
Formulation Design, Optimization and Evaluation of Mupirocin Emulgel

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ABSTRACT

Emulsified gels are stable and better vehicles for hydrophobic or poorly water soluble drugs. Mupirocin is an antibacterial drug. Mupirocin works to kill the bacteria which include strains of *Staphylococcus aureus* and *Streptococcus pyogenes*. Conventionally, Mupirocin is available in the form of ointments in most South East Asian countries. However, emulgels have been found to be better suited with an added advantage of sustained release, which is necessary for the treatment of skin infection. Mupirocin emulgels were formulated using PEGs, Carbomer 940 and flax seed oil. The pH of the emulgel formulations were found to be close to that of the human skin. Drug excipient compatibility analysis was carried out by FTIR. Emulgel was evaluated for its physical characterization, in-vitro release and antimicrobial properties. FTIR analysis confirmed drug excipient compatibility. Release studies indicated controlled release of drug carbomer 940 due to its higher gelling capacity. Formulation EG3 showed good results against *Staphylococcus aureus*. Mupirocin was stable in topical emulgel formulations and showed enhanced retention in the skin indicating better potential of the delivery system for treatment of primary and secondary skin infections, such as impetigo, eczema, and atopic dermatitis.

Key words: Emulgel; topical; mupirocin; antimicrobial; FTIR; carbomer 940.

INTRODUCTION

Emulgels

Gels are currently receiving increasing attention, especially hydrogel formulations, for topical application of drugs since they have an attractive appearance and develop pleasant cool feeling. Gels for topical use have several favorable properties such as being thixotropic, greaseless, easily spreadable, easily removable, non-staining, compatible with several excipients, and water-soluble or miscible [1]. Gel dosage forms are successfully used as drug delivery systems to control drug release and protect the medicaments from a hostile environment. In spite of many advantages of gels, a major limitation is in the delivery of hydrophobic drugs.

Since gels have a higher aqueous component, it limits the dissolution of hydrophobic drugs, and also retards migration of the drug through a vehicle that is essentially a liquid. To overcome this limitation, emulgels are prepared and used so that even a hydrophobic therapeutic moiety can enjoy the unique properties of gels [2].

Emulgels are emulsions either of oil-in-water or water-in-oil type, which is gelled by mixing with gelling agent. They have been recently used as vehicles to deliver various drugs to the skin and vagina [6]. Emulsified gels are stable and better vehicles for hydrophobic or poorly

water soluble drugs. They have the advantages of emulsions and gels. They are used to deliver various drugs to the mucosa due to enhanced drug absorption through the mucosa since the drug substance is in solution.

MATERIALS AND METHOD

The whole project was carried out at M/s Denium Laboratories Pvt. Ltd., Birgunj, Nepal. Mupirocin IP was sourced from Concord biotech Ltd., Gujarat., Carbomer 940 from Maruti Chemicals, Gujarat., Triethanolamine from Triveni Chemicals, Gujarat., PEG-400 from India Glycols Limited, Uttarakhand., PEG-4000 from India Glycols Limited, Uttarakhand., Di sodium EDTA from New Alliance Dye Chem, Mumbai., Glycerine from Salius Pharma Private Limited, Mumbai. Flax seed oil sourced from local purchase.

Preparation of emulgels comprises of simple and short steps which increase feasibility of the production. There are no specialized instruments needed for the production of emulgels. Moreover, materials used are easily available and cheap. All these; decrease the production cost of emulgels. The rheological properties and the breakdown behavior of gels filled with emulsions droplets can be varied by changing the interactions between oil droplets and gel matrix, the oil content and the oil droplet size [10].

Basically emulgel preparation consists of two steps; emulsification and incorporating the emulsion into a gel base (Fig.1 & 2). Important constituents of the emulgel are the aqueous phase, the oil phase and the gelling agents [2]. Commonly used aqueous agents are water and alcohols.

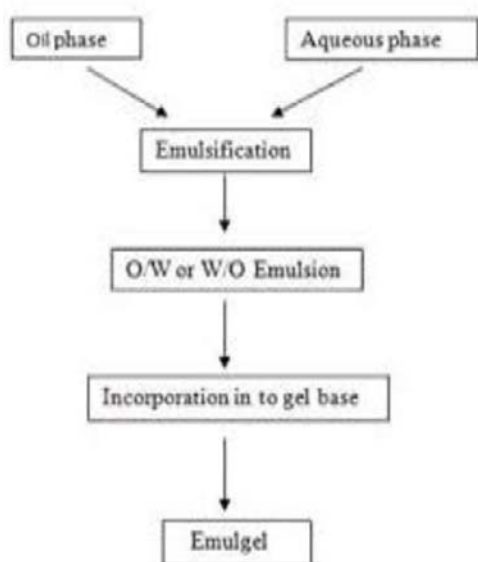


Fig.1. Preparation of Emulgel

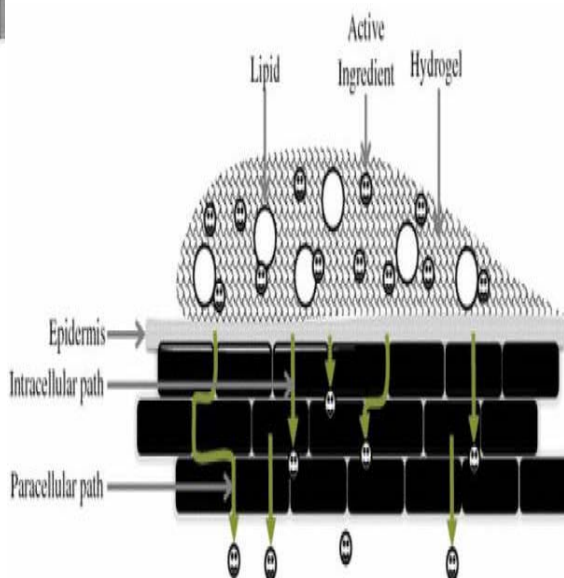


Fig.2. Schematic Representation of Emulgel System

PREFORMULATION STUDIES

Preformulation studies are important for the development of dosage form for achieving the goal of designing optimum drug delivery system.

