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Design and Evaluation of Topical Nanoemulgel Containing Curcumin and Aloe Vera for Antimicrobial Activity

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ABSTRACT

Aim: The aim of present work was to prepare curcumin (CUR) and aloe vera nanoemulgel and characterization for its antimicrobial activity.

Objective: The objective of the present work was to enhance solubility and permeability of CUR and aloe-vera.

Materials and Methods: Nanoemulsion of CUR was prepared by constructing pseudo ternary phase diagram with varying concentrations of surfactant (tween 80), co-surfactant (PEG 400) and oil (Isopropyl myristate) using aqueous phase titration method. Six formulations (FE1-FE6) of nanoemulsion were prepared on the basis of Smix ratios 1:1, 2:1, 1:2, 3:1. Nanoemulsion were further characterized for pH, particle size, zeta potential, drug content, Thermodynamic stability, in-vitro drug permeation study and flux. FE3 was selected as an optimized formulation depend on droplet size, zeta potential drug permeation study and further FE3 was incorporated into 1.5% carbopol 934 gel base. Eight nanoemulgel (FG1-FG8) were prepared by using cold method. Nanoemulgel were subjected to physical appearance, pH, rheology, spreadability, in-vitro drug release study, kinetic modeling and antimicrobial activity study. Optimized nanoemulgel formulation was compared with marketed formulation (POVIDONE IDODINE OINTMENT).

Result: It was observed that nanoemulgel FG4 (40±0.72µg/cm^2/hr) show two fold increase in transdermal flux as compared to marketed (17.37±0.52µg/cm^2/hr). In- vitro drug release data obtained was fit into different kinetic models like zero order, first order, Higuchi, Hixson-Crowell, Korsmeyer- Peppas plot. The coefficient (R^2) was considered to be main parameter for interpreting the release kinetics of drug from matrix systems. The optimized FG4 formulation drug release data was fitted to First order with R^2 value 0.996. FG4 showed maximum zone of inhibition i.e 8.14 mm was more as compared to marketed formulation (7.0 mm).

Conclusion: From the results it can be concluded that nanoemulgel formulation of CUR and aloe-vera gelis safe and effective for the treatment of microbial infections.

Key Words: nanoemulgel, nanoemulsion, curcumin, antimicrobial activity, carbopol 934, aloe vera.

Abbreviations

FE: Formulation of nanoemulsion
FG: Formulation of nanoemulgel
Ml: Milliliter
Nm: Nanometer
Mg: Milligram
µm: Micrometer
cm: Centimeter
ppm: Parts per million
rpm: rotations per minute
UV: Ultraviolet
Conc: Concentration
^{0}C: Degree celsius
Hrs: Hours
Min: Minutes
Lit: Litre
µg: Microgram
CUR: Curcumin

I. INTRODUCTION

Topical drug administration is a localized drug delivery system anywhere in the body through ophthalmic, rectal, vaginal and skin as topical routes. Topical drug delivery system consist the largest organ of body i.e skin which provides around 10% of body mass of an average person and it covers 1.7 m^2

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average area. The main advantage of topical delivery system is it bypasses first pass metabolism (Kumar et al., 2016).

Emulgel, as the name suggest are the combination of gel and emulsion. Both o/w and w/o type of emulsion used as vehicle to deliver varied drugs to the skin. Gels are newer type of dosage form i.e. made by trapping large amount of aqueous or hydro alcoholic liquid in a network of colloid solid particle that can be made of inorganic substance like aluminum salts or organic polymers of natural or synthetic origin. When compared to ointment and cream base, they have higher aqueous component which allows for drug dissolution and fast drug migration through vehicle i.e. liquid (Sanjay et al., 2019, Kute et al., 2013). These are superior in terms of ease of use and patient comfort.

Despite many benefits of gels one major drawback in delivery of drugs that are hydrophobic. So to overcome this, emulgel are prepared and used. So that allowing evens a hydrophobic therapeutic moiety to benefit from unique properties of gel (Shailendra et al., 2015).Emulgel for dermatological use have several favorable properties such as being thixotropic, greaseless, easily spreadable, easily removable, emollient, non-staining, water soluble, longer shelf life, bio friendly, transparent and pleasing appearance (Hardenia et al., 2014).

