators (surfactants) that improve skin permeability and drug penetration [3]. These nanovesicles possess the ability to encapsulate hydrophilic and lipophilic drugs, protecting them from enzymatic degradation and facilitating sustained release at the application site.

The formulation of invasomal gels represents a sophisticated approach to optimize drug delivery, combining the benefits of invasomes with the ease of application and patient compliance offered by gel formulations. This formulation strategy aims to overcome skin barriers, enhance drug permeation into deeper skin layers, and provide controlled release kinetics for sustained therapeutic effect.

This study focuses on the formulation and characterization of a topical invasomal gel of acyclovir, aiming to improve its efficacy and therapeutic outcomes in treating HSV infections. By encapsulating acyclovir within invasomes and formulating them into a gel matrix, the study seeks to enhance drug stability, skin permeation, and localized antiviral activity. Characterization will include evaluating vesicle size, encapsulation efficiency, drug release kinetics, and skin permeation studies to assess the formulation's performance and potential clinical application.

Through this research, we aim to contribute to the development of novel topical formulations that maximize the therapeutic benefits of acyclovir for HSV management, addressing current challenges in treatment efficacy and patient adherence.

## MATERIAL AND METHODS

The formulation development of invasomal gel containing acyclovir, several key materials were employed. Acyclovir, sourced from Bioplus Life Science in Bangalore, Soya phosphatidyl choline, obtained from Ash Chemie India in Thane. Other chemicals including disodium hydrogen phosphate, dipotassium hydrogen orthophosphate, and sodium chloride from S. D. Fine Chem Ltd. in Mumbai were utilized for buffer preparation and isotonicity adjustment. Solvents such as methanol, ethanol, and chloroform, supplied by Qualigens Fine Chemicals in Mumbai, facilitated the preparation and purification of invasomes. Carbopol 934p, methyl paraben, propyl paraben, and propylene glycol, also from S. D. Fine Chem Ltd., were employed for gel formation, viscosity adjustment, and preservative properties. Together, these materials were integrated into the

formulation to optimize the delivery and efficacy of acyclovir in treating HSV infections through enhanced topical delivery and sustained release mechanisms.

## Methods

### Formulation and optimization of acyclovir loaded Invasomes

Acyclovir (100mg) was loaded in to invasomes by mechanical dispersion technique. Soya Phosphatidylcholine (0.5 to 1.5% w/v) was added to ethanol and vortexed for 5 minutes [4]. Drug (500mg) and terpenes (Eugenol) (0.25%) were added under constant vortexing, this mixture was sonicated for 5 minutes. Fine stream of Phosphate buffer saline was added with syringe under constant vortexing. It was vortexed for additional 5 minutes to obtain final invasomal preparation.

**Table 1: Formulation optimization of Acyclovir loaded Invasomes**

| Ingredient (%) | F1 | F2 | F3 | F4 | F5 | F6 |
|---|---|---|---|---|---|---|
| Acyclovir (mg) | 500 | 500 | 500 | 500 | 500 | 500 |
| Phosphotidylcholine (%) | 1.0 | 1.5 | 2.0 | 1.0 | 1.5 | 2.0 |
| Terpenes (%) | 0.25 | 0.25 | 0.25 | 0.25 | 0.25 | 0.25 |
| Ethanol (ml) | 10 | 10 | 10 | 10 | 10 | 10 |

### Preparation of gel base

Carbopol 934 (1-3%w/v Invasome based gel formulation i.e. G-1 of 1%w/v, G-2 of 2%w/v, G-3 of 3%w/v) was accurately weighed and dispersed into double distilled water (80ml) in a beaker. This solution was stirred continuously at 800 rpm for 1 hour and then 10ml of propylene glycol was added to this solution [5]. The obtained slightly acidic solution was neutralized by drop wise addition of 0.05 N sodium hydroxide solutions, and again mixing was continued until gel becomes transparent. Volume of gel was adjusted to 100 ml and then sonicated for 10 min on bath sonicator to remove air bubbles. Final pH of the gel base was adjusted to 6.5. The same procedure was used to formulate Invasomes containing gel in which previously prepared Invasomes suspension was added. Invasomes preparation corresponding to 1% w/w of drug was incorporated into the gel base to get the desired concentration of drug in gel base.