Identification: The drug sample obtained was identified by various analytical techniques such as FTIR spectroscopy, UV spectroscopy, Melting point, Partition coefficient and Solubility etc.

Meting point: Melting point of Mupirocin was determined by taking small amount separately in a capillary tube closed at one end and placed in a Melting Point apparatus (PERFIT) and the temperature at which Mupirocin molten was recorded. This was performed in triplicate and average value was recorded. The melting point of Mupirocin was found to be 78°C, is similar to reported value indicating that no impurity is present in the sample.

Partition coefficient

Partition coefficient of Mupirocin was determined at 37 ± 0.5 °C by taking 5 ml of octanol which was saturated with 5 ml of water by shaking with externally driven magnetic stirrer. After shaking the system remained undisturbed for half an hour. About 100 mg of drug was added to this solution and was shaken on wrist action mechanical stirrer. Two layers were separate through separating funnel and filtered through Whatman grade filter, and the amount of Mupirocin solubilised, was determined by measuring the absorbance at 220 nm against reagent blank through double beam UV/Vis spectrophotometer (Shimadzu) in both the solution. Partition coefficient was determined as ratio of concentration of drug in octanol to the concentration of drug in water and the value were reported as log P. The Log p values were found to be 2.45 ± 0.12 .

Solubility Studies

Solubility studies were carried out with different solvents such as water, ethanol, and methanol in water bath shaker at 25°C and kept it for 24 hours.

<u>Solvents</u>	<u>Solubility (mg/ml) (mean \pmSD)*</u>
Distilled water	0.0265 \pm 0.009
Methanol	0.0229 \pm 0.007
DMSO	0.436 \pm 0.004

PREPARATION OF EMULGELS

Mupirocin was dissolved with flax seed oil and one half of the PEG-400 at 55°C under continuous stirring. Then the mixture of remaining PEG-400 and PEG-4000 (heated at 65°C and again cooled to 55°C) was incorporated with active solution. This emulsion is cooled to room temperature. In a separate SS container, purified water was poured and sequestered with Disodium EDTA. Now the carbomer 940 was wet with continuous stirring for 10 minutes. Then neutralized it to ~7 pH with Triethanolamine. A viscous transparent gel was obtained. Next, incorporation of the mupirocin emulsion to the gel under mild stirring for 20 minutes was done. In the meantime, glycerin added into it. Emulgels were immediately transferred into tubes for observation and stability study.

Table 1: Composition of different formulations

S. No.	Ingredients	EG1	EG2	EG3
1.	Mupirocin	2.04	2.04	2.04
2.	PEG-400	18	20	22
3.	PEG-4000	6	4	5
4.	Carbomer 940	4	3.3	3.0
5.	Triethanolamine	2	2	1.5
6.	Di sodium EDTA	0.1	0.1	0.1
7.	Glycerin	5	5	4
8.	Flax seed oil	8	10	12

9.	Purified water	qs to 100g	qs to 100g	qs to 100g
Total		100g	100g	100g

* Quantities in gram.

EVALUATION OF MUPIROCIN EMULGELS

Percentage (%) Yield

The prepared emulgels were collected and weighed. The measured weight was divided by the total weight of all the excipients and drug. The % yield was calculated.

Morphological Studies

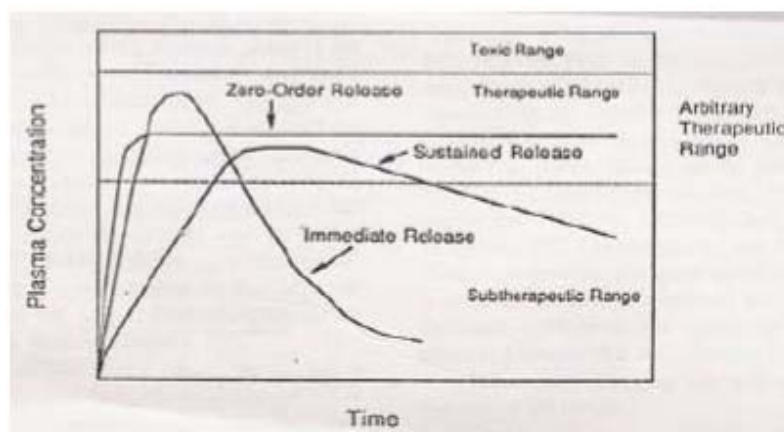
The morphology study of optimized formulation was performed by observing under compound microscope.

In-vitro Dissolution Studies

The emulgel equivalents to weight containing 100mg of mupirocin were immersed in dissolution medium. To assure the release of drug in solution at appropriate rate, dissolution test has been performed for optimized formulation in triplicate. The in-vitro release of mupirocin from the emulgel was examined using USP Type II dissolution apparatus. 6.8 phosphate buffer (900 ml) was used as the dissolution medium and maintained at $37 \pm 0.5^\circ\text{C}$ at a rotation speed of 100 rpm. An aliquot of 5 ml of the solution was withdrawn at predetermined time intervals and replaced by 5 ml of fresh dissolution medium. Samples were assayed with spectrophotometer at 220 nm after filtration through a $0.45 \mu\text{m}$ membrane filter (Millipore) against 6.8 phosphate buffer as blank.