II. MATERIAL AND METHODS

Materials

CUR, Aloe vera gel, Tween 80, Isopropyl myristate oil (IPM), Carbopol 934, Triethanolamine. CUR was obtained as a gift sample from Himalaya Drug Company, Bangalore. Aloevera gel was purchased Green Pharmacy, Pune and other chemicals were purchased from Loba chemicals, Mumbai. The microbial strain (Staphylococcus aureus) employed in the study was obtained from the Dr. Vikhe Patil Memorial Hospital and Medical College, Ahmednagar.

Equipment's

List of equipment's are shown in below table no.1

Methods

Preformulation studies

Appearance and colour

The drug (CUR) powder and aloe vera gel was examined for its organoleptic properties like colour and appearance.

Melting point determination

The melting point of the CUR and aloe vera gel was determined by open capillary method using melting point apparatus.

Solubility study

Excess amount of CUR was taken and added in 10 ml of different solvents. These solutions were shaken well and kept for 24 hrs at room temperature. Then the solubility was determined (Bele et al., 2017).

Determination of λ_{max} and calibration curve of CUR in phosphate buffer pH 7.4

10 mg of CUR weighed and transferred in 100ml volumetric flask and methanol was added upto the mark to get an amount of 100µg/ml of stock solution. 0.2, 0.4, 0.6, 0.8,1.0 ml solution were withdrawn from stock solution and diluted upto 10 ml with phosphate buffer pH7.4 to make 2, 4, 6, 8, 10 µg/ml respectively (Kadam et al., 2018).

Determination of solubility of CUR

Solubility of CUR was determined in different oils, surfactant, and co-surfactants. Excess amount of CUR added to solvents and allowed it to dissolve by mixing in orbital shaker for 48hrs at 150 rpm at ambient temperature. Centrifugation method undissolved CUR separated out at 5000 x g and then concentration of CUR in supernatant was determined by measuring absorbance at 421 nm. Sample was scanned between 200-800 nm using JASCO UV spectrophotometer to determine wavelength of maximum absorbance. The absorbance of each standard solution was determined spectrophotometrically at 421 nm. The Beer's-Lambert's plot was constructed by plotting concentration Vs its corresponding absorbance (Syed et al., 2018, Chandrasekaran et al., 2018, Ahmed et al., 2019)

Compatibility study (Fourier Transforms Spectroscopy)

The IR Spectra of CURand excipients were recorded by BRUKER FTIR spectrophotometer. Sample were prepared by KBr disc method and examined in the transmission mode spectrum was

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measured over frequency range of 4000-400 cm^{-1}.The peaks obtained in the spectra were then compared with the corresponding functional groups in structure of CUR (Naz et al., 2015).

Construction of Pseudo-Ternary Phase Diagram

On the basis of solubility study of drug, isopropyl myristate was selected as the oil phase. Tween 80 and PEG 400 were selected as surfactant and co-surfactant as per their emulsification capability for the system. For the construction of the phase diagram for the determination of the development area of nanoemulsion, distilled water was used as an aqueous phase. The aqueous titration method was used to construct pseudo ternary phase diagrams. Four weight ratios (1:1, 2:1, 1:2, and 3:1) of Tween 80 to PEG400 were optimized based on solubility to determine the optimum ratio which can result in maximum nanoemulsion existence area. To construct pseudo ternary phase diagrams the surfactant: co-surfactant was mix with oil phase used for titrations are 1:1, 1:2, 1:3,1:4,1:5,1:6,1:7,1:8,1:9 the mixture was titrated with distilled water until it turned turbid. The volume of water used was recorded water titration was continue until a clear, Isotropic and thermodynamically stable dispersion with low viscosity was obtained (Javed et al., 2013).

Method of preparation of nanoemulsion formulation

Manufacturing formula for nanoemulsion formulation is shown in table no.2

Method of preparation of gel base

Carbopol gels were prepared by incorporating different concentration, 1%, 1.5% w/v of carbopol in 1% w/v triethanolamine in double distilled water. Weighted amount of carbopol was taken and dispersed over in distilled water for 2 hrs till all the carbopol is soaked. After soaking add triethanolamine. Homogenized for 2 hrs at 600 rpm. After homogenization carbopol gel was subjected for two cycles of sonication for 15 min to expel out the entrapped air bubbles from the prepared gel. Similarly other gel formulation was prepared. (Enas et al., 2015) Manufacturing formula for gel base is shown in table no.3

Method of preparation of CUR and aloe vera nanoemulgel

Different formulations of nanoemulsion and nanoemulgel were prepared by using the varying amount of emulsifier by spontaneous emulsification method. Nanoemulgels were prepared by using spontaneous emulsification method. Optimized nanoemulsion was incorporated into gel base to obtain nanoemulgel. (Conxita et al., 2012). Varying amount of aloe vera and CUR nanoemulsion concentrations were combined with gel base for the synergistic effect of aloe vera gel against bacterial infection as shown in table no.4

Evaluation of nanoemulsion

Nanoemulsions were evaluated for following properties.

