Raman Gupta *et al*, International Journal of Pharmaceutical Sciences & Medicine (IJPSM),
Vol.7 Issue. 9, September- 2022, pg. 10-19

ISSN: 2519-9889
Impact Factor: 5.721

Preparation and Characterization of Luliconazole Nanoemulsion Based Hydrogel

Raman Gupta[1]; Twinkle Garg[2]; Dr. R.B. Sharma[2]

[1]Himalayan Institute of Pharmacy, Kala-Amb, H.P, e-mail: ramansinghal002@gmail.com

[2]Himalayan Institute of Pharmacy, Kala-Amb, H.P

DOI: 10.47760/ijpsm.2022.v07i09.002

Abstract

Fungal infection is one of the most common causes of skin diseases. The frequency of fungus infections is increasing worldwide. Oral treatment of fungal infections it has to do with the toxic effect, long-term treatment and patient intolerance while over-the-counter treatments for mild fungal infections are associated with reduced drugs, irritation to the skin and minimal clearance of the skin. So to improve drug penetration, to minimize side effects of drugs and rapid symptomatic relief from fungal infections, nanoemulsion based hydrogel (Carbopol 934) containing an antifungal drug was prepared. Excipients were screened on the bases of solubility of drug in these excipients. Cinnamon oil was selected as oil phase, Tween 80 as surfactant and PEG 400 as cosurfactant. Ternary phase diagram was constructed between oil Smix (Tween 80 and PEG 400 (1:1) and water. Luliconazole nanoemulsion was prepared and incorporated into hydrogel. Hydrogel (1%) was prepared by dispersing the Carbopol 934 in distilled water and kept for 24 hours for complete swelling of carbopol. Triethanolamine was added for crosslinking of gel. The prepared nanoemulsion based hydrogel formulations were then characterized for appearance, pH, rheology, and drug content and drug release. Drug content was found above 85% and drug release was sustained for 12 hrs. Viscosity and pH was found optimum and safe for topical use. Hence the formulation of antifungal LZL into Nanoemulsion for topical drug delivery proves to be promising carrier for the delivery of the drug.

Keywords: Fungal infection, Luliconazole, Nanoemulsion, Homogenizer, Hydrogel, Carbopol 934, Triethanolamine

1. Introduction

Nanoemulsion is a thermo-dynamically stable isotropically clear dispersion of two immiscible liquids, such as oil and water, stabilized by an interfacial film of surfactant molecules. In addition to the surfactant the Co-surfactant / Co-solvent is used with the oil and water phase. The dispersed phase typically comprises small particles or droplets, with a size range of 5-200 nm, and has very low oil / water interfacial tension (Jaiswal et al., 2015; Gupta et al., 2016).

ISSN: 2519-9889
Impact Factor: 5.721

Unlike coarse emulsions micronized with external energy, nanoemulsion is based on low interfacial tension which is achieved by adding a co-surfactant, which leads to spontaneous formation of a thermodynamically stable nanoemulsion (Kumar et al., 2012; Patel et al., 2011). The nanosized droplets leading to increase in interfacial surface area associated with nanoemulsion would affect the transport properties of the drug (Tadros et al., 2004). Nanoemulsions have a larger capacity for micellar solubilisation compared to simple solutions, give advantages in thermodynamic stability to unstable dispersions as it can be produced with less energy input, and have a greater shelf life a low viscosity, high kinetic stability against creaming or sedimentation, and large interfacial area make nanoemulsion of increasing use in different applications (Shen et al., 2011). Nanoemulsions have been increasingly developed for use as drug-delivery systems for parenteral, oral, ocular, and topical administration (Araujo et al., 2011; Hagigit et al., 2012). Specifically, in topical delivery, nanoemulsions offer several significant advantages including no skin irritation, powerful permeation ability, and high drug-loading capacity (Abdulkarim et al., 2010; Sonneville-Aubrun et al., 2004).