Drug release Kinetic Studies

In the present study, raw data obtained from in vitro release studies was analyzed, wherein data was fitted to different equations and kinetics model to calculate the percent drug release and release kinetics of mupirocin from emulgel formulations. The kinetic models used were a Zero-order equation, First-order, Higuchi's model and Korsmeyer- Peppas equation. When the data was plotted as log cumulative percent drug remaining versus time yields a straight line, indicating that the release follows first-order kinetics. The constant 'K' can be obtained by multiplying 2.303 with slope values.

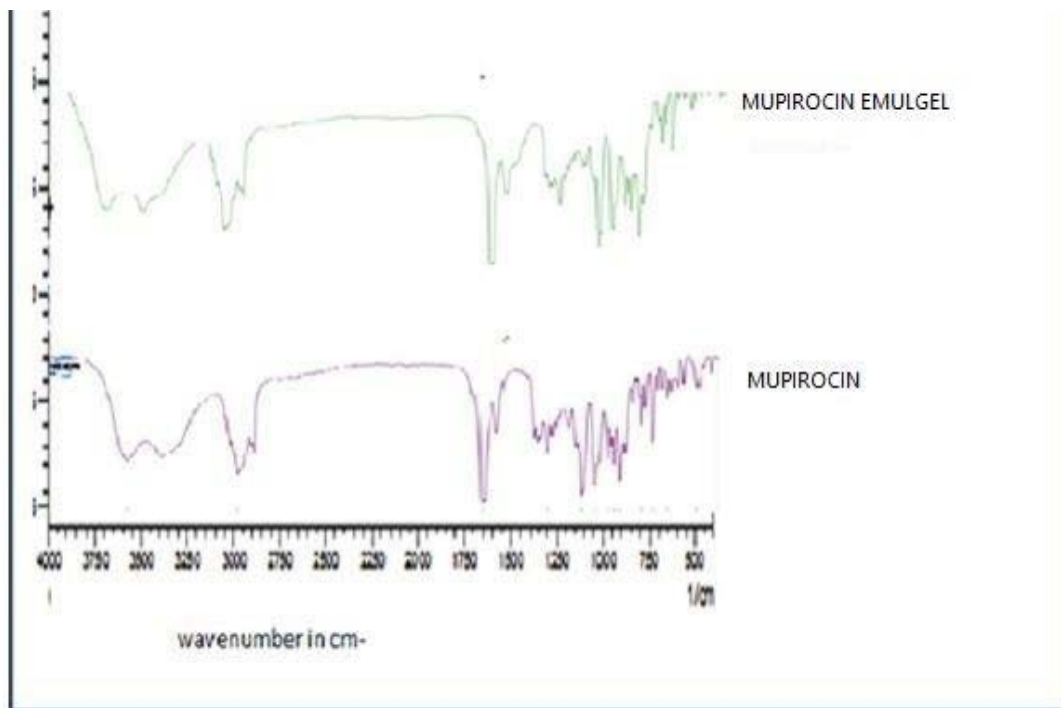


Graph1: Drug level versus time profile showing difference between zero order, controlled releases, slow first order sustained release and release from a conventional tablet

Accelerated Stability Studies

To assess the drug and formulation stability, stability studies were done according to the

International Council for Harmonization of Technical Requirements for Pharmaceuticals for Human Use (ICH) and World Health Organization recommended Good Manufacturing Practice (WHO-MGP) guidelines. The optimized Mupirocin Emulgel tubes were stored at $40\pm 2^\circ\text{C}/75\pm 5\%$ relative humidity for 6 months. Emulgels were analyzed at specified time intervals (0, 1, 2, 3 & 6 months) for the physicochemical properties and in vitro dissolution study.



Graph 2: FT-IR spectrum of Mupirocin Emulgel

RESULTS AND DISCUSSION

Physical Appearance

Emulgel formulations were white viscous creamy preparation with a smooth homogeneous texture and glossy appearance. Results have been discussed in Table 5.7.

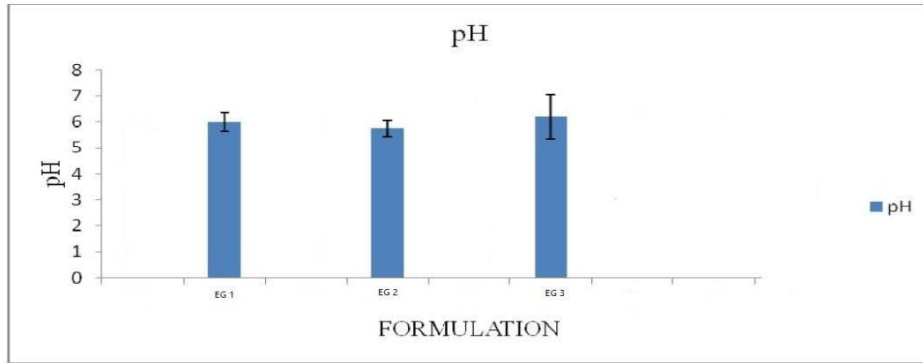
Table 2: Physicochemical characteristics of Mupirocin Emulgel formulations

S.N.	Formulation Code	Color	Phase Separation	Grittiness	Homogeneity	Consistency
1	EG1	White	None	-	Excellent	++
2	EG2	White	None	-	Good	+++
3	EG3	White	None	-	Excellent	+++

pH

The pH of the Emulgel formulations was in the range of 5.5 ± 0.54 to 6.4 ± 0.43 , which lies in the normal pH range of the skin and would not produce any skin irritation. There was no significant change in pH values as a function of time for all formulations.