Physical appearance

The prepared formulations were inspected visually for their colour and appearance (Mishra et al., 2014).

Droplet size and zeta potential measurement

Droplet size and zeta potential were determined by dynamic light scattering method (DLS), using a computerized inspection system (Malvern Zetamaster, ZEM 5002, Malvern, UK). The formulation were diluted by 1/4th distilled water before measurement and measured 3 times at a scattering angle of 90^0 between laser and detector. The polydispersity index (PI) was used as a measurement of the width of the size distribution. Polydispersity index (PI) less than 0.4 indicates a homogenous and monodisperse population. Zeta potential was measured as the particle electrophoretic mobility means of laser microelectrophoresis in a thermostated cell (Ambekar et al., 2017, Ambekar et al., 2018)

Viscosity determination of nanoemulsion

The viscosity of nanoemulsion of different formulations was measured at 10 rpm for 3 min at 25 ^{0}C by Brookfield type rotary viscometer with spindle 61.

Drug content

The amount of drug contained in the prepared nanoemulsion was determined by diluting required amount of prepared formulation using phosphate buffered saline (PBS) 7.4. This mixture was analyzed by UV spectrophotometer at 421nm against PBS 7.4 as blank. (Padsalg et al., 2007)

Thermodynamic stability test

Thermodynamic stability of the Nanoemulsions system was determined by performing following tests. (Ahmad et al., 2019)

Heating Cooling Cycle

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Nanoemulsion formulations were subjected to three cycles between refrigerator temperature 4 °C and 45 °C with storage for 48 hrs. Centrifugation test was donefor the optimized formulation.

Centrifugation

Nanoemulsion formulations were centrifuged at 3500 rpm for 30 min and those formulations which did not show any phase separation were taken for the freeze thaw stress test.

Freeze Thaw Cycle

In this the formulation were subjected to three freeze thaw cycles between -21°C and +25°C with storage at each temperature for not less than 48 hrs was done for the prepared formulations.

In- vitro drug permeation study

Franz diffusion cell was used for study of drug permeation. Prepared nanoemulsion was placed onto the surface of cellophane membrane (Dolphin, code -1540). The membrane was clasp between donor and receiver compartments. The receiver compartment was filled with phosphate buffer pH 7.4. The receiver compartment solution was stirred at 600 rpm by placing magnetic beads to it by maintaining 35^0C.The 1ml sample of aliquots withdrawn from 1 to 7 hrs intervals and to this fresh pH 7.4 phosphate buffer added to donor compartment to maintain sink condition. (Kaur et al., 2013) The samples were analysed using UV-VIS spectrophotometer at 421nm with appropriate dilutions.

Evaluation of gel

pH determination

pH determination of prepared formulations was done by using digital pH meter.

Rheology study of Gel

The viscosity of carbopol gel of different formulations was measured at 10 rpm for 3 mins at 25^0 C by Brookfield type rotary viscometer with spindle 64.

Evaluation of nanoemulgel

Nanoemulgel were evaluated for their physical appearance, pH, rheology, spreadability, extrudability, swelling index, drug content, In-vitro release study. (Kaur et al., 2013)

Physical appearance

The prepared nanoemulgel formulations were inspected visually for their colour, homogeneity, consistency, grittiness and phase separation. (Basera et al., 2015)

pH determination

pH determination of prepared formulations was done by using digital pH meter. The procedure was carried out by taking nanoemulgel in 250 ml beaker immersing pH meter into the formulation and readings of pH meter were recorded. Same process was repeated for 3 times with the same formulation. (Gadkari et al., 2019, Shadab et al., 2020)

Rheology study of nanoemulgel

The viscosity of nanoemulgel of different formulations was measured at 10 rpm for 3 min at 25 ^{0}C by Brookfield type rotary viscometer with spindle 63. (Gadkari et al., 2019)

