“FORMULATION DEVELOPMENT AND EVALUATION OF EMULGEL FOR SOME TOPICAL THERAPEUTICS AGENTS”

Synopsis of the PhD Thesis
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1. **Title of the thesis**

Formulation development and evaluation of emulgel for some topical therapeutics agents.

2. **Abstract**

Emulgel have been identified as an alternative approach to conventional dosage forms. Emulgel for dermatological use have several favourable properties such as being thixotropic, greaseless, easily spreadable, easily removable, emollient, non-staining, compatible with several excipients, and water-soluble or miscible. The present investigation was undertaken with the objectives of formulating an emulgel of Apremilast, used in psoriasis, and Lornoxicam, used as an analgesic and anti-inflammatory. The present research reveals the effectiveness of the quality by design methodology to optimize the formulation variables for the development of Apremilast and Lornoxicam emulgel. Various oils, surfactants, co-surfactants and co-solvents were selected on the basis of a solubility study of the drug, and concentrations of surfactants and co-surfactants were selected on the basis of a pseudo-ternary phase diagram study. First, nanoemulsions were prepared by spontaneous emulsification methods using a systematic approach to the design of experiments and evaluated for various parameters like physical properties, clarity, phase separation, pH, droplet size, polydispersibility index, zeta potential, % drug content, and % in vitro drug diffusion. The optimised formulation was further converted into gel form using different grades and concentrations of carbomer polymer and evaluated for different parameters like pH, viscosity, spreadability, extrudability, % drug content, % drug diffusion, release kinetic study, skin permeation and skin retention, and animal study. Apremilast nanoemulgel showed good skin permeation without any skin irritation, as well as a show positive response in psoriasis induced mice. Therefore, a developed emulgel of Apremilast could be a good alternative formulation approach for the management and treatment of psoriasis, with benefits such as targeted drug delivery, consumer compliance, and convenience. The optimised formulation of Lornoxicam nanoemulgel shows good in vitro diffusion as well as no skin irritation. Also, the optimised formulation showed good anti-inflammatory and analgesic activity as compared to plain gel. In both formulations, no significant change in the appearance, pH, viscosity, spreadability, extrudability, % drug content and % in vitro drug diffusion was observed during storage for 90 days.
3. Introduction

Topical drug delivery is the most common route of drug administration for the general population. Human skin is a uniquely engineered organ that permits terrestrial life by regulating heat and water loss from the body while preventing the ingress of noxious chemicals or microorganisms. It is also the largest organ of the human body, providing around 10% of the body mass of an average person, and it covers an average area of 1.7 m². (1, 2) It is an easier alternative to oral medicine that is useful for local delivery of agents, particularly those that have toxic effects if administered systemically, with benefits of easy termination of medications when needed, fewer risks of abuse, better patient compliance, and an effective cost for the pharmaceutical companies. (3) Drawbacks of various topical dosage forms, such as in cream phase inversion and breaking, in ointment rancidity due to an oily base, lump formation due to hygroscopicity in powder, liniment that requires rubbing at the affected area, and other issues, such as being very sticky and uncomfortable for the patient and having less spreadability. (4) Within the major group of semisolid preparations, the use of transparent gels has expanded widely, both in cosmetics and in pharmaceutical preparations. The USP defines gels as semisolid systems containing either suspensions made up of either small inorganic particles or large organic molecules interpenetrated by a liquid. Gel forms a cross-linked network where it captures small drug particles and provides their release in a controlled manner. It extends the contact period of medication over the skin due to its mucoadhesive property. (5) Within biphasic liquid dosage forms, emulsion is a controlled release system where entrapped drug particles in the internal phase pass through the external phase to the skin and slowly get absorbed. Internal phases act as drug reservoirs, slowly releasing drug through the external phase to the skin. (6) In spite of the many advantages of gels and emulsions, a major limitation in gel is their inability to deliver hydrophobic drugs and in emulsion instability during storage. To overcome these limitations, an emulsion-based approach, i.e., emulgel, is used, allowing a hydrophobic therapeutic moieties to be successfully incorporated while enjoying the unique properties of gels. (7)

Apremilast (APM), is a phosphodiesterase 4 (PDE4) inhibitor used to treat various types of symptoms resulting from certain inflammatory autoimmune diseases. Initially approved in 2014, it is marketed by Celgene. In July 2019, APM was granted a new FDA approval for the treatment of oral ulcers associated with Bechet’s disease, an autoimmune condition that causes recurrent skin, blood vessel, and central nervous system inflammation. (8) APM is indicated for the treatment of active psoriatic arthritis in adults and for the treatment of active moderate-to-severe psoriatic arthritis in patients who are eligible for phototherapy and systemic
treatment. (9) APM belongs to class IV drugs as per BCS classification, so improve its solubility and permeability by formulating nanoemulgel.

Lornoxicam (LOR) is a new nonsteroidal anti-inflammatory drug (NSAID) of the oxicams class with analgesic, anti-inflammatory, and antipyretic properties. LOR differs from other oxicams compounds in its potent inhibition of prostaglandin biosynthesis, a property that explains the drug’s particularly pronounced efficacy. LOR is used for the treatment of various types of pain, especially that resulting from inflammatory diseases of joints, osteoarthritis, surgery, sciatica, and rheumatoid arthritis. (10)