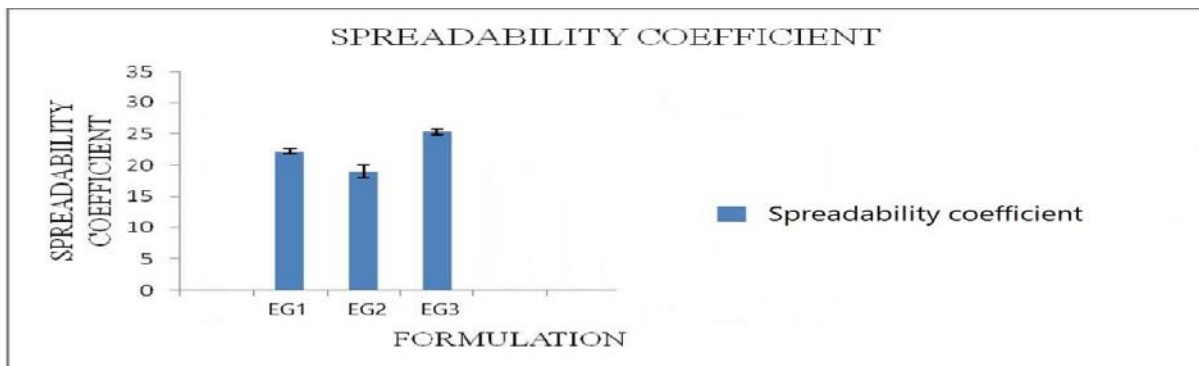
The data is shown below in Graph 3.



Graph 3: pH of Different Formulations EG1 - EG3

Spreadability Test

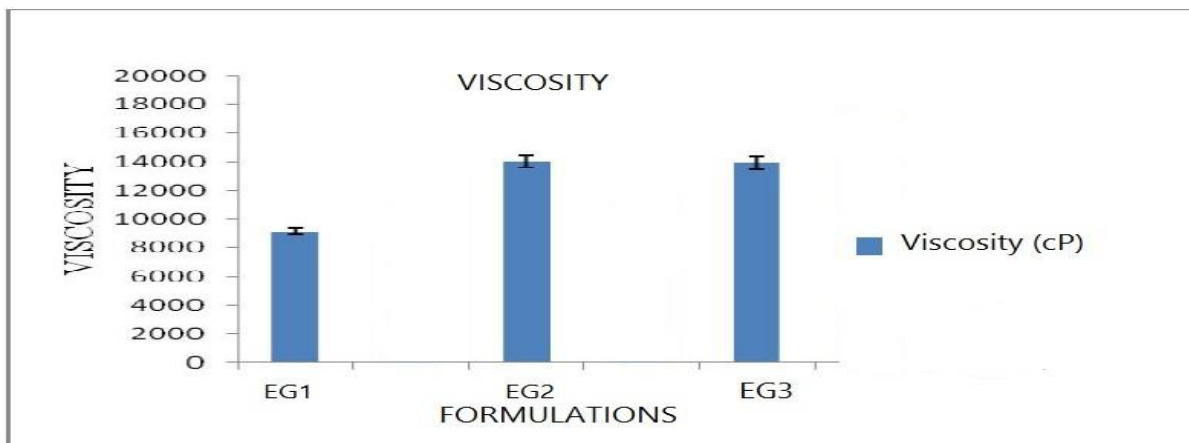
Spreadability tests were carried out for all the formulations. The spreadability indicates that the Emulgel is easily spreadable by small amount of shear. Spreadability of the Emulgel decreases with the increase in the concentration of the polymer. The spreadability is very much important as it shows the behaviour of Emulgel when it comes out from the tube as given in graph 2.



Graph 4: Spreading Coefficient of Different Formulations EG1 – EG3

Rheological Study

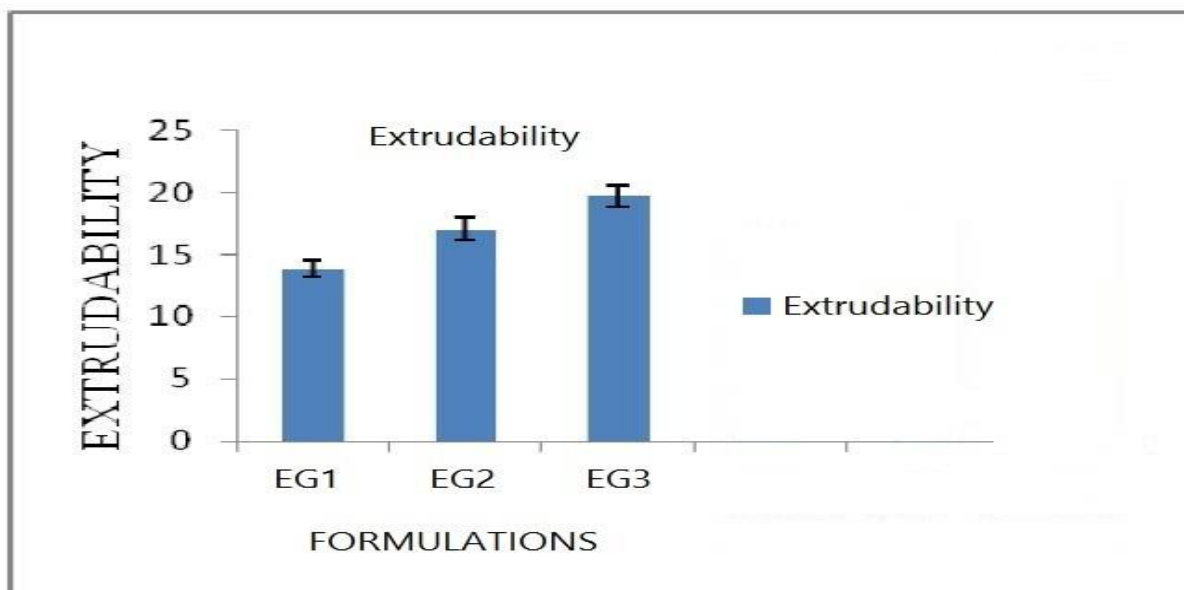
The Emulgel was rotated at 50 rpm for 10 min with spindle C25-1. The corresponding reading was noted. The viscosity of the Emulgel was obtained (Graph 3). The viscosity of the formulations increases as concentration of polymer increases.



Graph 5: Viscosities of Different Formulations EG1 - EG3

Extrudability

The gels were filled into collapsible tubes after formulating them. The extrudability of the formulation has been checked as shown below in graph 4.



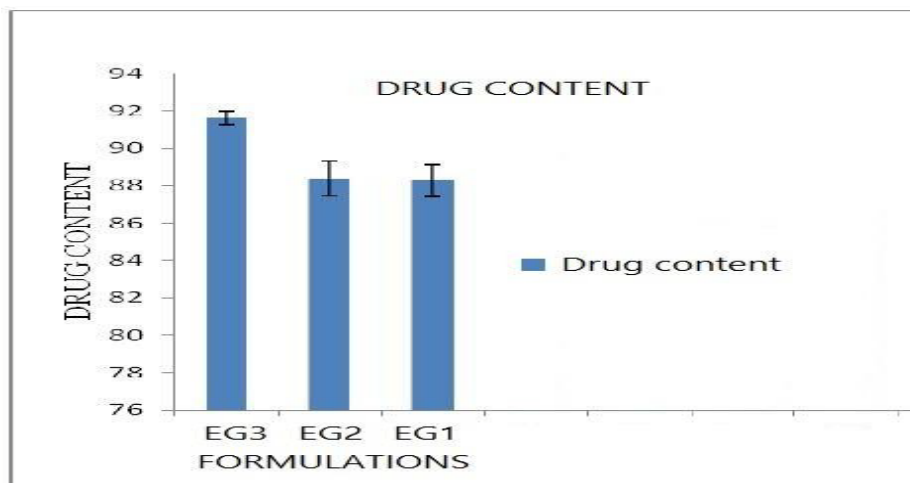
Graph 6: Extrudability of Formulations EG1 – EG3

Drug Content

The drug content of the formulated Emulgel was estimated spectrophotometrically at λ_{max} 220. The results were in the limits as shown in Table 4 and Graph 5.

Table 3: Drug content of all formulations

S.N.	Formulations	Drug Contents (% Assay)
1	EG 1	88.38 ± 0.92
2	EG2	88.26 ± 0.87
3	EG3	91.60 ± 0.37



Graph 7: Drug Content of Formulations EG1 - EG3

In Vitro Drug Release

The release of Mupirocin from the Emulgel was varied according to concentration of polymer. The release of the drugs from its emulsified gel formulation can be ranked in the following descending order: EG1 > EG2 > EG3.

Where the amounts of the drug released after 8 hours were 54.21%, 58.43%, 67.02%, respectively.

The progressive increase in the amount of drug diffusion through membrane from formulation attributed to gradual decrease in the concentration of polymer. It has been concluded that, if we increase the concentration of polymer, the diffusion of drug through the membrane also decreases. The cumulative % drug release profile of all the formulation batches has been shown in Table 4 and graph is plotted between cumulative % drug releases versus time as shown in graph 8.

Table 4: In-vitro drug release study of Mupirocin emulgel formulations

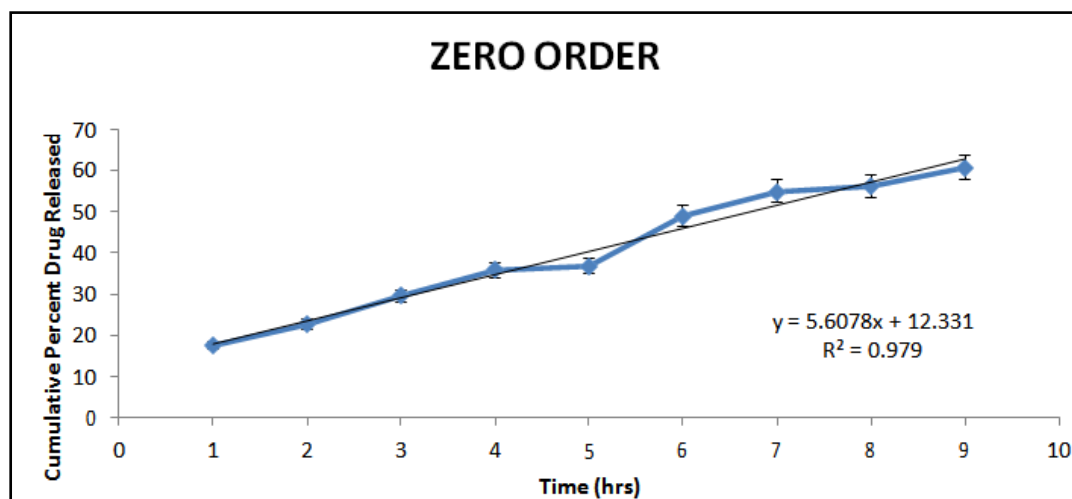
Time (hours)	EG1	EG2	EG3
0	0.00±0.00	0.00±0.00	0.00±0.00
0.5	11.98±1.77	15.44±1.25	14.68±0.015
1.0	16.36±1.51	20.85±0.31	19.19±0.005
1.5	20.61±1.96	24.83±0.05	22.53±0.02
2.0	23.22±0.55	27.81±1.46	27.39±0.003
2.5	26.68±0.56	31.25±2.24	32.11±0.36
3.0	28.46±2.43	35.23±0.57	36.04±0.23
3.5	30.88±2.02	37.66±1.98	38.93±0.47
4.0	34.77±2.67	40.08±1.11	44.27±0.15
4.5	37.42±1.90	42.17±2.87	47.45±0.89
5.0	40.27±0.32	43.39±0.91	52.19±0.20
5.5	43.76±1.86	46.19±1.32	57.21±0.115
6.0	45.48±1.61	47.56±0.65	58.64±0.65
6.5	47.14±0.34	50.85±1.81	59.74±0.11
7.0	50.12±0.56	53.65±1.90	62.09±1.18
7.5	52.20±1.43	56.39±2.49	64.32±0.32
8.0	54.21±0.33	58.43±1.09	67.02±1.04
8.5	54.92±0.79	60.41±0.26	69.32±2.09
9.0	58.56±0.53	61.9±2.65	72.56±0.42
9.5	59.88±0.7	63.06±0.32	74.48±0.01
10.0	61.84±0.83	64.48±0.51	76.18±2.46
10.5	63.2±2.22	66.55±2.54	77.54±2.06
11.0	69.6±1.48	70.33±1.86	78.7±0.69
11.5	69.8±1.27	71.04±0.74	79.14±2.13
12.0	69.91±1.16	71.65±0.65	80.47±1.27

Drug release Kinetic Study

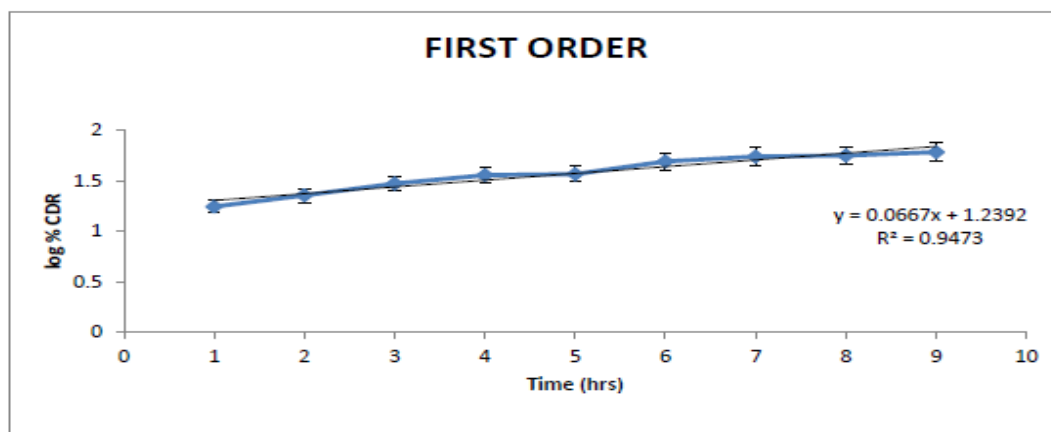
Raw data obtained from *in-vitro* release studies were analyzed, wherein data were fitted to different equations and kinetics model to calculate the percent drug release and release kinetics of Mupirocin from Emulgel.

Table 5: Drug Release Kinetic Data of Formulation EG3

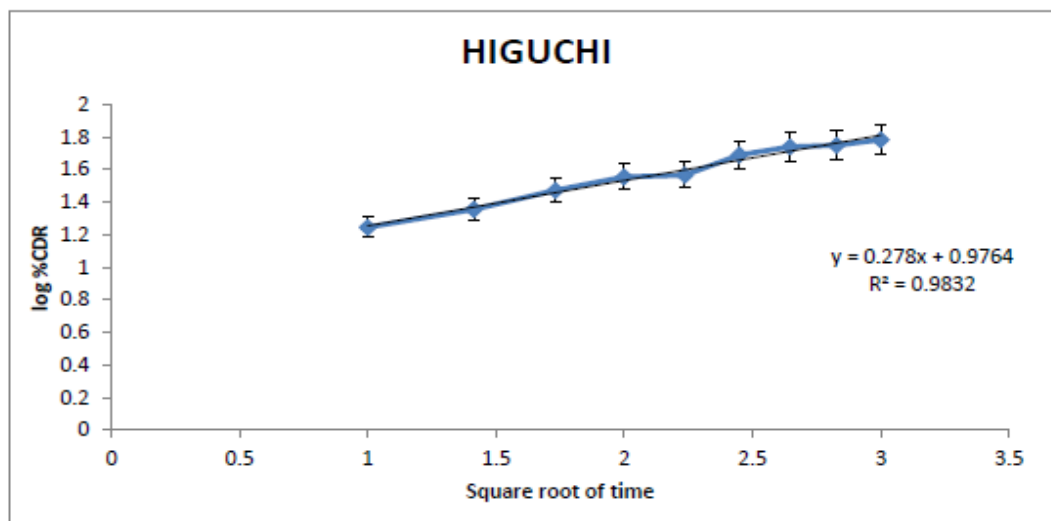
S.N.	Time (hrs)	Square root of time	Log time	Cumulative Percent Drug Release±SD	Log Cumulative Percent Drug Release	Cumulative Percent Drug Remaining	Log Cumulative Percent Drug Remaining
1	0	0	-	0	-	100	2.000
2	1	1	0	17.59±0.500	1.245399	82.41	1.915
3	2	1.414	0.301	22.66±0.438	1.355	77.34	1.888
4	3	1.732	0.477	29.64±0.681	1.471	70.36	1.847
5	4	2.0	0.602	35.87±1.022	1.554	64.13	1.807
6	5	2.236	0.698	36.82±0.520	1.566	63.18	1.800
7	6	2.449	0.778	48.99±0.521	1.690	51.01	1.707
8	7	2.645	0.845	54.87±0.563	1.739	45.13	1.654
9	8	2.828	0.903	56.15±0.691	1.749	43.85	1.641
10	9	3	0.954	60.70±0.522	1.783	39.30	1.594