Spreadability

A ground glass slide is fixed on this block. An excess of nanoemulgel (about 2 gm) under study is placed on this ground slide. The nanoemulgel was sandwiched between this slide and another glass slide having the dimension of fixed ground slide and provided with the hook. 500gm weight was placed on the top of the two slides for 5 min to expel air and to provide a uniform film of the nanoemulgel between the slides. Excess of the nanoemulgel was scrapped off from the edges. With the help of string attached to the hook and the time (in seconds) required by the top slide to cover a distance can be noted. A shorter interval indicates better spreadability. (Shadab et al., 2020) Spreadability was calculated by using the formula.

$$S = M.L/T$$

Where,
S = spreadability,
M = Weight tied to upper slide,
L = Length of glass slides
T = Time taken to separate the slides completely from each other.

Kinetic modeling of drug release data

Whenever a new dosage form is developed or produced, it is necessary to ensure that the drug dissolution/ diffusion occur in an appropriate manner. Drug diffusion from solid dosage form has been described by kinetic models in which the diffused amount of drug 'Q' is a function of time't'. Some analytical definitions of the 'Q','t' are commonly used such as zero order, first order, Hixson-Crowell

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and Higuchi equation, Korsmeyer-peppas models. (Higuchi et al., 1963)

In-vitro drug permeation study

Franz diffusion cell was used for study of drug permeation. Prepared nanoemulgel 1gm and marketed formulation was placed onto the surface of cellophane membrane. The membrane was clasp between donor and receiver compartments. The receiver compartment was filled with phosphate buffer pH 7.4. The receiver compartment solution was stirred at 600 rpm by placing magnetic beads to it by maintaining 25 ^{0}C. The 1ml samples of aliquots withdrawn from 1 to 7 hrs intervals and to this fresh pH 7.4 phosphate buffer added to donor compartment to maintain sink condition. (Bajjuri et al., 2018). The samples were analyzed using UV-VIS spectrophotometer at 421 nm with appropriate dilutions.

Antimicrobial activity study of nanoemulgel formulations

Microbial Strains

The microbial strain employed in the study was obtained from the Dr. Vikhe Patil Memorial Hospital and Medical College, Ahmednagar.

Staphylococcus aureus used to assess susceptibility patterns against the phytochemical extracts.

In vitro, Evaluation of antibacterial activity of CUR aloe vera nanoemulgel was measured by using the agar diffusion method (the cup plate method). Staphylococcus aureus bacteria were used in the study. Nutrient agar is used as a culture media for antimicrobial assay. Sterilized molten nutrient agar was dipped into sterilized petri dishes and leave for solidify. The plates were swabbed the 100ml culture of the microorganisms. By using a sterilized well bore at an equidistant position 6mm diameter uniform- sized cups were aseptically punched into seeded agar medium. The prepared gel samples were filled into the cylinder cup and incubated at 37± 0.5^{0}C for 48 hrs. The diameter (mm) of the zone of growth inhibition was estimated as the diameter (mm). (Patel et al., 2008, Kumar et al., 2020, Ahmad et al., 2019)

Composition of Nutrient agar media

Peptone- 50gm
Beef exract-3.0gm
Sodium chloride-8.0gm
Agar-15gm
Water -1litre

III. RESULT

Preformulation study

Color

The drug sample of CUR was found to be yellow colored powder.

Melting point

The melting point of CUR was found to be 185±2^{o}C.

Solubility study

CUR shows maximum solubility in tween 80 (3.03 ± 1.5 µg/ml) and PEG 400(12.91 ± 2.0µg/ml) hence they were selected as surfactant and co-surfactant. Isopropyl myristate as oil (15.68 ±1.21 µg/ml).The ratio of surfactant co-surfactant (1:1) was selected as optimized ratio for preparation of nanoemulsion.

Determination of λ_{max} and calibration curve of CUR in phosphate buffer pH 7.4

The Calibration curve of CUR was performed in phosphate buffer pH 7.4 and λ_{max} of CUR was found to be 421 nm. The calibration curve of CUR in this media concentration range of 1 to 7 µg/ml having coefficient of regression R^2 = 0.996 and was found to be linear in the slop m =0.211 as shown in figure no. 1

Compatibility study

Fourier transforms Infrared spectroscopy (FT-IR)

FTIR characteristic of CUR are observed in the physical mixture of drug and excipients, there was no major interaction except some minor physical interaction which might be due to hydrogen bonding and wander wall forces / covalent bonding. (Naz et al., 2015). Hence this proves there is no potential incompatibility with drug and excipient as shown in figure no. 2 &3.