3.1 Brief description on the state of the art of the research topic

Emulgel has the capability of delivering both hydrophilic and hydrophobic drug moieties due to the presence of both aqueous and non-aqueous phases. Emulgel itself is a controlled release system where entrapped drug particles in the internal phase pass through the external phases, act as a reservoir of drug, and slowly release drug in a controlled way through the external phase to the skin. Due to its bioadhesive property, it prolongs the contact period of medication with the skin. Since emulgel possesses the properties of both emulsion and gel, it acts as a dual-control release system. (2) Emulgels are a class of biphasic semisolid formulations. They are now being used for controlled delivery applications. It is suitably applied to the skin due to its non-greasy nature in comparison to other topical formulations such as ointments, creams, etc., which are very thick and require excess rubbing. (11) It is accepted that the utility of any topical preparation lies in its penetration ability, which refers to the disappearance of product or oiliness from the skin. Penetration into the skin is facilitated if the emulsion is thixotropic, that is, if it becomes less viscous during shearing. Thus, to improve emulsion stability and penetration ability, it is incorporated into gel. Furthermore, dermatological emulgels have several advantageous properties, including being thixotropic, having a longer shelf life, being bio friendly, and having a clear and pleasant appearance. (12)

3.2 The ideal features of Emulgel (2, 13)

- Easily spreadable
- Ease of handling and administration
- Easily removable
- Non staining
- Pleasing appearance
- Being greaseless
- Emollient

3.3 Advantages of Emulgel (12-14)
Incorporation of hydrophobic drugs
Better drug loading capacity
Better stability.
Production feasibility and low preparation cost
More patient compliance
No first-pass effect
Dual-controlled release of drug from emulsion and gel
From the market perspective, new drug delivery technologies offer the opportunity to extend revenue life cycles for pharmaceutical companies whose drug patents are about to expire and will soon be vulnerable to generic competition. Moreover, the grant of marketing exclusivity to the new dosage form would help to increase revenue.

4. Definition of the problem
Many widely used topical agents such as ointment, cream, and lotion have many disadvantages. They are very sticky and cause uneasiness in the patient when applied. Moreover, they also have a lower spreading coefficient and need to be applied by rubbing. They also exhibit the problem of stability. Due to all these factors within the major group of semisolid preparations, the use of transparent gels has expanded both in cosmetics and pharmaceutical preparations. A gel is a colloidal substance that is typically 99% liquid and is immobilised by surface tension and a macromolecular network of fibres built from a small amount of gelatin present. (15) In spite of the many advantages of gels, a major limitation is the delivery of hydrophobic drugs. Hence, to overcome this limitation, an emulsion-based approach is being used so that even a hydrophobic therapeutic moiety can be successfully incorporated and delivered through gels. (16)

APM is primarily used to treat psoriasis and other skin diseases, it also has anti-inflammatory properties at low doses. APM have low solubility and permeability; therefore, improve their solubility and permeability, develop a nanoemulgel topical formulation. APM is also an excellent candidate for emulgel formulation due to its low molecular weight (460.5 g/mol), low water solubility (0.0503 mg/ml), high log P value (1.79), small molecular size, non-irritant, and non-sensitizing properties. APM topical formulation also reduces the side effects, like worsening depression and weight loss. (17)

LOR is a highly potent NSAID belonging to the chemical class of oxicams. It is used in the treatment of mild to moderate pain, as well as rheumatoid arthritis and osteoarthritis. Because of its low molecular weight of 371.82 g/mol, low water solubility of 0.0155 mg/ml, high Log P value of 2.62, small molecular size, and non-irritating and non-sensitizing properties, LOR
is an ideal candidate for emulgel formulation. Also, LOR is 100 times more potent than tenoxicam as a COX inhibitor. LOR's analgesic potency is 12 and 10 times greater than that of piroxicam and tenoxicam, respectively. LOR, Meloxicam, and Piroxicam produce the same anti-inflammatory effect, but only LOR is effective for the prevention of hyperalgesia. (18) So the aim of this study to develop and evaluate a topical formulation of APM and LOR in emulgel to overcome side effects and improve patient compliance.

5. Objective and scope of work

The aim of the present study to formulate and evaluate nanoemulgels of APM and LOR.

Specific Objectives are as follows

- Preformulation study for identification of drugs by melting point, FTIR, and UV maxima
- Standard calibration curve for pure drug in methanol and pH 6.8 phosphate buffer
- Solubility study of drugs in different oils, surfactants, co-surfactants, and co-solvents
- Preparation of a pseudo-ternary phase diagram for selecting the concentration ratio of surfactant : cosurfactant
- Compatibility studies between the drug and excipients by FTIR
- To optimise the formulation through experimental design
- The formulated emulsion was evaluated for various parameters such as optical transparency, clarity, pH, centrifugation, droplet size, zeta potential, drug content, and % in vitro diffusion study, and so on.
- Statistical analysis of all the results and selection of the optimised batch for conversion into gel form
- Formulation of gel from a selection of different grades and concentrations of carbomer polymers.
- Evaluate the prepared emulgel for different parameters like appearance, pH, viscosity, spreadability, extrudability, swelling index, drug content, and % in vitro diffusion study.
- Evaluate the optimised batch for kinetic studies, ex vivo drug permeation and retention studies, skin irritation tests, and the effect of the drug on animals.
- To conduct stability studies as per the ICH guidelines.

Scope of the research work:

The main aim of topical formulations of emulgel is to fulfil targeted drug delivery and give effect directly on the affected part, which may also contribute to a reduction of the side effects observed after oral administration. Incorporation of emulsion into gel makes it a dual-control
release system; further problems such as phase separation and creaming associated with emulsion get resolved, and its stability improves. APM and LOR applied in the form of emulgel will be a potential dosage form for the general population by providing topical delivery of a hydrophobic drug and better patient compliance.

By developing an effective delivery system for the existing drug, Emulgels offer the opportunity to extend the product life cycle for pharmaceutical companies whose drug patents are about to expire and will soon be vulnerable to generic competition.

6. **Original contribution by the thesis:**

The entire work in this synopsis, as well as the thesis, is original. An extensive literature review was done to identify the challenges associated with the conventional dosage form for a special group of people and approaches that can resolve them. Despite the fact that many researchers have worked on emulgel development, the idea of developing emulgels of APM and LOR drugs through a systematic approach to the design of experiments for optimization of various parameters and to target increasing solubility and drug diffusion remains.

7. **Methodology of research, Results / Comparisons:**

7.1 **Design of the Experiment**

A literature review was conducted in order to identify and determine the quality target product profile (QTPP), critical quality attributes (CQA), methods for emulgel formulation, and various process and formulation attributes influencing product CQA. The QbD method was used for the investigations. Different preliminary experiments were performed for the selection of suitable excipients or polymers that may directly or indirectly influence critical quality attributes. (19) The compatibility of the drug excipients was checked before the optimization of various process and formulation variables. With the selected excipients and polymers, various critical process variables identified through literature were optimised sequentially as per the steps involved in the formulation. (20) Finally, the formulation variables were optimised using a suitable factorial design and Design Expert® software for analysing the data statistically and graphically using response surface plots.

7.2 **Analytical Methods**

**UV-spectrophotometric method**

UV spectrophotometric methods have been reported for the estimation of APM and LOR in formulations. Calibration curves were plotted in methanol and phosphate buffer pH 6.8 as solvents for the determination of APM and LOR in formulation using the UV-1800 spectrophotometer (Shimadzu) at 230 nm and 380 nm, respectively. (21, 22) In APM, linearity was observed in the concentration range of 1–10 μg/ml with a regression coefficient value of
0.9983 in methanol and 0.9989 in a pH 6.8 phosphate buffer. Linearity was observed in LOR at the concentration range of 1–20 μg/ml with a regression coefficient value of 0.9923 in methanol and 0.9960 in a pH 6.8 phosphate buffer.

### 7.3. Selection of Excipients:
Drug release from emulsion-based delivery systems is regulated and influenced by the drugs’ partitioning from the lipid phase into the aqueous phase via interactions with surfactant and co-surfactant molecules. That’s why it’s so important to pick the appropriate lipids and stabilising surfactants. In addition, the excipient selection is based on the maximal loading of poorly soluble medication in the formulation; therefore, the excipients with the highest solubility for the lipophilic drug are selected. (23, 24) APM showed the highest solubility in Captex 355 EP/NF, which was chosen as the oil, Cremophore RH 40 as the surfactant, Labrafil as the co-surfactant, and Propylene Glycol as the co-solvent based on the solubility study. LOR showed the highest solubility in Captex 200, which was chosen as the oil, Cremophore RH 40 as the surfactant, Capmul MCM C8 as the co-surfactant, and Propylene glycol as the co-solvent based on the solubility study.

### 7.4 Selection of Smix ratio
Several combinations of surfactant and co-surfactant (Smix) (1:1, 1:2, 1:3, 2:1, and 3:1) were investigated based on the rising concentration of surfactant in relation to co-surfactant and the rising concentration of co-surfactant in relation to surfactant. When the oil concentration and Smix ratio were increased from 1:9 to 9:1, mixtures were titrated gradually with small amounts of water. Titration samples were shaken to ensure consistency and examined visually to determine how well they emulsified. (25) Thus, the optimal microemulsion of APM was obtained by following the pseudo-ternary phase diagram, which involved a 3:1 surfactant: co-surfactant ratio. Thus, the optimal microemulsion of LOR was obtained by following the pseudo-ternary phase diagram, which involved a 1:1, 2:1, and 3:1 surfactant: co-surfactant ratio.

### 7.5 Formulation development and optimization of APM emulgel
The objective of this study is to formulate a stable emulsion with a small droplet size and higher drug diffusion. To achieve this objective, the contribution of independent formulation variables was examined using an experimental design.

The formulation of an APM nanoemulsion (NE) was optimised using a simplex lattice design. The simplex lattice design is a three-component system that is represented by an equilateral triangle in two-dimensional spaces. In this design, three factors were assessed by altering their concentrations simultaneously while keeping their total concentration constant. For
formulating an APM emulsion, select oil, Smix, and water as independent factors and droplet size and % in vitro drug diffusion as dependent variables. Seven batches were created in total: three at vertexes (A, B, and C), three at halfway points between vertexes (AB, AC, and BC), and one at the midpoint (ABC). (26)

7.5.1 Apremilast emulsion preparation:

APM emulsion prepared using the spontaneous emulsification method and evaluated for various parameters, as shown in Table: 1.

Table: 1 Evaluation result of prepared APM nanoemulsion using experiment design

<table>
<thead>
<tr>
<th>Formula</th>
<th>pH</th>
<th>% Transmittance</th>
<th>Droplet size(nm)</th>
<th>PDI</th>
<th>Zeta potential (mv)</th>
<th>% Drug Content</th>
<th>% drug diffusion</th>
</tr>
</thead>
<tbody>
<tr>
<td>A1</td>
<td>5.85 ± 0.04</td>
<td>98.54 ± 0.06</td>
<td>59.86 ± 0.04</td>
<td>0.199 ± 0.04</td>
<td>-8.49 ± 0.06</td>
<td>98.96 ± 0.05</td>
<td>89.21 ± 0.08</td>
</tr>
<tr>
<td>A2</td>
<td>5.23 ± 0.06</td>
<td>92.34 ± 0.09</td>
<td>111.4 ± 0.06</td>
<td>0.252 ± 0.06</td>
<td>-10.1 ± 0.04</td>
<td>98.03 ± 0.08</td>
<td>78.41 ± 0.07</td>
</tr>
<tr>
<td>A3</td>
<td>5.34 ± 0.08</td>
<td>94.59 ± 0.06</td>
<td>95.87 ± 0.07</td>
<td>0.297 ± 0.05</td>
<td>-10.3 ± 0.05</td>
<td>99.50 ± 0.06</td>
<td>86.11 ± 0.03</td>
</tr>
<tr>
<td>A4</td>
<td>5.59 ± 0.04</td>
<td>97.98 ± 0.06</td>
<td>72.01 ± 0.05</td>
<td>0.155 ± 0.04</td>
<td>-6.99 ± 0.07</td>
<td>98.81 ± 0.04</td>
<td>88.24 ± 0.04</td>
</tr>
<tr>
<td>A5</td>
<td>5.76 ± 0.05</td>
<td>98.94 ± 0.07</td>
<td>53.19 ± 0.07</td>
<td>0.261 ± 0.07</td>
<td>-9.64 ± 0.06</td>
<td>99.23 ± 0.04</td>
<td>94.87 ± 0.05</td>
</tr>
<tr>
<td>A6</td>
<td>5.19 ± 0.04</td>
<td>98.01 ± 0.05</td>
<td>67.46 ± 0.04</td>
<td>0.174 ± 0.07</td>
<td>-8.85 ± 0.04</td>
<td>99.27 ± 0.08</td>
<td>84.54 ± 0.06</td>
</tr>
<tr>
<td>A7</td>
<td>5.25 ± 0.04</td>
<td>84.32 ± 0.08</td>
<td>552.5 ± 0.06</td>
<td>1 ± 0.05</td>
<td>-10.3 ± 0.05</td>
<td>98.52 ± 0.04</td>
<td>74.35 ± 0.08</td>
</tr>
</tbody>
</table>

Simple lattice design was employed for optimization of formulation variables, and Design Expert® 13 software was used for statistical analysis by ANOVA, generating ease to construct contour plots and 3D surface plots for each response.

Factorial plot of Droplet size of APM:

Figure: 1 Surface Response Plot & 3D Surface Plot of droplet size of APM

Full model equation for droplet size: 47.63 + 62815.2 X1 + 198.64 X2 + 1438.29 X3 – 75163.2 X1X2 – 84229.2 X1X3 + 951.732 X2X3

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Simple lattice design analysis showed that the coefficients $b_1$, $b_2$, and $b_3$ showed a positive sign for $X_1$, $X_2$, and $X_3$, so it indicated that as the amount of that increased, the droplet size of the formulation also increased. In this equation, find the highest value of factor $X_1$ as compared to $X_2$ and $X_3$. So, it indicate oil concentration increase give more effect on droplet size as compare to other. The coefficient $b_{23}$ has more than a 0.05 $P$ value, so this is omitted from the full model.

**Factorial plots for diffusion rate of APM:**

![Factorial plots for diffusion rate of APM](image)

**Figure: 2 Surface Response Plot & 3D Surface Plot of diffusion rate of APM**

Full model equation for diffusion rate: $78.66 + 69.01X_1 + 89.91X_2 + 82.29X_3$

Simple lattice design analysis revealed that the coefficients $b_1$, $b_2$, and $b_3$ have a positive sign for $X_1$, $X_2$, and $X_3$, implying that as the amount of the increase increases, so does the diffusion rate of the formulation. Find the highest value of factor $X_2$ in this equation compared to the other factors, indicating that an increase in SMIX concentration has a greater effect on increasing diffusion than oil.

**Experimental validation of design space:**

Experimental validation of DoE trials for formulation variables was undertaken through the formulation and evaluation of gel at the checkpoint batch suggested by the software. The prediction power of the model was validated by comparing the predicted value to the observed or experimental value for each CQA listed in Table 2. The press value was calculated for each CQA, and it was found to be less than ± 5%, which confirms the prediction power of the applied design.

**Table: 2 Check point batch preparation**

<table>
<thead>
<tr>
<th>Response</th>
<th>Check point batch A8</th>
<th>Check point batch A9</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Predicted</td>
<td>Observed</td>
</tr>
<tr>
<td>Droplet size (nm)</td>
<td>36.75</td>
<td>37.64</td>
</tr>
</tbody>
</table>
7.5.2 Conversion of APM nanoemulsion to nanoemulgel:

Optimized NE formulation incorporated into gel matrix prepared in 1:1 NE gel matrix using carbopol (934, 940, or 980) polymer formulation composition shown in Table 3 and evaluate for different parameter and result shown in Table-4.

Table: 3 Formulation Composition of APM Nanoemulgel.

<table>
<thead>
<tr>
<th>Ingredients (%)</th>
<th>C1</th>
<th>C2</th>
<th>C3</th>
<th>C4</th>
<th>C5</th>
<th>C6</th>
<th>C7</th>
<th>C8</th>
<th>C9</th>
</tr>
</thead>
<tbody>
<tr>
<td>Apremilast</td>
<td>0.5</td>
<td>0.5</td>
<td>0.5</td>
<td>0.5</td>
<td>0.5</td>
<td>0.5</td>
<td>0.5</td>
<td>0.5</td>
<td>0.5</td>
</tr>
<tr>
<td>Captex 355</td>
<td>10</td>
<td>10</td>
<td>10</td>
<td>10</td>
<td>10</td>
<td>10</td>
<td>10</td>
<td>10</td>
<td>10</td>
</tr>
<tr>
<td>S mix</td>
<td>35</td>
<td>35</td>
<td>35</td>
<td>35</td>
<td>35</td>
<td>35</td>
<td>35</td>
<td>35</td>
<td>35</td>
</tr>
<tr>
<td>Propylene Glycol</td>
<td>0.5</td>
<td>0.5</td>
<td>0.5</td>
<td>0.5</td>
<td>0.5</td>
<td>0.5</td>
<td>0.5</td>
<td>0.5</td>
<td>0.5</td>
</tr>
<tr>
<td>Methyl Paraben</td>
<td>0.18</td>
<td>0.18</td>
<td>0.18</td>
<td>0.18</td>
<td>0.18</td>
<td>0.18</td>
<td>0.18</td>
<td>0.18</td>
<td>0.18</td>
</tr>
<tr>
<td>Propyl Paraben</td>
<td>0.02</td>
<td>0.02</td>
<td>0.02</td>
<td>0.02</td>
<td>0.02</td>
<td>0.02</td>
<td>0.02</td>
<td>0.02</td>
<td>0.02</td>
</tr>
<tr>
<td>Carbopol 934</td>
<td>0.5</td>
<td>0.75</td>
<td>1.0</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td></td>
</tr>
<tr>
<td>Carbopol 940</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>0.5</td>
<td>0.75</td>
<td>1.0</td>
<td>-</td>
<td>-</td>
<td></td>
</tr>
<tr>
<td>Carbopol 980</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>0.5</td>
<td>0.75</td>
<td>1.0</td>
<td></td>
</tr>
<tr>
<td>Water</td>
<td>q.s.</td>
<td>q.s.</td>
<td>q.s.</td>
<td>q.s.</td>
<td>q.s.</td>
<td>q.s.</td>
<td>q.s.</td>
<td>q.s.</td>
<td>q.s.</td>
</tr>
</tbody>
</table>

(Smix containing cremophore RH40 and labrafil in 3:1)

Table: 4 Evaluation result of APM nanoemulgel

<table>
<thead>
<tr>
<th>Formula</th>
<th>pH</th>
<th>Viscosity</th>
<th>Spreadability</th>
<th>Extrudability</th>
<th>% Drug Content</th>
<th>% Drug Diffusion</th>
</tr>
</thead>
<tbody>
<tr>
<td>C1</td>
<td>5.12 ± 0.04</td>
<td>12100 ± 0.02</td>
<td>35.33 ± 0.02</td>
<td>13.65 ± 0.03</td>
<td>98.30 ± 0.06</td>
<td>83.57 ± 0.08</td>
</tr>
<tr>
<td>C2</td>
<td>5.70 ± 0.03</td>
<td>13600 ± 0.04</td>
<td>32.33 ± 0.04</td>
<td>15.41 ± 0.02</td>
<td>99.25 ± 0.07</td>
<td>81.34 ± 0.07</td>
</tr>
<tr>
<td>C3</td>
<td>5.94 ± 0.04</td>
<td>14967 ± 0.03</td>
<td>27.33 ± 0.03</td>
<td>18.48 ± 0.05</td>
<td>98.14 ± 0.05</td>
<td>76.26 ± 0.06</td>
</tr>
<tr>
<td>C4</td>
<td>5.28 ± 0.05</td>
<td>17100 ± 0.02</td>
<td>32.67 ± 0.01</td>
<td>16.28 ± 0.02</td>
<td>98.37 ± 0.04</td>
<td>86.34 ± 0.05</td>
</tr>
<tr>
<td>C5</td>
<td>5.49 ± 0.02</td>
<td>19300 ± 0.05</td>
<td>31.33 ± 0.05</td>
<td>18.92 ± 0.03</td>
<td>99.15 ± 0.03</td>
<td>92.46 ± 0.03</td>
</tr>
<tr>
<td>C6</td>
<td>5.79 ± 0.06</td>
<td>22267 ± 0.06</td>
<td>27.67 ± 0.02</td>
<td>22.48 ± 0.04</td>
<td>99.93 ± 0.05</td>
<td>81.35 ± 0.04</td>
</tr>
<tr>
<td>C7</td>
<td>5.62 ± 0.05</td>
<td>21100 ± 0.02</td>
<td>28.33 ± 0.04</td>
<td>20.62 ± 0.01</td>
<td>98.32 ± 0.07</td>
<td>80.51 ± 0.07</td>
</tr>
<tr>
<td>C8</td>
<td>5.81 ± 0.01</td>
<td>23367 ± 0.04</td>
<td>26.67 ± 0.03</td>
<td>22.64 ± 0.04</td>
<td>99.64 ± 0.06</td>
<td>77.08 ± 0.08</td>
</tr>
<tr>
<td>C9</td>
<td>5.98 ± 0.02</td>
<td>26400 ± 0.05</td>
<td>24.67 ± 0.02</td>
<td>24.60 ± 0.05</td>
<td>97.64 ± 0.04</td>
<td>75.58 ± 0.06</td>
</tr>
</tbody>
</table>
7.5.3 Evaluation of optimization formulation for different parameter
From all the emulgel formulations, select one optimised formulation for further study based on the formulation having a good physical appearance, good spreadability, good extrudability, and the highest in vitro diffusion. Batch C5 was selected as an optimised batch and further tested for a release kinetic study, skin permeation and skin retention, a skin irritation test, the effect of APM-loaded gel on psoriasis-induced mice, and a stability study. From the release kinetic study, we conclude that batch C5 follows the Higuchi model (diffusion-controlled release) and that the Korsmeyer-Peppas model is the most fitted model with the highest $R^2$ value. The data from the Korsmeyer-Peppas model revealed that the (n) values of 0.9148 were in the range of 0.5 to 1, implying that the medication was released via a non-fickian (anomalous) mechanism (a combination of erosion and diffusion). The amount of drug permeated through the skin in 8 hours in optimised batch C5 was 40.25%, with drug retention in the skin at 41.4% and remaining on the skin at 18.35%. The animal protocol was approved by the Arihant School of Pharmacy and Bioresearch Institution’s animal ethics committee (ASPBR/IAEC/2020-21/02) for animal study. The skin irritation study on rats, which found no evidence of skin irritation. The effect of APM-loaded gel on psoriasis-induced mice was studied, which showed a positive response. Stability studies conducted as per the ICH guidelines found the product to be physically and chemically stable, as no significant difference was observed.

7.6 Formulation development and optimization of Loroxicam emulgel
LOR nanoemulsion formulation was optimised by using $3^2$ full factorial designs; batches were prepared using two factors, each at three levels, and experimental trials were performed at all nine possible combinations. The ratio of oil to Smix (1:9, 2:8, and 3:7) and the ratio of surfactant to co-surfactant (1:1, 2:1, and 3:1) were selected as independent variables at three different levels. (27)

7.6.1 LOR emulsion preparation:
LOR emulsion was prepared using the spontaneous emulsification method and evaluated for various parameters, as shown in Table 5.

Table 5: Evaluation result of prepared LOR nanoemulsion using experiment design

<table>
<thead>
<tr>
<th>Formula</th>
<th>pH</th>
<th>Transmitance</th>
<th>Droplet size(nm)</th>
<th>PDI</th>
<th>Zeta potential (mv)</th>
<th>%Drug Content</th>
<th>% drug diffusion</th>
</tr>
</thead>
<tbody>
<tr>
<td>L1</td>
<td>5.85 ±</td>
<td>94.83 ±</td>
<td>40.23 ±</td>
<td>0.593 ±</td>
<td>-3.72 ±</td>
<td>99.24 ±</td>
<td>87.56 ±</td>
</tr>
<tr>
<td></td>
<td>0.04</td>
<td>0.06</td>
<td>0.05</td>
<td>0.07</td>
<td>0.04</td>
<td>0.07</td>
<td>0.08</td>
</tr>
<tr>
<td>L2</td>
<td>5.23 ±</td>
<td>98.92 ±</td>
<td>26.54 ±</td>
<td>0.296 ±</td>
<td>-6.01 ±</td>
<td>98.80 ±</td>
<td>90.21 ±</td>
</tr>
<tr>
<td></td>
<td>0.06</td>
<td>0.05</td>
<td>0.06</td>
<td>0.08</td>
<td>0.05</td>
<td>0.06</td>
<td>0.07</td>
</tr>
</tbody>
</table>
3² factorial design was employed for optimization of formulation variables, and Design Expert® 13 software was used for statistical analysis by ANOVA, generating ease to construct contour plots and 3D surface plots for each response.

**Factorial plot of Droplet size of LOR:**

![3D Surface Response Plot & 3D Surface Plot of droplet size of LOR](image)

**Full droplet size model equation:**

\[ 29.93 + 64.29 X1 + 5.88 X2 + 12.34 X1 X2 + 67.14 X1^2 - 0.65 X2^2 \]

3² Factorial design analysis showed that the coefficients b1 and b2 bear a positive sign for the coefficient value of X1 and X2. So it indicated that both X1 and X2 had a positive effect on droplet size, as a result of which the droplet size of the formulation also increased. In this equation, find the highest value of factor X1 in comparison to X2, indicating that increasing the oil to Smix concentration has a greater effect on droplet size than increasing X2. The coefficients b2 and b22 have more than a 0.05 p value, so they are omitted from the full model.

**Factorial plot of Diffusion rate of LOR:**

<table>
<thead>
<tr>
<th>L3</th>
<th>5.34 ± 0.08</th>
<th>96.79 ± 0.05</th>
<th>30.24 ± 0.04</th>
<th>0.322 ± 0.06</th>
<th>-7.32 ± 0.07</th>
<th>99.67 ± 0.07</th>
<th>85.11 ± 0.03</th>
</tr>
</thead>
<tbody>
<tr>
<td>L4</td>
<td>5.59 ± 0.04</td>
<td>98.33 ± 0.06</td>
<td>31.34 ± 0.03</td>
<td>0.259 ± 0.09</td>
<td>-3.06 ± 0.06</td>
<td>98.59 ± 0.06</td>
<td>91.18 ± 0.04</td>
</tr>
<tr>
<td>L5</td>
<td>5.76 ± 0.05</td>
<td>98.83 ± 0.06</td>
<td>25.62 ± 0.05</td>
<td>0.289 ± 0.03</td>
<td>-9.32 ± 0.03</td>
<td>99.61 ± 0.04</td>
<td>95.65 ± 0.05</td>
</tr>
<tr>
<td>L6</td>
<td>5.19 ± 0.04</td>
<td>97.63 ± 0.02</td>
<td>31.52 ± 0.07</td>
<td>0.164 ± 0.05</td>
<td>-9.11 ± 0.04</td>
<td>99.49 ± 0.07</td>
<td>89.24 ± 0.06</td>
</tr>
<tr>
<td>L7</td>
<td>5.25 ± 0.04</td>
<td>90.32 ± 0.09</td>
<td>138.6 ± 0.06</td>
<td>0.262 ± 0.06</td>
<td>-6.77 ± 0.05</td>
<td>98.38 ± 0.06</td>
<td>81.24 ± 0.08</td>
</tr>
<tr>
<td>L8</td>
<td>5.69 ± 0.05</td>
<td>87.53 ± 0.05</td>
<td>166.2 ± 0.04</td>
<td>0.204 ± 0.07</td>
<td>-6.49 ± 0.07</td>
<td>99.29 ± 0.06</td>
<td>78.41 ± 0.06</td>
</tr>
<tr>
<td>L9</td>
<td>5.53 ± 0.06</td>
<td>85.60 ± 0.08</td>
<td>178.0 ± 0.05</td>
<td>0.251 ± 0.04</td>
<td>-6.60 ± 0.05</td>
<td>99.08 ± 0.02</td>
<td>74.35 ± 0.05</td>
</tr>
</tbody>
</table>
Figure: 4 Surface Response Plot & 3D Surface Plot of diffusion rate of LOR

Full diffusion rate model equation: 93.90 – 6.42 X1 + 1.02X2 + 0.80X1X2 - 9.77 X1² – 4.84 X2²

3² Factorial design analysis showed that the coefficients b1 show negative sign for coefficient value for X1 and b2 bear a positive sign for coefficient value for X2. So it indicated that as oil : Smix conc. increased, the percentage of drug diffusion decreased, but as Smix concentration increased, the percentage of drug diffusion increased. The coefficient b12 have more than 0.05 p value so this omitted from the full model.

Experimental validation of design space:

Formulation and evaluation of gel at the checkpoint batch suggested by the software in Table 6 were used to conduct experimental validation of DoE trials for formulation variables.

Table: 6 Check point batch preparation

<table>
<thead>
<tr>
<th>Response</th>
<th>Check point batch L10</th>
<th></th>
<th></th>
<th></th>
<th></th>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>Droplet size (nm)</td>
<td>Predicted</td>
<td>Observed</td>
<td>Difference</td>
<td>% bias</td>
<td>Predicted</td>
<td>Observed</td>
<td>Difference</td>
</tr>
<tr>
<td>% Drug Diffusion</td>
<td>92.92</td>
<td>92.46</td>
<td>0.46</td>
<td>0.49</td>
<td>92.06</td>
<td>91.54</td>
<td>0.52</td>
</tr>
</tbody>
</table>

7.6.2 Conversion of Lornoxicam Nanoemulsion to Nanoemulgel:

Optimized NE formulation incorporated into a 1:1 NE gel matrix prepared with Carbopol (934, 940, or 980) polymer formulation composition shown in Table 7 and evaluated for various parameters and results shown in Table 8.

Table: 7 Formulation Composition of LOR Nanoemulgel.

<table>
<thead>
<tr>
<th>Ingredients (%)</th>
<th>F1</th>
<th>F2</th>
<th>F3</th>
<th>F4</th>
<th>F5</th>
<th>F6</th>
<th>F7</th>
<th>F8</th>
<th>F9</th>
</tr>
</thead>
<tbody>
<tr>
<td>Lornoxicam</td>
<td>0.5</td>
<td>0.5</td>
<td>0.5</td>
<td>0.5</td>
<td>0.5</td>
<td>0.5</td>
<td>0.5</td>
<td>0.5</td>
<td>0.5</td>
</tr>
<tr>
<td>Captex 200</td>
<td>20</td>
<td>20</td>
<td>20</td>
<td>20</td>
<td>20</td>
<td>20</td>
<td>20</td>
<td>20</td>
<td>20</td>
</tr>
</tbody>
</table>
Table: Evaluation result of LOR nanoemulgel

<table>
<thead>
<tr>
<th>Formulation</th>
<th>pH</th>
<th>Viscosity (cps)</th>
<th>Spreadability (gm.cm/sec)</th>
<th>Extrudability (gm/cm²)</th>
<th>% Drug Content</th>
<th>% Drug Diffusion</th>
</tr>
</thead>
<tbody>
<tr>
<td>F1</td>
<td>5.85 ± 0.05</td>
<td>12600 ± 0.03</td>
<td>36.17 ± 0.29</td>
<td>11.52 ± 0.17</td>
<td>99.23 ± 0.08</td>
<td>90.23 ± 0.08</td>
</tr>
<tr>
<td>F2</td>
<td>5.76 ± 0.04</td>
<td>18000 ± 0.02</td>
<td>32.67 ± 0.58</td>
<td>23.63 ± 0.19</td>
<td>99.25 ± 0.04</td>
<td>93.65 ± 0.07</td>
</tr>
<tr>
<td>F3</td>
<td>5.97 ± 0.02</td>
<td>23200 ± 0.03</td>
<td>27.50 ± 0.50</td>
<td>19.67 ± 0.15</td>
<td>98.90 ± 0.03</td>
<td>88.54 ± 0.06</td>
</tr>
<tr>
<td>F4</td>
<td>5.63 ± 0.07</td>
<td>16500 ± 0.04</td>
<td>33.67 ± 0.58</td>
<td>14.46 ± 0.23</td>
<td>98.66 ± 0.04</td>
<td>87.59 ± 0.05</td>
</tr>
<tr>
<td>F5</td>
<td>5.73 ± 0.02</td>
<td>22500 ± 0.05</td>
<td>30.83 ± 0.76</td>
<td>20.60 ± 0.48</td>
<td>99.65 ± 0.04</td>
<td>85.65 ± 0.03</td>
</tr>
<tr>
<td>F6</td>
<td>5.92 ± 0.04</td>
<td>26100 ± 0.06</td>
<td>25.50 ± 0.50</td>
<td>18.58 ± 0.16</td>
<td>99.90 ± 0.05</td>
<td>83.56 ± 0.04</td>
</tr>
<tr>
<td>F7</td>
<td>5.64 ± 0.04</td>
<td>23000 ± 0.03</td>
<td>26.67 ± 0.58</td>
<td>21.66 ± 0.11</td>
<td>99.00 ± 0.04</td>
<td>82.54 ± 0.07</td>
</tr>
<tr>
<td>F8</td>
<td>5.86 ± 0.08</td>
<td>26400 ± 0.05</td>
<td>21.50 ± 0.50</td>
<td>18.58 ± 0.16</td>
<td>99.17 ± 0.05</td>
<td>81.74 ± 0.08</td>
</tr>
<tr>
<td>F9</td>
<td>5.94 ± 0.03</td>
<td>32100 ± 0.04</td>
<td>17.67 ± 0.58</td>
<td>15.60 ± 0.16</td>
<td>98.65 ± 0.08</td>
<td>80.59 ± 0.06</td>
</tr>
</tbody>
</table>

7.6.3 Evaluation of LOR nanoemulgel for different parameters

From all the emulgel formulations, select one optimised formulation for further study based on the formulation having a good physical appearance, good spreadability, good extrudability, and the highest in vitro diffusion. Batch L5 was selected as an optimised batch and further tested for release kinetics, skin permeation and skin retention, skin irritation, anti-inflammatory and...
analgesic activity, and a stability study. From the release kinetic study, we conclude that batch L2 follows the Higuchi model (diffusion-controlled release) and the Korsmeyer-Peppas model is the most fitted model with the highest $R^2$ value. The data from Korsmeyer-Peppas' model indicated that the $n$ values of 0.7521 were in the range of 0.5 to 1, implying a non-fickian (anomalous) drug release mechanism (couple of erosion and diffusion). The amount of drug permeated through the skin in 8 hours in optimised batch L2 was 60.35%, with drug retention in the skin at 24.28% and remaining on the skin at 15.37%. LOR skin irritation study on rats, which found no evidence of skin irritation. LOR nanoemulgel anti-inflammatory and analgesic activity also compare with the plain LOR gel result, which is also shown in Table 9. Stability studies conducted as per the ICH guidelines found the product to be physically and chemically stable, as no significant difference was observed.

Table 9: Result of LOR emulgel anti-inflammatory study.

<table>
<thead>
<tr>
<th>Group</th>
<th>Paw edema at different time after carrageenan injection</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Initial</td>
</tr>
<tr>
<td>Control (Plain base)</td>
<td>1.56 ± 0.07</td>
</tr>
<tr>
<td>Mean % Edema</td>
<td>17.94</td>
</tr>
<tr>
<td>Standard (LOR plain gel)</td>
<td>1.49 ± 0.08</td>
</tr>
<tr>
<td>Mean % Edema</td>
<td>16.11</td>
</tr>
<tr>
<td>Percent age of inhibition</td>
<td>10.26</td>
</tr>
<tr>
<td>Test (LOR Emulgel)</td>
<td>1.51 ± 0.07</td>
</tr>
<tr>
<td>Mean % Edema</td>
<td>15.23</td>
</tr>
<tr>
<td>Percent age of inhibition</td>
<td>15.13</td>
</tr>
</tbody>
</table>

(Each value was expressed as mean ± S.E.M. (Std error of mean) for 4 rat, and anti-inflammatory data was statistically analyzed using one way ANOVA. Reaction time for Std and test were compared with control group and ***P<0.001 was considered significant.)
Figure: 5 LOR emulgel Analgesic activity study compare with control and plain gel.

8. Achievements with respect to objectives

Different oils and surfactants were screened and statistically analysed for proper selection. Emulgels with desired quality could be achieved by employing QbD studies, which, in the present investigation, helped in optimising product and process variables impacting the CQA of emulgels. It was demonstrated that emulgels can be successfully prepared on a small scale using a suitable method. DoE is used to optimise formulation. Emulgels are evaluated for different parameters like pH, viscosity, spreadability, extrudability, drug content, % in-vitro drug diffusion, skin permeation, skin retention, skin irritation studies, etc. Stability studies conducted as per the ICH guidelines found the product to be physically and chemically stable, as no significant difference was observed in appearance, pH, viscosity, spreadability, extrudability, % drug content, and % in vitro drug diffusion.

9. Conclusion

The present research reveals the effectiveness of the quality by design methodology to optimise the formulation variables for the development of APM emulgel. A simple lattice mixture design was used to learn about the process of making emulsion formulations and improve their quality. Choose the oil, Smix, and water as the independent variables and the droplet size and percent in vitro drug diffusion as the dependent variables for constructing an emulsion. After testing several batches of prepared emulsion for different parameters, batch A5 was chosen as the optimised batch. In order to boost stability and permeability, a nanoemulsion formulation was transformed into an emulgel, which demonstrated considerable skin penetration and retention for effective topical delivery without causing skin irritation. Formation of emulgel by utilizing various grades and concentrations of carbopol polymer and evaluation of several parameters choose the optimised batch C5, which was made with carbopol 940 at a concentration of 0.75%, out of all the formulation batches since it has a more effective effect than the other batches and
produces a more stable formulation. Further studies determine the optimal formulation of the emulgel with regards to skin penetration and retention, kinetics, skin irritation, efficacy in psoriasis-induced mice, and stability. The best-fitting model based on release kinetics was found to be the Higuchi model with non-fickian diffusion. No skin irritation was noticed, and the psoriasis-induced mice used in the animal research responded favorably. Physical stability was confirmed during the stability investigation. Therefore, it was determined that nanoemulgel formulations have the potential to replace conventional topical formulations.

A 3²-factorial design was employed to understand the LOR emulsion formulation development process and improve its quality. In this study, Captex 200 was used as oil, Cremophore RH 40, Capmul MCM C8 as Smix, and Propylene glycol as a co-solvent to prepare a stable nanoemulsion to increase the effect of LOR. In the current study, the major factors influencing the critical quality attributes of the emulsion were oil and Smix ratio and S:Cos ratio, so they were chosen as independent variables and droplet size and% in vitro drug diffusion as dependent variables. After testing several batches of prepared emulsion for different parameters, batch L5 was chosen as the optimised batch. An optimised nanoemulsion formulation was converted into gel form by utilizing various grades and concentrations of carbopol polymer and evaluating it for different parameters. Choose the optimised batch F2, which was made with carbopol 934 at a concentration of 0.75%, out of all the formulation batches since it has a more effective effect than the other batches and produces a more stable formulation. Optimized batch further tested for skin penetration and retention, release kinetics, skin irritation, anti-inflammatory, analgesic, and stability study. The best-fitting model based on release kinetics was found to be the Higuchi model with non-fickian diffusion. No skin irritation was noticed, and the anti-inflammatory and analgesic activity in LOR nanoemulgel was found to be higher than that of LOR plain gel. Physical stability was confirmed during the stability investigation. Therefore, it was determined that LOR nanoemulgel formulations have the potential to replace conventional topical formulations.

10. Publications


**Oral presentation in conference:**


**11. References**