**Table 2: Formulation optimization of gel base**

| Ingredient (%) | G-1 | G-2 | G-3 |
|---|---|---|---|
| Drug (Invasomes equivalent to 5%) | 1 | 1 | 1 |
| Carbopol 934 | 1 | 2 | 3 |
| Propylene glycol | 0.2 | 0.2 | 0.2 |
| Water (ml) | 100 | 100 | 100 |

**Evaluation of Invasomes**

**Entrapment efficiency**

Entrapment efficiency of Acyclovir Invasomes formulation was determined using centrifugation method [6]. The entrapment efficiency of acyclovir in invasomes vesicle was determined by ultracentrifugation, 10mL of invasomes formulation were collect in test tube. The amount of drug not entrapped in the invasomes was determined by centrifuging at 3,000 rpm and collect the supernatant, the supernatant layer was separated, diluted with water suitably and drug concentration was determined at 256nm using UV spectrophotometer.

$$\%\ Entrapment\ Efficiency\ = \frac{Theroticaldrugcontent - Practicaldrugcontent}{\text{Therotical drug content}} \times 100$$

**Vesicle Size**

Microscopic analysis was performed to determine the average size of prepared invasomes [7]. Formulation was diluted with distilled water and one drop was taken on a glass slide and covered with cover slip. The prepared slide was examined under trinocular microscopic at 400 X. The diameters of more than 150 vehicles were randomly measured using calibrated ocular and stage micrometer. The average diameter was calculated using the following formula.

$$AverageDiameter = \frac{\Sigma n.d}{\Sigma\ \text{n}}$$

Where n = number of vesicles; d = diameter of the vesicles

**Evaluation of Invasomes containing gel**

**Measurement of viscosity**

Viscosity measurements of prepared topical Invasomes based gel were measured by Brookfield viscometer using spindle no. 63 with the optimum speed of 10rpm.

**pH measurements**

pH of selected optimized formulations was determined with the help of digital pH meter. Before each measurement of pH, pH meter should be calibrated with the help of buffer solution of pH 4, pH 7 and pH 9.2. After calibration, the electrode was dipped into the vesicles as long as covered by the vesicles. Then pH of selected formulation was measured and readings shown on display were noted [8].

**Drug content**

Accurately weighed equivalent to 100 mg of topical Invasomes gel was taken in beaker and added 20 ml of methanol. This solution was mixed thoroughly and filtered using Whatman filter paper no.1. Then 1.0 mL of filtered solution was taken in 10 mL capacity of volumetric flask and volume was made upto 10 mL with methanol. This solution was analyzed using UV-Spectroscope at $\lambda_{max}$256 nm.

**Extrudability study**

Extrudability was based upon the quantity of the gel extruded from collapsible tube on application of certain load. More the quantity of gel extruded shows better extrudability. It was determine by applying the weight on gel filled collapsible tube and recorded the weight on which gel was extruded from tube [9].

**Spreadibility**

Spreadibility of formulation is necessary to provide sufficient dose available to absorb from skin to get good therapeutic response. It was determined by method reported by Multimer *et al.* An apparatus in which a slide fixed on wooded block and upper slide has movable and one end of movable slide tied with weight pan.To determine spreadibility, placing 2-5 g of gel between two slide and gradually weight was increased by adding it on the weight pan and time required by the top plate to cover a distance of 10 cm upon adding 80g of weight was noted. Good spreadibility show lesser time to spread [10].

$$\textbf{\textit{Spreadability}} = \frac{Weight tide to Upper Slide x\ Length\ moved\ on\ the\ glass\ slide}{Time\ taken\ to\ slide}$$

***In-vitro* drug diffusion study**

The *in-vitro* diffusion study is carried by using franz diffusion cell. Egg membrane is taken as semi permeable membrane for diffusion. The franz diffusion cell has receptor compartment with an effective volume approximately 60 mL and effective surface area of permeation 3.14sq.cms. The egg membrane is mounted between the donor and the receptor compartment. A two $cm^2$ size patch taken and weighed then placed on one side of membrane facing donor compartment. The receptor medium is phosphate buffer pH 7.4. The receptor compartment is surrounded by water jacket so as to maintain the temperature at 32±0.5°C. Heat is provided using a thermostatic hot plate with a magnetic stirrer. The receptor fluid is stirred by Teflon coated magnetic bead which is placed in the diffusion cell. During each sampling interval, samples are withdrawn and replaced by equal volumes of fresh receptor fluid on each sampling. The samples withdrawn and analyzed spectrophotometrically at wavelength of 256nm [11].

**Results and Discussion**

The study evaluates various formulations of acyclovir-loaded invasomes, focusing on parameters such as entrapment efficiency and vesicle size. Among the formulations tested, F5 demonstrates the highest entrapment efficiency at 75.65% with a relatively smaller average vesicle size of 210.32 nm. This indicates F5 as a promising candidate for further optimization due to its efficient encapsulation of acyclovir within smaller vesicles, potentially enhancing drug delivery efficacy.

Moving to the gel formulations of invasomes, characterized in Table 5, formulations like IG-2 stand out with a viscosity of 3465 cps and a pH of 6.74. These characteristics are crucial for topical applications, influencing factors such as stability on storage and skin compatibility. IG-2 also exhibits significant drug content (98.95%) and demonstrates intermediate values for extrudability and spreadability, which are essential for user-friendliness and uniform application.

The in vitro drug release studies presented in Table 6 highlight IG-2's sustained release profile over 12 hours, with cumulative drug release reaching 94.45% by the end of the period. This sustained release pattern suggests a potential for prolonged therapeutic effect, aligning with patient compliance and reduced dosing frequency.

Further analysis in Table 7 delves into the kinetics of drug release from IG-2, fitting well with the Higuchi and Korsmeyer-Peppas models, indicating diffusion-controlled release mechanisms. This

detailed characterization underscores the formulation's predictability in drug release kinetics, crucial for optimizing therapeutic outcomes and dosage regimens.

The regression analysis in Table 8 reaffirms the suitability of the Higuchi and Korsmeyer-Peppas models for IG-2, with high coefficients of determination ($R^2$), validating the formulation's controlled release mechanism.

**Table 3: Entrapment efficiency and average vesicle size of acyclovir loaded invasomes**

| Formulation Code | % Entrapment efficiency | Average vesicle size (nm) |
|---|---|---|
| F1 | 68.89±0.32 | 295.65±0.25 |
| F2 | 65.85±0.15 | 267.78±0.14 |
| F3 | 70.36±0.26 | 248.96±0.36 |
| F4 | 71.12±0.22 | 238.87±0.32 |
| F5 | 75.65±0.18 | 210.32±0.44 |
| F6 | 69.98±0.33 | 226.65±0.25 |

**Table 4: Characterization of optimized formulation of invasomes**

| Formulation | Average vesicle size (nm) | % Entrapment efficiency | Zeta Potential (mV) |
|---|---|---|---|
| F5 | 75.65±0.18 | 210.32±0.44 | -37.45 |

**Table 5: Characterization of Invasomes gel**

| Gel formulation | Viscosity (cps) | pH | Drug Content (%) | Extrudability (g) | Spreadibility (g.cm/sec) |
|---|---|---|---|---|---|
| IG-1 | 3572 | 6.95 | 97.74 | 165 | 11.32 |
| IG-2 | 3465 | 6.74 | 98.95 | 172 | 9.85 |
| IG-3 | 3345 | 6.32 | 96.32 | 185 | 7.45 |

**Table 6: *In vitro* drug release study of gel formulation**

| S. No. | Time (hr) | % Cumulative Drug Release* | | |
|---|---|---|---|---|
| | | IG-1 | IG-2 | IG-3 |
| 1 | 0.5 | 23.32 | 20.32 | 18.85 |
| 2 | 1 | 36.65 | 33.12 | 29.98 |
| 3 | 2 | 49.98 | 45.65 | 39.98 |
| 4 | 4 | 68.85 | 62.23 | 46.65 |
| 5 | 6 | 86.65 | 74.45 | 59.98 |
| 6 | 8 | 97.74 | 83.32 | 68.85 |
| 7 | 10 | 98.85 | 94.45 | 78.85 |
| 8 | 12 | 99.45 | 98.85 | 89.98 |

**Table 7: *In-vitro* drug release data for optimized formulation IG-2**

| Time (h) | Square Root of Time(h)$^{1/2}$ | Log Time | Cumulative* % Drug Release | Log Cumulative % Drug Release | Cumulative % Drug Remaining | Log Cumulative % Drug Remaining |
|---|---|---|---|---|---|---|
| 0.5 | 0.707 | -0.301 | 20.32 | 1.308 | 79.68 | 1.901 |
| 1 | 1 | 0 | 33.12 | 1.520 | 66.88 | 1.825 |
| 2 | 1.414 | 0.301 | 45.65 | 1.659 | 54.35 | 1.735 |
| 4 | 2 | 0.602 | 62.23 | 1.794 | 37.77 | 1.577 |
| 6 | 2.449 | 0.778 | 74.45 | 1.872 | 25.55 | 1.407 |
| 8 | 2.828 | 0.903 | 83.32 | 1.921 | 16.68 | 1.222 |
| 10 | 3.162 | 1 | 94.45 | 1.975 | 5.55 | 0.744 |
| 12 | 3.464 | 1.079 | 98.85 | 1.995 | 1.15 | 0.061 |

**Table 8: Regression analysis data of optimized gel formulation IG-2**

**Table 8: Regression analysis data of optimized gel formulation IG-2**

| Batch | Zero Order | First Order | Higuchi | Korsmeyer Peppas |
|---|---|---|---|---|
| | $R^2$ | $R^2$ | $R^2$ | $R^2$ |
| **IG-2** | 0.944 | 0.910 | 0.994 | 0.992 |

## CONCLUSION

In conclusion, the data collectively supports the potential of formulation F5 for invasomes and IG-2 for gel formulations in delivering acyclovir effectively. These formulations exhibit favorable characteristics such as high entrapment efficiency, controlled release kinetics, and suitable physicochemical properties for topical application. Future research could explore enhancing stability, modifying release kinetics, or investigating novel applications to further optimize these formulations for clinical use.

## REFERENCES

1. Corey L, Nahmias AJ, Guinan ME, Benedetti JK, Critchlow CW and Holmes KK. A trial of topical acyclovir in genital herpes simplex virus infections. N Engl J Med. 1983;308(12):693-696.
2. Spruance SL, Overall JC Jr, Kern ER, Krueger GG, Pliam V and Miller W. The natural history of recurrent herpes simplex labialis: implications for antiviral therapy. N Engl J Med. 1982;306(15):945-949.
3. Touitou E, Dayan N, Bergelson L, Godin B and Eliaz M. Ethosomes - novel vesicular carriers for enhanced delivery: characterization and skin penetration properties. J Control Release. 2000;65(3):403-418.
4. El-Nabarawi MA, Shamma RN, Farouk F and Nasralla SM. Dapsone-loaded invasomes as a potential treatment of acne: preparation, characterization, and in vivo skin deposition assay. Aaps Pharmscitech. 2018;19(5):2174-84.
5. P. K. Lakshmi, B. Kalpana and D. Prasanthi. Invasomes-novel Vesicular Carriers for enhanced skin permeation (2014).

6. Haag SF, Fleige E, Chen M, Fahr A, Teutloff C, Bittl R, et al. Skin penetration enhancement of core-multishell nanotransporters and invasomes measured by electron paramagnetic resonance spectroscopy. Int J Pharm 2011; 416:223-8.
7. Ota Y, Hamada A, Nakano M and Saito H. Evaluation of percutaneous absorption of midazolam by terpenes. Drug Metab. Pharmacokinet. 2003;18:261–266.
8. Vaddi H, Ho P, Chan Y and Chan S. Terpenes in ethanol: Haloperidol permeation and partition through human skin and stratum corneum changes. J. Control. Release. 2002;81:121–133.
9. Multimer, M.N. Riffskin, C. and Hill J.A.J. Am. Pharm. Assso., 45, 212-214.
10. Higuchi T. Mechanism of sustained-action medication: Theoretical analysis of rate of release of solid drugs dispersed in solid matrices. J Pharm Sci. 1963; 52:1145–9.
11. Korsmeyer RW, Gurny R, Doelker EM, Buri P and Peppas NA. Mechanism of solute release from porous hydrophilic polymers. Int J Pharm. 1983; 15:25–35.
12. Balouiri M, Sadiki M, Ibnsouda SK. Methods for in vitro evaluating antimicrobial activity: A review. J. Pharm. Anal. 2016, *6*, 71– 79.