Construction of Pseudo-Ternary Phase Diagram

IPM oil, Tween 80, PEG 400 and water were taken to construct the pseudo-ternary phase diagrams for development of the CUR nanoemulsion using Past Software. Figureno. 4 shows the results for this study. 1:1 Smix ratios showed the maximum amount of emulsification area at 5 and 6 point with dark zone. Decrease concentration of oil increase in area of emulsification. (Shah et al., 2017)

Phase diagrams developed by the aqueous phase titration method for nanoemulsion zones of CUR (dotted area) for IPM, Tween 80 and PEG 400 at Smix ratios of 1:1.

Evaluation of nanoemulsion

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Physical appearance

Formulation was examined for appearance which shows transparent formulation .They does not show any turbidity.

Evaluation data of nanoemulsion

Nanoemulsion was evaluated for uniformity of droplet size within the formulation is determined by PDI. The least PDI of formulation FE3 (0.269) indicating higher uniformity of droplet size distribution as compared to FE6 (0.664). Lowest zeta potential and PDI shows greater stability. Zeta potential of FE3 is (-9.05) hence it shows good stability. Rheology study of nanoemulsion formulation was carried out. Viscosity of formulations was found from range of 4320±1.421cps to 7960±1.241cps (table no.5). It is observed that increase in concentration of surfactant and co-surfactant leads to increase the viscosity while lowering the amount of surfactant and co-surfactant leads to decrease the viscosity.The drug content was determined for all the formulations by UV spectrophotometer method. The drug content of nanoemulsion gel was found in range of 87.50 ± 1.314% to 95.80 ± 1.121%.The higher drug content found in FE3 i.e.95.80 ± 1.121% and lower drug content found in FE1 i.e.87.50 ± 1.314% as shown in table no.5.

Thermodynamic stability

From the above table no.6 it was shows that formulation FE3, FE4 and FE6 are thermodynamically stable and formulation FE1, FE2 and FE5 are thermodynamically unstable.

In-vitro drug permeation study[39]

The in-vitro drug permeation of nanoemulsion was varied in amount according to droplet size of formulations as shown in figure no.8. The drug permeation from nanoemulsion in following ascending order FE1□ FE2□ FE5□ FE4□ FE6□ FE3. Where the amount of % drug permeated per cm^2 160.17□ 205.78□ 280.73□ 297.16□ 320.2□ 365.89. From the study it was shows that nanoemulsion with smaller particle size FE3 shows better drug release than other nanoemulsion within 10 hrs. (Kryscio et al., 2008)

Evaluation of gel

The pH of gel in between 6 to 6.5 which lies in between normal pH range of skin which does not produce any skin irritation as shown in table no.7

Evaluation of nanoemulgel

Nanoemulgel formulation evaluation data

The pH of nanoemulgel in between 6 to 6.5 which lies in between normal pH range of skin which does not produce any skin irritation. Formulation FG4 showed less viscosity i.e. 46800 cps and spreadability of formulation showed direct relationship with viscosity. If viscosity increases spreadability increases. (Kryscio et al., 2008). Here FG4 showed 30.5gm.cm/sec spreadability as shown in table no.8

In-vitro drug release study of nanoemulgel

Based on the pH, rheology, spreadability, drug content FG4 was selected as optimized nanoemulgel formulation and was further evaluated for drug release profiles. Amongst the formulations nanoemulsion showed in figure no. 9 drug release in following order FG4>FG8 (Marketed)>FG7>FG6>FG5>FG3>FG2>FG1. Where the amount of % drug release 78%>65.4%>59.76%>53.2%>49.93%> 40.88%>35.18%>20.22%. Here FG4 showed higher drug release i.e., 78% within 7 hrs and further drug release data was fitted in kinetic modeling and studied best fitted model and drug release profile of nanoemulgel formulation.

Kinetic modeling of drug release data

The in-vitro drug release data as shown in figure 10 to figure 14 of the formulation FG4 was fitted to various mathematical models, it showed linear nature between cumulative percentage drug released and time suggesting that it followed first-order kinetics. The best fit with higher correlation was found with first order R^2=0.996.