Nanoemulsion-based cosmeceuticals have improved efficacy; whereby, the active ingredient will have better skin penetration and a higher rate in successful drug delivery to the target site due to its small particle size. Besides, the long-term colloidal stability of nanoemulsion can be achieved through high potential due to the increase of the repulsive force between droplets. Nanoemulsion is a non-equilibrium colloidal system where oil phase is dispersed as fine droplets, usually with particle size from 20–200 nm, throughout the aqueous phase stabilized by surfactants. Nanoemulsions can be prepared by high-energy emulsification or by low-energy emulsification methods. However, nanoemulsions are thermodynamically unstable colloidal systems that are highly dependent on their physicochemical properties, usually based on the preparation method (Mason et al., 2014; Dhadde et al., 2020).

Nanoemulgel

Nanoemulgel which known as the formation of nanoemulsion based on hydrogel is the addition of nanoemulsion system intergraded into hydrogel matrix which influences a better skin penetration (Mou et al., 2008). This mixture of nanoemulgel has attracted the attention of many scientists for the development of numerous drugs that function to treat various kinds of skin disorders. The formulation of nanoemulgel for the topical delivery system acts as drug reservoirs which, influence the release of drugs from the inner phase to the outer phase and then further onto the skin (Bernard 2012). These release mechanism depends on the composition of the network polymer chains and the crosslink density (Alves et al., 2007). Besides that, the ability of a drug to permeate the skin and successfully release of therapeutic agent is influenced by drug affinity to diffuse out from the vehicle and permeate through barrier (Babihav et al., 2011). Nanoemulgel on intact with skin will release the oily

ISSN: 2519-9889
Impact Factor: 5.721

droplets from the gel network. The oil droplets then will penetrate into the stratum corneum of the skin and directly deliver the drug molecules without a transfer via hydrophilic phase of nanoemulsions (Mou et al., 2008).

Luliconazole

Luliconazole (LUL), an antifungal drug containing imidazole moiety with ketone dithioacetate, is a broad-spectrum agent, which has shown its potential against wide varieties of fungi, especially against filamentous fungi, for example, dermatophytes.

Although the exact mechanism of this novel agent for antifungal efficacy is unknown, it has been reported that LUL acts by inhibiting the fungal cytochrome P450; that is, 14-α demethylase enzyme thus prevents the biosynthesis of ergosterol from lanosterol and interrupts cellwall synthesis within the fungi. Solubility of LUL is low which restricts permeation of the drug across the skin upon topical delivery. Alternatively, conventional topical cream formulations possess several drawbacks of low permeation from the stratum corneum along with reduced retention at the site of application. Moreover, the rate-limiting step for LUL permeation is its solubility in the lipid phase of the stratum corneum that limits its dermal availability (Nabil et al., 2021). There is an urgent requirement for novel deliveries for improved retention and penetration from the site of skin application.

In context of above principle a strong need was recognized for the development of sustained release drug delivery system for the drug. An approach known to fulfil both the needs is Luliconazole nanoemulgel. This is useful in sustaining the drug release. The prepared formulation will be characterized for several parameters such as appearance, pH, rheology, and drug content and drug release.

2. MATERIAL AND METHODS

Materials

Luliconazole was received as gift sample from Allastir Pvt. Ltd. Chennai. Other excipients like Carbopol 934, Castor oil, cinnamon oil, PEG, triethanolamine, Tween 40 and Tween 80 etc. were obtained from college laboratory.

Experimental Methods

Formulation of Nanoemulsion

a. Construction of Pseudo ternary phase diagram

On the basis of the solubility studies and drug excipient compatibility studies, Cinnamon oil was selected as the oil phase. Tween-80 and PEG400 were selected as surfactant and co-surfactant, respectively. Distilled water was used as an aqueous phase. Surfactant and co-surfactant (Smix) were mixed at (1:1) ratio and prepared phase diagram. For phase diagram, oil and Smix at a specific ratio was mixed thoroughly at different mass ratios from 1:9 to 9:1 in different glass vials (Chang et al., 2020). The mixtures of oil, surfactant and co-surfactant at certain

ISSN: 2519-9889
Impact Factor: 5.721

weight ratios were titrated by adding water dropwise, under moderate magnetic stirring. After being equilibrated, the mixtures were assessed visually and at the same time examined for the transparency. Samples with low viscosity, single phase and transparent nature were considered as stable formulation. The data obtain after titration was used for the construction of ternary phase diagram.

b. Preparation of Nanoemulsion

The preparation of NE was performed using high speed homogenizer. In this technique, the drug was dissolved in the lipid. The lipid phase was then added to the aqueous Smix (surfactant and co-surfactant v/v) phase heated at 70-80^{o}C under stirring at 11000 rpm with the help of high speed homogenizer for at least 15 min (Gupta et al., 2016). Three formulations were designed by altering the concentration of formulation variables like Lipid, S mix as shown in Table I. A 5% w/v PVA solution was prepared by adding 0.25 gm of PVA to 4ml deionized water and heated at 60^{o}C to dissolve PVA added.