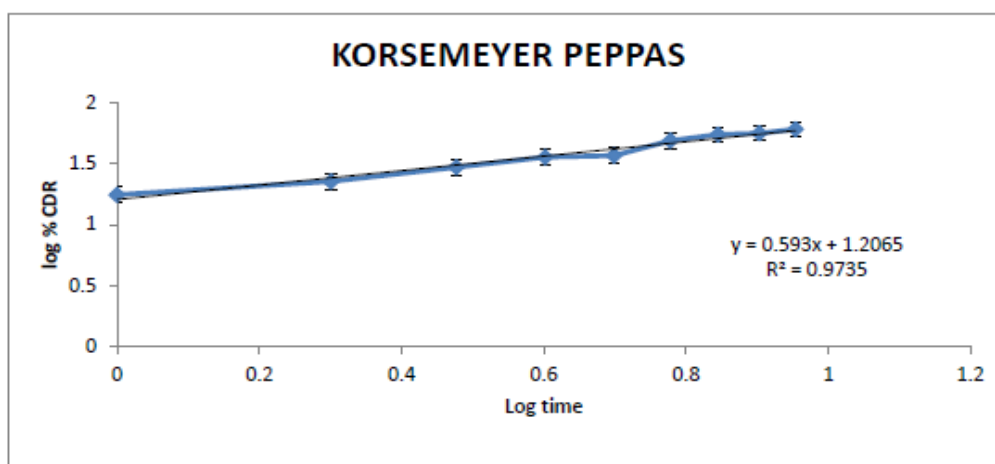
Graph 8: Cumulative Percent Drug Released Vs Time Plot (Zero Order)



Graph 9: Log Cumulative Percent Drug Remaining Vs Time Plot (First Order)



Graph 10: Log of cumulative Drug Release Vs Square Root of Time (Higuchi's Plot)



Graph 11: Log Cumulative Percent Drug Released Vs Log Time (Peppas Plot)

Linear regression analysis and model fitting shows that formulation EG3 follows Higuchi kinetics, which has higher value of correlation coefficient (R^2). The final results are reproduced in Table 5.11

Table 6: Regression Coefficient (R^2) Values of Drug Release Data obtained from various Kinetic Models

Formulation Code	Zero Order (R^2)	Higuchi Model (R^2)	First Order (R^2)	Peppas Model (R^2)
EG3	0.979	0.983	0.947	0.9735

Microbiological Assay

The use of control plates allowed that the plain emulgel bases were microbiologically inert toward the *staphylococcus aureus* strain. Percentage inhibition was taken as a measure of the drug's antimicrobial activity.

The highest activity was observed with EG3 where the percentage inhibition found to be $64.3 \pm 1.52\%$ while the lowest activity found with EG1 where the percentage of inhibition is 41.6

$\pm 0.57\%$ and the percentage inhibition of marketed mupirocin ointment was found to be 51.03 ± 1.23 which is less than the best (EG3) formulation.

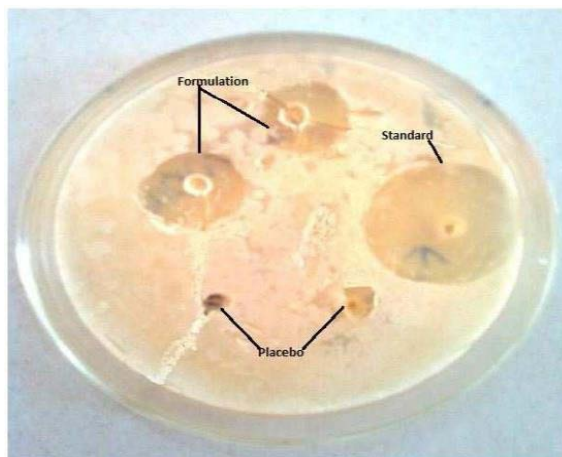


Fig.3: Zone of inhibition obtained with Formulation EG3



Fig.4: Showing % inhibition by marketed Mupirocin ointment

Accelerated Stability Studies

The optimised formulation was stored at $40 \pm 2^\circ\text{C}/75 \pm 5\% \text{RH}$ in lamitubes for 6 months. Emulgels were analysed at specified time intervals (1,2,3,4th weeks) for various parameters.

Table 7: Effect on Various Parameters during Accelerated Stability Studies of EG3

S. NO.	Time intervals(months)	pH	Consistency & Appearance	In vitro Drug Release (8 th hr)	% CDR
1	0	6.4	+++	67.02	66.70
2	1	6.3	+++	66.95	67.45
3	2	6.4	+++	67.79	68.23
4	3	6.5	+++	68.14	68.41
5	6	6.4	+++	68.52	69.63

There are no significant changes in the parameters during accelerated stability condition. Hence, Mupirocin Emulgel is stable at accelerated conditions.

CONCLUSION

After thorough literature survey and these extensive works, we reached into a conclusion that emulgels have proven as most convenient, better, and effective delivery system. Due to its non-greasy, gel-like property and better release of drugs as compared to other topical drug delivery system, it is proved to be one of the better delivery system.

Incorporation of emulsion into gel makes it a dual control release system further problem such as phase separation, creaming associated with emulsion gets resolved, and its stability improves. Emulgel loaded with specific drugs has been found effective in some topical disorders, and it is emerging as potential drug delivery system in the area of dermatology. In future, emulgel will provide a solution for topical delivery of hydrophobic drugs.

From the above results we can conclude that Mupirocin Emulgel formulations prepared with Carbomer 940, Flax seed oil, PEGs, Glycerin, *etc.* showed acceptable physical properties, drug release, and antimicrobial activity, which remained unchanged upon storage for 6 months. However, the Carbomer 940 based Emulgel in its low concentration with the formulation code EG3 proved to be the formula of choice, since it showed the highest drug release and very good antimicrobial activity when compared to the marketed Azithromycin gel. So, Mupirocin Emulgel can be used as an anti-microbial broad spectrum medication for topical drug delivery.