In-vitro drug permeation study

In-vitro drug permeation study of nanoemulgel and marketed formulation is shown in table no. 9 and figure no. 15

Antimicrobial activity study of nanoemulgel formulation

Minimum inhibitory concentration in formulation of nanoemulgel was studied using staph aureus as a microbial strain. Study was done for 48 hrs at 37±0.5 ^{0}C. Results are as shown in figure no.16

Evaluation of nanoemulgel as compared to marketed

Nanoemulgel formulation was evaluated for several evaluation parameters as compared to marketed to determine its effectiveness. Those evaluation parameters are as below (table no 10). pH

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of nanoemulgel formulation was found to be 6.48±0.16 and marketed formulation 6.49±0.546 which lies in between normal pH range of skin which does not produce any skin irritation. Spreadability of formulation showed direct relationship with viscosity. Viscosity of Nanoemulgel formulation was determined by Brookfield viscometer and it was found to be 46800±0.214 (cps). Formulation should be easily spreadable, hence spreadability of both formulations are determined using glass slides and it was found to be 54.5gm.cm/sec. for Nanoemulgel and 52.68 ± 0.652 for marketed formulation.
From these results we can say that Nanoemulgel offers good spreadability as compare to marketed gel.

IV. DISCUSSION

In present work, nanoemulsion of CUR was formulated by spontaneous emulsification method and characterized for particle size, polydispersity index, zeta potential, drug content and viscosity. Droplet size of all the formulated nanoemulsions was found to be in the nanoemulsion size range. Polydispersity index indicates homogeneous population of nanoemulsion droplet in formulation. All the formulations shows homogenous dispersion of droplets. FE3 formulation showed highest transdermal flux across cellophane membrane than other formulations. From the characterization study of nanoemulsions FE3 was selected as the optimized formulation which was formulated into nanoemulgel by using 1.5% carbopol-934 as hydrogel and aloe-vera gel in different concentrations and antimicrobial activity is determined by petri-plate method and zone of inhibition was calculated. FG4 shows maximum(8.14±0.21mm) zone of inhibition which was then compared with marketed (POVIDONE IODINE OINTMENT) for various parameters i.e. viscosity, extrudability and drug content. It was observed that nanoemulgel formulation (40±0.72µg/cm^2/hr) show two fold increase in transdermal flux as compared to marketed (17.37±0.52µg/cm^2/hr). From the results it can be concluded that nanoemulgel formulation is potential and effective transdermal drug delivery system for CUR and aloevera gel. Thus, it can be concluded that nanoemulgel was prepared to enhance solubility, permeability as well as bioavailability of curcumin. Nanoemulgel of CUR and aloe vera for the treatment of antimicrobial activity and is a safe, effective and promising formulation for the topical treatment of microbial infections.

V. CONCLUSION

In present study, CUR aloe vera nanoemulgel were prepared by spontaneous emulsification method. Drug excipient compatibility study was confirmed by FT-IR. Nanoemulsion was optimized on the basis of particle size, zeta potential drug permeation and flux. Further FE3 was incorporated into 1.5%carbopol 934 gel and eight nanoemulgel (FG1-FG8) formulations were prepared by cold method. Formulation FG4 shows maximum (8.14±0.21mm) zone of inhibition which was then compared with marketed (POVIDONE IODINE OINTMENT) for various parameters i.e. viscosity, extrudability and drug content. It was observed that nanoemulgel formulation (40±0.72 µg/cm^2/hr) show two-fold increase in transdermal flux as compared to marketed (17.37±0.52 µg/cm^2/hr). From the results it can be concluded that nanoemulgel formulation is potential and effective transdermal drug delivery system for CURand aloe-vera gel. Thus, it can be concluded that nanoemulgel was prepared with enhance solubility, permeability as well as bioavailability of CURand is a safe, effective and promising formulation for the treatment of microbial infections.

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REFERENCES

[1]. Ahmad N et al. A novel nano formulation development of eugenol and their treatment in inflammation and periodontitis (2019). Saudi Pharm. J. 17:778-790.
[2]. Ahmad N et al. A novel self-nanoemulsifying drug delivery system for curcumin used in the treatment of wound healing and inflammation (2019). Biotech 9:1-20.
[3]. Ahmed N et al. Preparation of a novel curcumin nanoemulsion by ultrasonication and its comparative effects in wound healing and