Ingredients	Role	F1	F2	F3
Drug (mg)	API	100	100	100
Cinnamon Oil (ml)	Carrier	0.9	1.4	1.9
S mix (ml)	Emulsifier	3.5	2.5	1.3
Water (ml)	Carrier	5.1	5.6	6.3

Table I: Compositions of various nanoemulsion formulations

Preparation of Hydrogel

Hydrogel was prepared by dispersing the 1gm of Carbopol 934 in sufficient quantity of distilled water. After complete dispersion, the solution was kept for 24 hours for complete swelling of carbopol 934. Triethanolamine was added for crosslinking. Clear and transparent 1% gel of Carbopol 934 was prepared (Helal et al., 2012).

Incorporation of Nanoemulsion into Hydrogel

Nanoemulsion was incorporated to hydrogel (1:1 w/w) with continuous stirring. Prepared nanoemulsion based hydro gel was placed in a suitable container (Helal et al., 2012).

Characterization of nanoemulsion based hydrogel

Appearance

Prepared nanoemulsion based hydrogel was visually checked for its colour and clarity.

Drug content

The drug content of nanoemulsions was determined by diluting 1mL of the formulation with 100 mL methanol, further diluted 10ml to 50ml with methanol followed by analysis with UV-visible spectrophotometer (Shishu et al., 2006).

ISSN: 2519-9889
Impact Factor: 5.721

pH

The pH of nanoformulations was determined using a pH meter at 25 ± 0.5°C.

Rheology study of Nanoemulsion based hydrogel

The viscosity of Nanoemulsion based hydro gel of different formulation was measured at 10 rpm for 3 min at 25 C by Brookfield type rotary viscometer with spindle (Kaplan et al., 2019).

***In vitro* release**

About 2gm of prepared gel were placed on dialyzing membrane containing diffusion cell phosphate buffer 7.4 in its receptor compartment, 1ml of phosphate buffer was taken after 1 hr interval up to 12 hr. Mention steady state condition in diffusion cell by placing 1ml of fresh phosphate buffer after removal of 1ml of sample from receptor compartment (Barradas et al., 2017).

3. RESULT AND DISCUSSION

Preparation of Nanoemulsion

Construction of Pseudo Ternary Phase diagram

The solubility of oil (Cinnamon oil, castor oil, soyabean oil and oleic acid), surfactant (Span 80, Span 20, Tween 20 and Tween 80) and co-surfactant (PEG 400, PEG 300 and PG) was determined and displayed in Table II.

S. No.	Compound	Solubility (mg/ml)
OIL		
1	Cinnamon Oil	105.1 ±0.08
2	Castor Oil	17.85±0.38
3	Soyabean Oil	3.96±0.09
4	Oleic acid	5.11±0.06
SURFACTANT		
1	Span 80	0.11±0.11
2	Span 20	4.17±0.28
3	Tween 80	7.63±0.07
4	Tween 20	2.77±0.05
CO-SURFACTANT		
1	PEG 400	22.39±0.2
2	PEG 300	12.38±0.06
3	PG	2.89±0.03

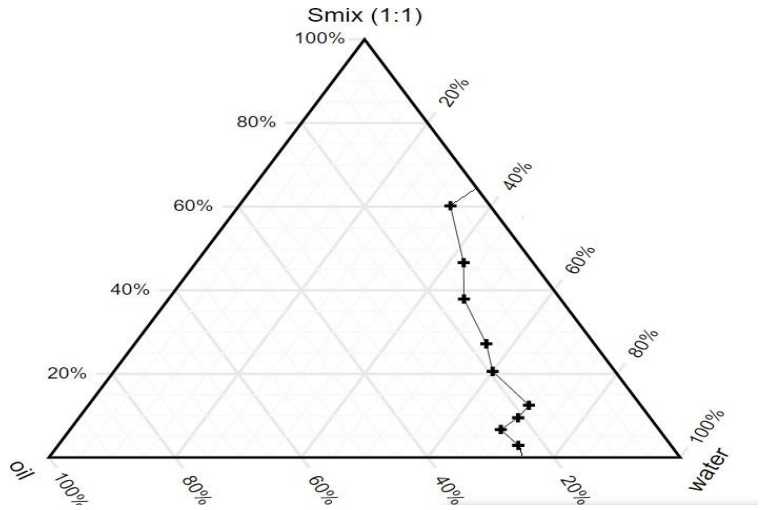
Table II Solubility of Luliconazole in oil, surfactant and co-surfactant (mean ±SD, n=3)

ISSN: 2519-9889
Impact Factor: 5.721

Ternary phase diagram

On the basis of the solubility studies and drug excipient compatibility studies, Cinnamon oil was selected as the oil phase. Tween-80 and PEG400 were selected as surfactant and co-surfactant, respectively. Distilled water was used as an aqueous phase. Surfactant and co-surfactant (Smix) were mixed at (1:1) ratio and prepared phase diagram (Fig. I).

Figure I Pseudo Ternary Phase Diagram



Characterization of nanoemulsion based hydrogel

Prepared nanoemulsions were characterized for appearance; pH, viscosity and drug content (Table III).

Appearance

Prepared formulations were visually checked for its colour, and clarity. They were found yellow and clear.

Drug content

The drug content of formulation F1, F2 & F3 was found to be 96.62%± 1.13%, 99.51%±0.4% and 87.4%±2.10% respectively.

pH

pH of nanoemulsions F1, F2 and F3 was found to be 7.29±0.02, 7.33± 0.04 and 7.13 ± 0.02 respectively.

Viscosity

The viscosity of formulation F1, F2 & F3 was found to be 4910 ±2.59 mPa.S, 4851±2.56 mPa.S and 4893 ±3.66 mPa.S respectively.

ISSN: 2519-9889
Impact Factor: 5.721

Formulation Code	Appearance	pH	Viscosity (mPa.S)	Drug content
F1	Yellow colored clear transparent gel	7.29±0.02	4910 ±2.59	96.62%±1.13%
F2	Yellow colored clear transparent gel	7.33±0.04	4851±2.56	99.51%±0.4%
F3	Yellow colored clear transparent gel	7.13±0.02	4893 ±3.66	87.4%±2.10%

Table III Characterization of nanoemulsion based hydrogels

In vitro release

The in vitro release of all formed batches exhibited a sustained delivery of LZL (>70% in 12 h) (Table IV, Fig. II). The first hour release was found to be moderate (10-25%) i.e. no initial burst release was observed**.**

Time (hrs)	% drug release		
	F1	**F2**	**F3**
1	13.58	16.04	13.67
2	22.12	24.75	18.36
3	28.34	30.85	26.74
4	35.65	37.35	35.18
5	48.05	51.78	45.67
6	55.76	57.5	57.64
7	62.12	67.77	64.37
8	66.03	76.26	69.48
9	71.41	79.84	74.58
10	82.31	88.37	77.37
11	86.13	90.76	79.48
12	91.11	94.87	83.76

Table IV % Drug release of nanoemulsion based hydrogels

ISSN: 2519-9889
Impact Factor: 5.721

In vitro release graph of all three formulations (figure II) shows that formulation F2 give the better release as compared to formulation F1 and F3. So further studies done on formulation F2.

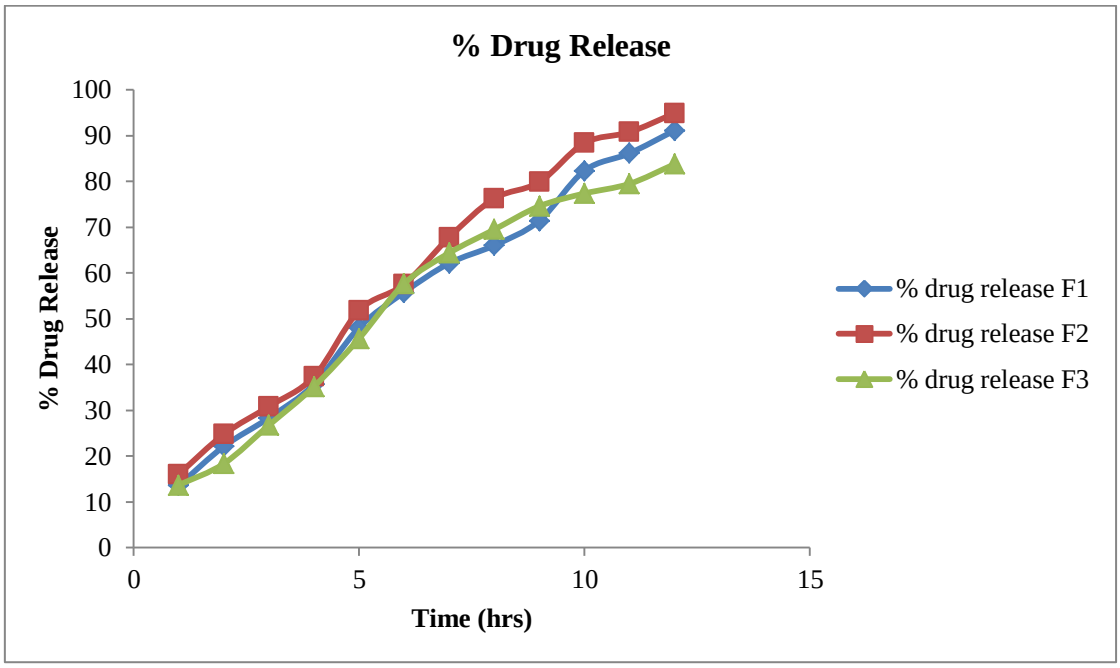


Figure II % Drug Release of nanoemulsion based hydrogels

4. CONCLUSION

In conclusion, nanoemulsions composed of antifungal drug and natural novel excipients were prepared. A homogenization method was employed. Different characterization study and in vitro studies supports the successful formulation of nanoemulsion based hydrogel, safe and effective of antifungal drugs in the skin.

5. ACKNOWLEDGEMENT

I would like to express my special thanks of gratitude to my guide Ms. Twinkle Garg for their able guidance and support in completing my research.

ISSN: 2519-9889
Impact Factor: 5.721

REFERENCES

[1]. Jaiswal M., Dudhe R., Sharma PK., 2015, Nanoemulsion: an advanced mode of drug delivery system, *Biotech*, 5, 2, pp. 123-127.

[2]. Gupta A., Eral B., Hatton T., Doyle P., 2016, Nanoemulsions: Formation, Properties and Applications, *Soft Matter*, 12, pp. 1-16.

[3]. Kumar SL., Singh V., 2012, Nanoemulsification - A Novel Targeted Drug Delivery Tool, *Journal of Drug Delivery and Therapeutics*, 2, 4, pp. 40-45.

[4]. Patel R., Patel G., Bharadia P., 2011, Nanoemulsion: An Advanced Concept of Dosage Form, *IJPI's Journal of Pharmaceutics and Cosmetology*, 2011, pp. 1-5.

[5]. Tadros T., Izquierdo P., Esquena J., Solans C., 2004, Formation and stability of nanoemulsions, *Advances in Colloid and Interface Science*, 108, 109, pp. 303-318.

[6]. Shen Q., Wang Y., Zhang Y., 2011, Improvement of colchicine oral bioavailability by incorporating eugenol in the nanoemulsion as an oil excipient and enhancer, *International Journal of Nanomedicine*, 6, pp. 1237-1243.

[7]. Araújo FA., Kelmann RG., Araújo BV., Finatto RB., Teixeira HF., Koester LS., 2011, Development and characterization of parenteral nanoemulsions containing thalidomide, *European Journal of Pharmaceutical Science*, 42, 3, pp. 238–245.

[8]. Hagigit T., Abdulrazik M., Valamanesh F., Behar-Cohen F., Benita S., 2012, Ocular antisense oligonucleotide delivery by cationic nanoemulsion for improved treatment of ocular neovascularization: an in-vivo study in rats and mice, *Journal of Controlled Release*, 160, 2, pp. 225–231.

[9]. Abdulkarim MF., Abdullah GZ., Chitneni M., Salman IM., Ameer OZ., Yam MF., Mahdi ES., Sattar MA., Basri M., Noor AM., 2010, Topical piroxicam in vitro release and in vivo anti-inflammatory and analgesic effects from palm oil esters-based nanocream. *International Journal of Nanomedicine*, 5, pp. 915-24.

[10]. Sonneville-Aubrun O., Simonnet JT., Alloret F., 2004, Nanoemulsions: a new vehicle for skincare products, *Advances in Colloid and Interface Science*, 108–109, pp. 145 –149.

[11]. Mason TG., Wilking JN., Meleson K., Chang CB., Graves SM., Nanoemulsions: formation, structure, and physical properties, *Journal of Physics: Condesed Matter*, 18, 41, pp. 635–666.

[12]. Dhadde SB., Patil JS., Chandakavathe BN., Thippeswamy BS., Kavatekar MG., 2020, Relevance of Nanotechnology in Solving Oral Drug Delivery Challenges: A Perspective Review, *Critical Reviews™ in Therapeutic Drug Carrier Systems*, 37, 5, pp. 407-34.

[13]. Mou D., Chen H., Du D., Mao C., Wan J., Xu H., Yang X., 2008, Hydrogel-thickened nanoemulsion system for topical delivery of lipophilic drugs. *International Journal of Pharmaceutics*, 353, (1-2), pp. 270-6.

[14]. Bernard PB., 2012, Modern Aspects of Emulsion Science, Emulsions -Recent Advances in Understanding, UK: Royal Science of Chemistry.

ISSN: 2519-9889
Impact Factor: 5.721

[15]. Alves MP., Scarrone AL., Santos M., Pohlmann AR., Guterres SS., 2007, Human skin penetration and distribution of nimesulide from hydrophilic gels containing nanocarriers, *International Journal of Pharmaceutics*, 341, (1-2), pp. 215-20.

[16]. Baibhav J., Gurpreet S., Seema S., Vikas S., 2011, Emulgel: A Comprehensive Review On The Recent Advances In Topical Drug Delivery, *International Research Journal of Pharmacy*, 2, 11, pp. 66-70.

[17]. Luliconazole, National Center for Biotechnology Information. Pub Chem Compound Database; CID=3003141.

[18]. Nabil A., Shadab Md., Alam Md., Rasheed S., Javed A., Abrar A., Hussam K., Ahmad N., Alaa B., Douha B., Bapi G., Ponnurengam S., 2021, Development, Optimization, and Evaluation of Luliconazole Nanoemulgel for the Treatment of Fungal Infection, *Journal of Chemistry*, 2021, pp. 1-13.

[19]. Chang M., Guo Y., Jiang Z., Shi L., Zhang T., Wang Y., Gong M., Wang T., Lin R., Liu R., Wang Y., Jin Q., Wang X., 2020, Sea buckthorn pulp oil nanoemulsions fabricated by ultra-high pressure homogenization process: A promising carrier for nutraceutical, *Journal of Food Engineering*, 287, 110129.

[20]. Helal D., El-Rhman D., Abdel-Halim SA., El-Nabarawi M., 2012, Formulation and evaluation of fluconazole topical gel, International Journal of Pharmacy and Pharmaceutical Sciences, 4, pp. 176-183.

[21]. Aggarwal SN., 2006, Preparation of hydrogels of griseofulvin for dermal application, *International Journal of Pharmaceutics*, 326, (1-2), pp. 20-4.

[22]. Uğur Kaplan AB., Cetin M., Orgul D., Taghizadehghalehjoughi A., Hacimuftuoglu A., Hekimoglu S., 2019, Formulation and in vitro evaluation of topical nanoemulsion and nanoemulsion-based gels containing daidzein, *Journal of Drug Delivery Science and Technology*, 52, pp. 189-203.

[23]. Barradas TN., Senna JP., Cardoso SA., Nicoli S., Padula C., Santi P., Rossi F., de Holanda E Silva KG., Mansur CRE., 2017, Hydrogel-thickened nanoemulsions based on essential oils for topical delivery of psoralen: Permeation and stability studies, *European Journal of Pharmaceutics and Biopharmaceutics*, 116, pp. 38-50.