Many of drugs that have utility in the treatment of skin disorders are hydrophobic in nature. Such drugs can be delivered in the form of emulgel where they can be incorporated in the oil phase of the emulsion and combined with gel. Drugs which are still unexplored in this area are Retinoic acid, Adapalene, Tolnaftate, Betamethasone, Dexamethasone, *etc.*

REFERENCES

- 1) Aher, S.D., Banerjee, S.K., Gadhave, M.V., and Gaikawad, D.D. Emulgel: A new dosage form for topical drug delivery. *Int J Inst Pharm Life Sci* 2013; 3: 1-10.
- 2) Ajazuddin A., Alexander A., Khichariya A., Gupta S., and Patel R.J. Recent expansion in an emergent novel drug delivery technology: Emulgel. *J Controlled Release* 2013; 171:122-32.
- 3) Ashara K., and Shah K. Emulgel: A novel drug delivery system. *J Prev Alzheimer' Dis* 2016; 243-49.
- 4) Chain, E.B., and Mellows, G. Pseudomonic acid. Part 1. The structure of pseudomonic acid A, a novel antibiotic produced by *Pseudomonas fluorescens*. *J. Chem. Soc. Perkin Trans. 1*. 1977; 294-309.
- 5) Chien Y.W. Rate-control drug delivery systems: controlled release vs. sustained release, *Med Prog. Techn.* 1989; 21-46.
- 6) Dadwal, M. Emulgel: A novel approach to topical drug delivery. *Int J Pharm Sci* 2013; 4: 847-56.
- 7) El-Sayed, A.K., Hothersall, J., and Thomas, C.M. Quo-rum-sensing-dependent regulation of biosynthesis of the polyketide antibiotic mupirocin in *Pseudomonas fluorescens*. *Microbiology* 2001; 147, p. 2127-39.
- 8) Fuller, A.T., Mellows, G., Woodford, M., Banks, G.T., Barrow, K.D., and Chain, E.B. Pseudomonic acid: an antibiotic produced by *Pseudomonas fluorescens*. *Nature* 234. 1971;
- 9) Hardenia A., Jayronia S., Jain S. Emulgel: An emergent tool in topical drug delivery. *Int J Pharm Sci Res* 2014; 5: 1653-60.
- 10) Jain, A., Jain, S., Vyas, N., and Deveda, P. Development of antifungal emulsion based

- gel for topical fungal infection. *Int J PeriodontRestorat Dent* 2011; 2: 18- 23.
- 13) Joshi, B., Singh, G., Rana, A., and Saini, S. Development and characterization of clarithromycin emulgel for topical delivery. *Int J Drug Dev Res* 2012; 4: 310-23.
 - 14) Lademann, J., et al., Hair follicles—an efficient storage and penetration pathway for topically applied substances. *Skin pharmacology and physiology*, 2008.21. (3): p.150-155.
 - 15) Martin, F.M., and Simpson, T.J. Biosynthetic studies on pseudomonic acid (mupirocin), a novel antibiotic metabolite of *Pseudomonas fluorescens*. *J. Chem. Soc. Perkin Trans.* 1.1989; 207–209.
 - 16) McGrath, J., Eady, R., and Pope, F. Anatomy and organization of human skin.
 - 17) *Rook's textbook of dermatology*, 2004. 3: p. 1-15.
 - 18) Meguro, S., et al., Relationship between covalently bound ceramides and transepidermal water loss (TEWL). *Arch Dermatol Res*, 2000.292 (9): p. 463-8.
 - 19) Mulye, S., Wadkar, K., and Kondawar, M. Formulation development and evaluation of indomethacin emulgel. *Pelagia Res Der Pharm Sin* 2013; 4: 31-45.
 - 20) Odland, G.F. Structure of the skin. *Physiology, biochemistry, and molecular biology of the skin*, 1991.1: p. 3-62.
 - 21) Pant S., Badola A., Baluni S., and Pant W. A review on emulgel novel approach for topical drug delivery system. *World J Pharm PharmSci* 2015; 4: 1728-43.
 - 22) Parenti M.A., Hatfield S.M., and Leyden J.J. Mupirocin: a topical antibiotic with a unique structure and mechanism of action. *Clin Pharm.* 1987; 10: 761-70.
 - 23) Poovelikunnel, T., Gethin. G., and Humphreys. H. Mupirocin resistance: clinical implications and potential alternatives for the eradication of MRSA. *J AntimicrobChemother.* 2015; 70(10):2681-92.
 - 24) Prausnitz, M.R., et al., Skin barrier and transdermal drug delivery. *Dermatology*, 2012.3: p. 2065-2073.
 - 25) Proksch, E., Brandner, J.M., and Jensen, J.M. The skin: an indispensable barrier. *Experimental Dermatology*, 2008. 17 (12): p. 1063-1072.
 - 26) Putnam, C.D., and Reynolds, M.S. Mupirocin: a new topical therapy for impetigo.
 - 27) *J Pediatr Health Care.* 1989; 3(4): 224-27.
 - 28) Rieger, M.M., Lachman, L., Lieberman, H.A., and Kanig, J.L. *The Theory and Practice of Industrial Pharmacy*. 3rd ed. Philadelphia, PA: Lea and Febiger; 1986. p. 502-33.
 - 29) Sawant, A., and Mohit, S. Formulation and evaluation of itraconazole emulgel for topical drug delivery. *Asian J PharmacolToxicol* 2015; 5:91-96.
 - 30) Singla, V., Saini, S., Joshi, B., and Rana, A.C. Emulgel: A new platform for topical drug delivery. *Int J Pharm Sci* 2012; 3: 485-98.