the treatment of inflammation (2019). The Royal Society of Chemistry 9: 20192–20206.
[4]. Ambekar AW, Nagaraju R. Development characterization and in-vitroevaluation of ethosomal gel for transdermal delivery of saxagliptin (2017). J. Global Trends Pharm Sci 8(4):4429-4437.
[5]. Ambekar AW, Nagaraju R, Subhash Chandra BP. Glibenclamide loaded ethosomal gel for transdermal delivery: formulation, optimization and ex- vivo study (2018).Asian J Pharm Pharmacol. 4: 630-636.
[6]. Bajjuri S, Dr.Vinod KR, Dr. Goud S. Formulation development and evaluation of topical curcumin emulgel (2018). Eur. J. biomed. Pharm. Sci. 5:399-405.
[7]. Balasubramani S, Moola AK, Vivek K, Ranjitha Kumari BD. Formulation of nanoemulsion from leaves essential oil of ocimum basilicum l. and its antibacterial, antioxidant and larvicidal activities (culexquinquefasciatus) (2018). Microbial patho. 125: 475-485.
[8]. Basera K, Bhatt G, Kothiyal P and Gupta P. Nanoemulgel: a novel formulation for topical delivery of hydrophobic drugs (2015). World J Pharm Pharm Sci. 4:1871-1886.
[9]. Bele MH, Shaikh AK, Paralkar SG. To enhance the solubility of curcumin by solid self-micro emulsifying drug delivery system (smedds) (2017). Indo Am. j. pharm. 7:8587-8607.
[10]. Cecchini ME et al. Nanoemulsion of minthostachys verticillata essential oil. In-vitro evaluation of its antibacterial activity (2021). Heliyon7: 1-8.
[11]. Chandrasekaran N et al. Preparation and characterization of edible oil nanoemulsions for enhanced stability and oral delivery of curcumin (2018). Int J App Pharm. 10:139-146.
[12]. Conxita S, Isabel S. Nano-emulsions: Formation by low-energy methods (2012). Curr Opin Colloid Interface Sci. 2:246–254.
[13]. Enas M, Ahmed. Hydrogel: Preparation, characterization, and applications: A review (2015) . J. Adv. Res 6:105-121.
[14]. Gadkari PN, Patil PB, Saudagar RB. Formulation, development and evaluation of topical nanoemulgel of tolnaftate (2019). J. drug deliv. ther. 9:208-213.
[15]. Hardenia A, Jayronia S andJain S.Emulgel: an emergent tool in topical drug delivery (2014).Int. J. Pharm. Sci. Res. 5:1653-1660.
[16]. Haritha, Syed B, Rao K, Chakravarthi V. A brief introduction to methods of preparation, application, and characterization of nanoemulsion drug delivery system. Indian j. res. Pharm. Biotech. 1:25-28.
[17]. Higuchi T. Mechanism of sustained action medication: Theoretical analysis of rate of release of solid drugs dispersed in solid matrices (1963). J. Pharm. Sci 52:1145-1149.
[18]. Javed A, Amin S, Kohli K, Mir SR .Construction of pseudo ternary phase diagram and its evaluation: development of self-dispersible oral formulation (2013) . Int. J. Drug Dev. Res.5: 84-90.
[19]. Kadam PV, Yadav KN, Bhingare CL, Patil MJ. Standardization of curcumin from curcuma longa extracts using UV visible spectroscopy and HPLC (2018). J. pharmacogn. phytochem..7:1913-1918.
[20]. Kaur LP, Guleri TK. Topical Gel: A Recent Approach for Novel Drug delivery (2013). Asian j. biomed. pharm. 3: 10-15.
[21]. Kryscio DR et al. Spreadability measurement to assess structural equivalence (Q3) of topical formulations- A technical note (2008). AAPS PharmSciTech 9:84-86.
[22]. Kumar D, Singh J, Antil M and Kumar V (2016). Emulgel-novel topical drug delivery system–a comprehensive review. Int. J. Pharm. Sci. Res. 7: 4733-4742.
[23]. Kumar M, Kaur P, Garg R, Patil R, Patil H. A study on antibacterial property of curcuma longa-herbal and traditional medicine (2020). Adesh. Uni. J. Medi. Sci. Res. 2:103-108.
[24]. Kumar M, Bishnoi RS, Shukla AK, Jain CP. Techniques for formulation of nanoemulsion drug delivery system: a review (2018). Prev. Nut. Food 24: 225–234.
[25]. Kute S, Saudagar R. Emulsified gel anovel approach for delivery of hydrophobic drugs: an overview (2013). J. Adv. Pharm. Educ. 3: 368-376).
[26]. Mishra RK, Soni GC, