

Formulation and Evaluation of Microsphere Loaded Gel Containing Tretinoin and Fusidic Acid and Its Anti-Microbial Activity

Rampal Dhakad¹, Dr. Brajesh Kumar Arjariya², Mrs. Sheenam Mansuri³

¹Student- Malhotra College of Pharmacy, Badwai, Near Karond, Bhopal (Madhya Pradesh)

²Professor - Malhotra College of Pharmacy, Badwai, Near Karond, Bhopal, (M.P.)

³Associate Professor - Malhotra College of Pharmacy, Badwai, Near Karond, Bhopal, (M.P.)

Date of Submission: 25-01-2026

Date of Acceptance: 05-02-2026

ABSTRACT

The present investigation focused on the physicochemical evaluation, analytical method development, and microsphere formulation of Tretinoin and Fusidic acid for novel drug delivery applications. Organoleptic evaluation confirmed that Tretinoin is a yellow, odorless solid powder and Fusidic acid is a white, odorless solid, both complying with Indian Pharmacopoeia specifications. Solubility studies revealed that Tretinoin was freely soluble in dimethyl sulfoxide and methanol and soluble in ethanol and chloroform, while Fusidic acid was freely soluble in dimethyl sulfoxide, methanol, and ethanol but insoluble in water. The pH values of Tretinoin and Fusidic acid were found to be 6.4 and 5.7, respectively, and melting points were observed at 178 °C and 193 °C, confirming drug purity and stability. UV-visible spectrophotometric analysis showed λ_{max} values of 353 nm for Tretinoin and 228 nm for Fusidic acid, with a common wavelength of 263 nm suitable for simultaneous estimation. Linearity studies demonstrated good correlation over concentration ranges of 2–14 $\mu\text{g/mL}$ for Tretinoin ($R^2 = 0.9865$) and 5–35 $\mu\text{g/mL}$ for Fusidic acid ($R^2 = 0.9941$), indicating the reliability of the analytical method. Microspheres were successfully prepared using the solvent evaporation technique with HPMC and ethyl cellulose as polymers. Particle size analysis using a Malvern zeta sizer showed that all formulations were in the nanometer range (150.4–217.8 nm), with formulation F1 exhibiting the smallest particle size, suggesting uniformity and stability. Overall, the study confirms that the developed microsphere formulations meet the required physicochemical and analytical standards and demonstrate potential as an effective and stable novel drug delivery system for topical and other pharmaceutical applications.

Keywords: Tretinoin; Fusidic acid; Microspheres; Solubility studies; UV-visible spectroscopy; Particle size analysis

I. INTRODUCTION

Microspheres defined as solid spherical particles, approximately the size ranges from 1 to 1000 μm containing dispersed drug molecules either in solution or crystalline forms (Lengyel et al., 2019). They are shallow spherical, free-flowing powders consisting of proteins polymers or synthetic polymers which are biodegradable in nature. Microspheres are a polymeric matrix system which contains the drug in a state of uniform distribution throughout the matrix (Lengyel et al., 2019). Polymers such as ethyl cellulose are used for the preparation of matrix-type microspheres of water-soluble drugs to control the dissolution rate of drugs from the dosage forms. Transdermal gels are a semisolid system, they prepared from a liquid which is thickened with other ingredients. The drug release through skin membrane and preparation of gelling agent sodium alginate is used (Bhuyan et al., 2021).

Tretinoin (TRT) is a widely used drug in the topical treatment of acne, photo-aged skin, psoriasis and other skin disorders. It also has been used to treat mottled hyper pigmentation, roughness, and fine wrinkling of photo damaged skin and to induce remission in acute promyelocytic leukemia (Rahman et al., 2015). In addition, topical TRT has been tried in a wide range of skin conditions such as rosacea, keratinisation disorders, pigmentation disorders, and some neoplastic disorders. The effect of TRT in the treatment of acne is to reduce the number and the size of comedones and therefore, TRT is commonly used at various dosage forms, such as lotions, hydrogels or creams (Mwamba et al., 2023).

Despite having these potentials advantageous its photo degradability can cause skin sensitization. Furthermore, topical TRT often causes transitory stinging, erythema, peeling, edema, dryness, or itching, which can result in poor patient compliance. To overcome problems associated with topical delivery of TRT, microsphere based formulations for topical delivery has been considered in the present study(Rai and Ravikumar 2016). The particulate delivery systems may be considered for topical delivery of medications because of their potentials of providing a controlled release rate and enhanced deposition of drug into the hair follicles, where drug is most needed. The use of microspheres has provided for a higher concentration of drugs in deeper layers of the skin and a reduction in Percutaneous absorption and unwanted side-effects(Midhaet al., 2015).

Fusidic acid is mainly used in skin and soft tissue infections. The common skin infections in which fusidic acid is used are impetigo, erythrasma, bullous impetigo, psoriasis, folliculitis, furuncles, carbuncles, contagiosa, infected wounds, and burns(Ingen-Housz-Oro et al., 2016).

Fusidic acid is bacteriostatic agent obtained from the fungus *Fusidium coccineum*. It belongs from class fusidanes. It is a steroid antibiotic of narrow spectrum, which is predominantly active against gram-positive bacteria. It is mainly active against *Staphylococcus aureus*, *S. epidermis*, *Clostridium* spp., and corynebacterial (Silva-Santana et al., 2021). *S. aureus* is one of the species that is a leading threat to public health and causes morbidity or mortality. Fusidic acid inhibits protein synthesis of bacteria by interfering with its elongation factor G (translocase) and may be by other mechanisms. Fusidic acid acts through 4 phases, i.e., Initiation, elongation, translocation, and release(Fernandes, 2016).

The present study aims to formulate and evaluate a microsphere loaded gel containing tretinoin and Fusidic acid and microbial activity.

II. MATERIALS AND METHODS

2.1 Chemicals

Methyl paraben, HPMC, n-octanol, Ethyl cellulose, DCM, DMSO, Propylene glycol, Sodium hydroxide and Ethanol were obtained from Merck, a reputable supplier of analytical reagents. Sulab provided the Carbopol 934, and Rankem provided the Methanol. KBr and EC were obtained from Sigma-Aldrich.

2.2 Pre-formulation studies

2.2.1 Organoleptic Properties

The organoleptic studies of Tretinoin and Fusidic acid like general appearance like color, odor, state, etc. were observed.

2.2.2 Solubility study

Qualitative solubility of Tretinoin and Fusidic acid in different solvents was determined according to USP NF, 2007. Approximately 1 mg of Tretinoin and Fusidic acid (separately) was weighed and transferred into a 10 ml test tube and dissolved in the respective solvents (1 ml each of methanol, ethanol, DMSO, chloroform and water) (Jain and Verma 2020).

2.2.3 pH determination

pH was determined by Electrochemical method. Digital pH meter is used to determine the pH of Tretinoin and Fusidic acid.

2.2.4 Melting Point

Melting point was analyzed by open Capillary method using Thiele's tube(Chowk, M. I. 2020).

2.2.5 Determination of Lambda max and calibration curve

Preparation of standard stock solution:

- **Preparation of standard stock solution:**

About 5mg of Tretinoin and 5 mg Fusidic acid was weighed and transferred into 5ml volumetric flask (Separately). The volume was made up to 5ml using respective solvent to obtain a solution that has a concentration 1000 µg/ml. 1ml of this stock solution was taken and then diluted up to 10 ml using methanol solvent to obtain a solution that has a concentration 100 µg/ml which is standard stock solution (Both tretinoin and Fusidic acid).

- **Lambda max**

From the above stock solution (Tretinoin and Fusidic acid) 0.5 ml sample was transferred into a 5 ml volumetric flask (separately) and the volume was made up to mark with solvent to prepare a concentration of 10 µg/ml. The sample was scanned by UV-VIS Spectrophotometer in the range of 200- 400 nm for tretinoin and Fusidic acid, using methanol solvent as a blank. The wavelength corresponding to the maximum absorbance (max) was found (Kumbhar and Salunkhe 2013).

• **Linearity**

Aliquots of 2 to 14 µg/ml and 5 to 35 µg/ml prepared utilizing 100 µg/ml Tretinoin and Fusidic acid working standard solution were accurately transferred into a series of 5 ml calibrated flask and made up to the mark with solvent. The absorbance of the resulting solution was measured 353.0 nm for Tretinoin and 228.0 nm for Fusidic acid against solvent blank. Calibration curve was prepared by plotting the absorbance vs concentration of drugs.

Seven points calibration curve were obtained in a concentration range from 2-14 and 5 to 35 µg/ml for drugs. The response of the drug was found to be linear in the investigation concentration range and the linear regression equation (Tretinoin was $y = 0.0514x + 0.0416$ with correlation coefficient $R^2 = 0.9865$ and (Fusidic acid) was $y = 0.0172x + 0.0166$ with correlation coefficient $R^2 = 0.9941$ (Behera et al., 2012).

2.2.6 Fourier transmission Infra-Red Spectroscopy

FT-IR spectrum of Tretinoin and Fusidic acid were recorded over the range of 4000 to 400

cm⁻¹ by KBr pellet method using a FT-IR spectrophotometer. The KBr disc was prepared using 1 mg of each Tretinoin and Fusidic acid in 100 mg of spectroscopic grade KBr which has been dried using IR lamp. Both KBr and drug was mixed and subjected to hydraulic pressure to form disc. This disc was placed in FT-IR chamber. Infrared spectrum was recorded in the 4000 - 400 cm⁻¹ region (Chowk, M. I. 2020).

2.3 Formulation of microspheres by Solvent Evaporation method

Microspheres containing Tretinoin and Fusidic acid as a core material were prepared by Solvent Evaporation method. Tretinoin and Fusidic acid, HPMC and EC were dissolved in a mixture of ethanol and dichloromethane (1:1) at room temperature (As in table 6). This was poured into 250 mL water containing 0.01% Tween-80 maintained at a temperature of 30–40 °C and subsequently stirred at 300 rpm agitation speed for 45 minutes to allow the volatile solvent to evaporate. The microspheres formed were filtered, washed with water and dried in oven at 37°C (Fartyalet al., 2011).

Table 1: Composition of microsphere formulation

Formulations (Code)	Polymer HPMC (mg)	Polymer Ethyl cellulose (mg)	Tretinoin and Fusidic acid (%)	Temperature °C	Solvent ratio(1:1) ethanol/DCM
F1	300	50	0.025 and 2.0	30-40°C	5ml:5ml
F2	250	100	0.025 and 2.0	30-40°C	5ml:5ml
F3	200	150	0.025 and 2.0	30-40°C	5ml:5ml
F4	150	200	0.025 and 2.0	30-40°C	5ml:5ml
F5	100	250	0.025 and 2.0	30-40°C	5ml:5ml

2.4 Evaluation parameter of drug loaded microsphere

2.4.1 Particle size

The size of microspheres was measured using Malvern Zeta sizer (Malvern Instruments) (Singh and Vingkar 2008).

2.4.2 Zeta potential

The zeta potential was measured for the determination of the movement velocity of the particles in an electric field and the particle charge. In the present work, the microspheres was diluted 10 times with distilled water and analyzed by Zetasizer Malvern instruments (Martín et al., 2015).

2.4. 3 Quantitative analysis (Entrapment Efficiency)

%Entrapment efficiency was determined by indirect estimation. Drug -loaded microspheres were centrifuged at 15,000 rpm for 30 min using REMI Ultra Centrifuge. The non-entrapped drug (free drug) was determined in the supernatant solution using UV spectrophotometer (Guillot et al., 2015).

Entrapment efficiency % = Total drug conc. - Supernatant drug conc. / total drug conc.*100

2.4.4 Scanning Electron Microscopic (SEM)

The electron beam from a scanning electron microscope was used to attain the morphological features of the drug loaded microspheres were coated with a thin layer (2–20

nm) of metal(s) such as gold, palladium, or platinum using a sputter coater under vacuum. The pretreated specimen was then bombarded with an electron beam and the interaction resulted in the formation of secondary electrons called auger electrons. From this interaction between the electron beam and the specimen's atoms, only the electrons scattered at 90° were selected and further processed based on Rutherford and Kramer's Law for acquiring the images of surface topography (Ahmed et al., 2020).

2.5 Formulation of Microsphere loaded Gel

Initially carbopol-934 was immersed in 50 mL of warm water (A) for 2 hr and was

homogeneously dispersed using magnetic stirrer at 600 rpm. In separate container carboxymethyl cellulose and methyl paraben was added into 50 ml warm water (B) and stirred continuously to make stiff gel. Both the mixtures A and B were mixed with the continuous stirring. Then tri-ethanol amine (Drop wise) was added to neutralize the pH and microsphere of optimized formulation was incorporated into the dispersion to obtained Gel. At this stage, permeation enhancer (Propylene glycol) was added. The final dispersion was agitated until smooth gel was formed without lumps (Ansari et al., 2024).

Table 2: Composition of gel formulation

Excipients	Quantity
Carbopol 934	1.00 gm
Carboxymethyl cellulose	1.00 gm
Propylene glycol	0.5 ml
Methyl paraben	0.2 ml
Microsphere	1.0 gm
Tri-ethanolamine	q.s
Water	100 ml

2.6 Characterization of microsphere loaded Gel

2.6.1 Physical appearance

The prepared Gel formulation was evaluated for appearance, Color, Odor, and homogeneity by visual observation (Robiatunet al., 2022).

2.6.2 pH

pH of the formulation was determined by using Digital pH meter (EI).

2.6.3 Viscosity

The viscosity of the gel formulations was determined using Brookfield viscometer with spindle no. 61 at 100 rpm at the temperature of 25°C (Giriet al., 2019).

2.6.4 Spreadability

An ideal topical gel should possess a sufficient spreading coefficient when applied or rubbed on the skin surface. This was evaluated by placing about 1g of formulation on a glass slide. Another glass slide of the same length was placed above that, and a mass of 50 mg was put on the glass slide so that the gel gets sandwiched between the two glass slides and spreads at a certain distance. The time taken for the gel to travel the

distance from the place of its position was noted down. Spreadability was determined by the following formula

$$S = \frac{M \cdot L}{T}$$

Where, S-Spreadability, g.cm/s M-Weight put on the upper glass L-Length of glass slide T-Time for spreading gel in sec (Jha et al., 2024).

2.7 Antibacterial activity of Microsphere by Well diffusion assay

• Preparation of Nutrient Agar Media

28 g of Nutrient Media was dissolved in 1 litre of distilled water. pH of media was checked before sterilization. Media was sterilized in autoclave at 121°C at 15 lbs pressure for 15 minutes. Nutrient media was poured into plates and placed in the laminar air flow until the agar was get solidified.

• Well Diffusion Assay

The bacterial suspension of E. coli was standardized to 10⁸ CFU/ml of bacteria and kept into the shaker. Then, 100µl of the inoculums from the broth (containing 10⁸ CFU/ml) was taken with a micropipette and then transferred to fresh and sterile solidified Agar Media Plate (Mohammadi-Sichaniet al., 2012). The agar plate was inoculated

by spreading the inoculums with a sterile spreader, over the entire sterile agar surface. Three wells of 6 mm were bored in the inoculated media with the help of sterile cork-borer. The wells were then formed for the inoculation of the microsphere, Microsphere (0.5, 1 and 2mg/ml) solution. 100 µl of the sample was loaded. It was allowed to diffuse for about 30 minutes at room temperature and incubated for 18-24 hours at 37° C. After incubation, plates were observed for the formation of a clear zone around the well which corresponds to the antimicrobial activity of tested compounds. The zone of inhibition (ZOI) was observed and measured in mm. Zones were measured to a nearest millimeter using a ruler, which was held on the back of the inverted Petri plate. The Petri plate was held a few inches above a black, non-reflecting background. The diameters of the zone of complete inhibition (as judge by unaided eye) were

measured, including the diameter of the well (Manandharet al., 2019).

2.8 Stability studies

The drug loaded Microsphere formulation was packed and were placed in the stability test chamber and subjected to stability studies at accelerated testing (25°C±2°C and 60 ± 5% RH) and (40°C±2°C and 70 ±5% RH) for 3 months. The formulation was checked for evaluation parameter particle size and entrapment efficiency studies at the interval of 30, 45, 60, 90 days (3 month) months. The formulation was tested for stability under accelerated storage condition for 3 months in accordance to International Conference on Harmonization (ICH) guidelines. Formulation was analyzed for the change in evaluation parameter particle size and entrapment efficiency studies.

III. RESULT AND DISCUSSION

3.1 Pre-formulation study of drug

3.1.1 Organoleptic properties

Table 3: Organoleptic properties of Tretinoin and Fusidic acid

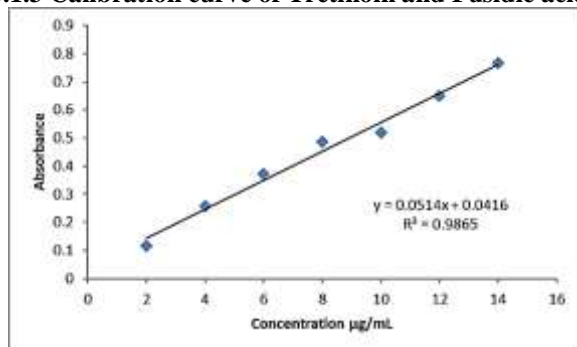
Drug	Organoleptic properties	Observation (Tretinoin)	Observation (Fusidic acid)
Tretinoin and Fusidic acid	Color	Yellow	White
	Odor	Odour less	Odour less
	Appearance	Solid powder	Solid powder
	State	Solid	Solid

3.1.2 pH and Melting point determination of Tretinoin and Fusidic acid

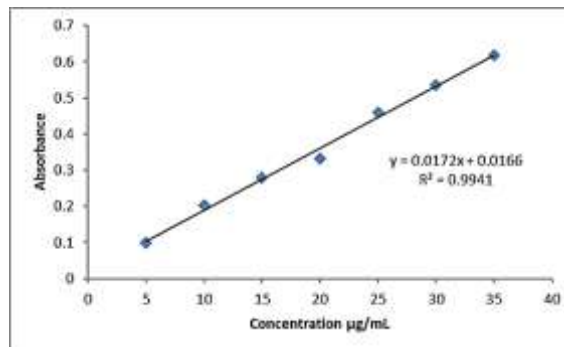
Table 4: pH and Melting point determination of Tretinoin and Fusidic acid

Drugs	Observed (pH)	Observed (Melting point)	Reference (Melting point)
Tretinoin	6.4	178°C	175°C-180°C
Fusidic acid	5.7	193°C	192°C-193°C

3.1.3 Calibration curve of Tretinoin and Fusidic acid

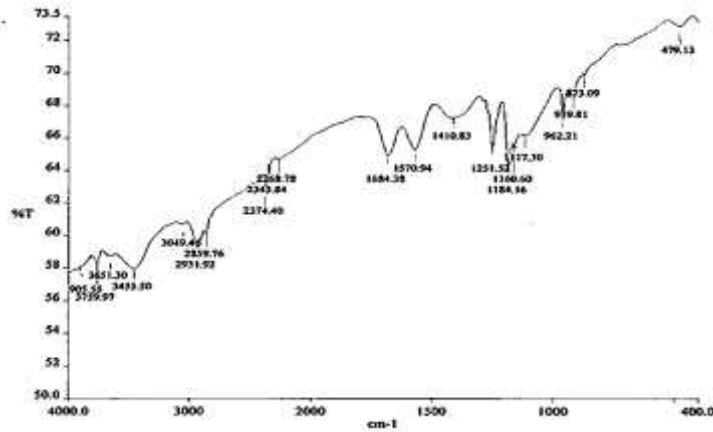


Graph 1: Calibration curve of Tretinoin



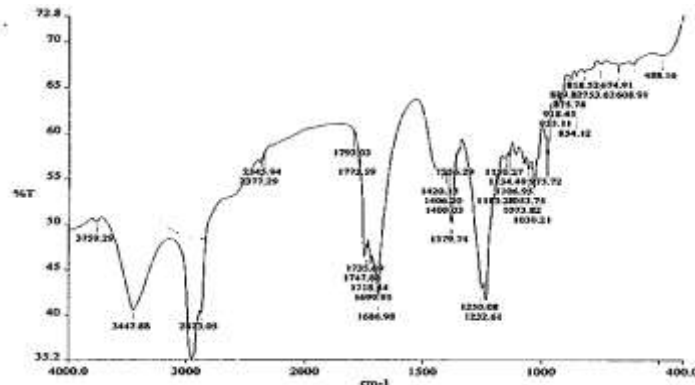
Graph 2: Calibration curve of Fusidic acid

3.1.4 Functional group identified by Fourier transform infrared (FTIR) study
1. FTIR of Tretinoin



Graph 3: Tretinoin

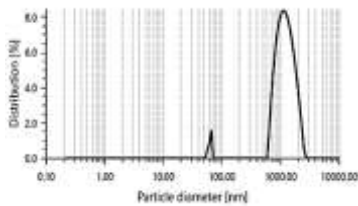
2. FTIR of Fusidic acid



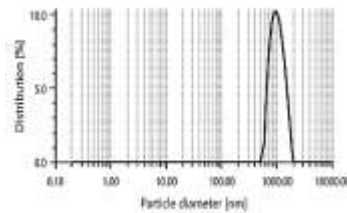
Graph 4: Fusidic acid

3.2 Evaluation parameter of microsphere formulations

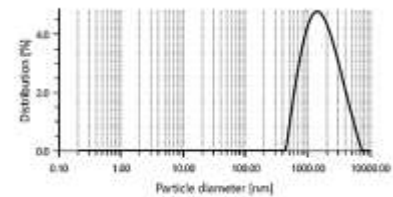
3.2.1 Particle size determination



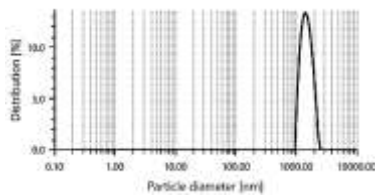
Graph 5: Particle size (F1)



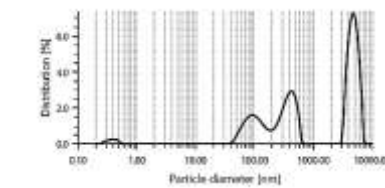
Graph 6: Particle size (F2)



Graph 7: Particle size (F3)



Graph 8: Particle size (F4)



Graph 9: Particle size (F5)

Table 5: Result of Particle size of all formulations

Formulations	Particle size (nm)	PI Value
Microsphere (F1)	153.4 nm	0.226
Microsphere (F2)	179.5 nm	0.395
Microsphere (F3)	220.8 nm	0.446
Microsphere (F4)	154.5nm	0.318
Microsphere (F5)	154.5nm	0.213

Discussion

The physicochemical and analytical evaluation of Tretinoin and Fusidic acid confirmed their compliance with Indian Pharmacopoeia specifications and suitability for formulation development. Organoleptic studies showed that Tretinoin was a yellow, odorless solid, while Fusidic acid was a white, odorless solid, matching official standards. Solubility studies revealed that Tretinoin was freely soluble in DMSO and methanol and soluble in ethanol and chloroform, whereas Fusidic acid was freely soluble in DMSO, methanol, and ethanol but insoluble in water, supporting the need for appropriate solvent selection.

The pH values of Tretinoin (6.4) and Fusidic acid (5.7) and their melting points (178 °C and 193 °C, respectively) were within specified limits, indicating purity and stability. UV-visible analysis showed λ_{max} values of 353 nm for Tretinoin and 228 nm for Fusidic acid, with a common wavelength of 263 nm suitable for simultaneous estimation. Linearity studies demonstrated good correlation within the tested concentration ranges for both drugs.

Particle size analysis of the microsphere formulations indicated sizes ranging from 150.4 to 217.8 nm, confirming successful microsphere formation, with formulation F1 exhibiting the smallest particle size. Overall, these results support the suitability of the drugs and the developed microsphere formulation for further pharmaceutical application.

IV. CONCLUSION

Tretinoin and Fusidic acid complied with official specifications for organoleptic properties, pH, melting point, and UV-visible absorption maxima, confirming their suitability for formulation. Drug-loaded microspheres were successfully prepared by the solvent evaporation method using HPMC and ethyl cellulose. SEM studies showed spherical, porous microspheres with smooth surfaces, while particle size analysis confirmed nanometer-sized particles (150.4–217.8 nm) with good stability and high entrapment

efficiency. The microsphere gel exhibited acceptable viscosity, pH, and spreadability, indicating suitability for topical application. Overall, the study demonstrates the potential of the developed microspheres as a stable and effective novel drug delivery system.

REFERENCES

- [1]. Lengyel, M., Kállai-Szabó, N., Antal, V., Laki, A. J., & Antal, I. (2019). Microparticles, microspheres, and microcapsules for advanced drug delivery. *Scientiapharmaceutica*, 87(3), 20.
- [2]. Lengyel, M., Kállai-Szabó, N., Antal, V., Laki, A. J., & Antal, I. (2019). Microparticles, microspheres, and microcapsules for advanced drug delivery. *Scientiapharmaceutica*, 87(3), 20.
- [3]. Bhuyan, C., Saha, D., & Rabha, B. (2021). A brief review on topical gels as drug delivery system. *J. Pharm. Res. Int*, 33, 344-357.
- [4]. Rahman, S. A., Abdelmalak, N. S., Badawi, A., Elbayoumy, T., Sabry, N., & Ramly, A. E. (2015). Formulation of tretinoin-loaded topical proniosomes for treatment of acne: in-vitro characterization, skin irritation test and comparative clinical study. *Drug Delivery*, 22(6), 731-739.
- [5]. Mwamba, R. N., Ekwonu, A., Guimaraes, P. V., & Raheem, O. A. (2023). The efficacy, safety, and outcomes of testosterone use among transgender men patients: A review of the literature. *Neurourology and Urodynamics*, 42(5), 921-930.
- [6]. Rai, S. Y., & Ravikumar, P. (2016). Development and evaluation of microsphere based topical formulation using design of experiments. *Indian Journal of Pharmaceutical Sciences*, 78(2), 182-192.

- [7]. Midha, K., Nagpal, M., & Arora, S. (2015). Microspheres: a recent update. *Int. J. Recent. Sci. Res*, 50(8), 5859-67.
- [8]. Ingen-Housz-Oro, S., Del Giudice, P., & Chosidow, O. (2016). Common skin bacterial infections. In *Antibiotic and antifungal therapies in dermatology* (pp. 1-20). Cham: Springer International Publishing.
- [9]. Silva-Santana, G., Silva, C. M. F., Olivella, J. G. B., Silva, I. F., Fernandes, L. M. O., Sued-Karam, B. R., ... & Mattos-Guaraldi, A. L. (2021). Worldwide survey of *Corynebacterium striatum* increasingly associated with human invasive infections, nosocomial outbreak, and antimicrobial multidrug-resistance, 1976–2020. *Archives of Microbiology*, 203(5), 1863-1880.
- [10]. Fernandes, P. (2016). Fusidic acid: a bacterial elongation factor inhibitor for the oral treatment of acute and chronic staphylococcal infections. *Cold Spring Harbor perspectives in medicine*, 6(1), a025437.
- [11]. Jain, N., & Verma, A. (2020). Preformulation studies of pilocarpine hydrochloride as niosomal gels for ocular drug delivery. *Asian Journal of Pharmaceutical and Clinical Research*, 149-155.
- [12]. Chowk, M. I. (2020). Preformulation study of terbinafine for novel drug delivery system formulation. *Tablet*, 250, 500mg.
- [13]. Kumbhar, S. C., & Salunkhe, V. R. (2013). UV Spectrophotometric Method development for Capecitabine Eudragit and Chitosan based Microspheres and its Validation. *Indian Journal of Pharmaceutical and Biological Research*, 1(03), 32-38.
- [14]. Behera, S., Ghanty, S., Ahmad, F., Santra, S., & Banerjee, S. (2012). UV-visible spectrophotometric method development and validation of assay of paracetamol tablet formulation. *J Anal Bioanal Techniques*, 3(6), 151-7.
- [15]. Chechare, D. D., & Siddaiah, M. (2024). Formulation and evaluation of mucoadhesive microspheres of metronidazole. *Journal of Applied Pharmaceutical Research*, 12(1), 93-99.
- [16]. Fartyal, S., Jha, S. K., Karchuli, M. S., Gupta, R., & Vajpayee, A. (2011). Formulation and evaluation of floating microspheres of boswellic acid. *Int J Pharm Tech Res*, 3, 76-81.
- [17]. Singh, K. K., & Vingar, S. K. (2008). Formulation, antimalarial activity and biodistribution of oral lipid nanoemulsion of primaquine. *International Journal of Pharmaceutics*, 347(1-2), 136-143.
- [18]. Martín, M. J., Calpena, A. C., Fernández, F., Mallandrich, M., Gálvez, P., & Clares, B. (2015). Development of alginate microspheres as nystatin carriers for oral mucosa drug delivery. *Carbohydrate polymers*, 117, 140-149.
- [19]. Guillot, A., Couffin, A. C., Sejean, X., Navarro, F., Limberger, M., & Lehr, C. M. (2015). Solid Phase Extraction as an Innovative Separation Method for Measuring Free and Entrapped Drug in Lipid Nanoparticles. *Pharmaceutical research*, 32(12).
- [20]. Ahmed, M. M., Fatima, F., Kalam, M. A., Alshamsan, A., Soliman, G. A., Shaikh, A. A., & Anwer, M. K. (2020). Development of spray-dried amorphous solid dispersions of tadalafil using glycyrrhizin for enhanced dissolution and aphrodisiac activity in male rats. *Saudi Pharmaceutical Journal*, 28(12), 1817-1826.
- [21]. Ansari, S. A., Bagre, A., & Jain, S. (2024). Preparation and evaluation of liposomal gel containing *Neolamarckiacadamba* leaves extract for anti-inflammatory activity. *Journal of Drug Delivery & Therapeutics*, 14(8).
- [22]. Robiatun, R. R., Pangondian, A., Paramitha, R., Rani, Z., & Gultom, E. D. (2022). Formulation and evaluation of hand sanitizer gel from clove flower extract (*Eugenia aromatica* L.). *International Journal of Science, Technology & Management*, 3(2), 484-491.
- [23]. Giri, M., Abhale, A., Ahire, M., & Bhalke, R. D. (2019). Formulation, Characterization, and Evaluation of Topical Anti-inflammatory Herbal Gel. *Int. J. Pharm. Biol. Arch*, 10, 190-195.
- [24]. Jha, S., Singh, P., Yadav, K., Rai, A., Saket, N., & Chouhan, M. (2024). Formulation And Development Of Diclofenac Topical Emulgels.
- [25]. Mohammadi-Sichani, M., Karbasizadeh, V., Aghai, F., & Mofid, M. R. (2012).



- Effect of different extracts of *Stevia rebaudiana* leaves on *Streptococcus mutans* growth. *J Med Plants Res*, 6(32), 4731-34.
- [26]. Manandhar, S., Luitel, S., & Dahal, R. K. (2019). In vitro antimicrobial activity of some medicinal plants against human pathogenic bacteria. *Journal of tropical medicine*, 2019.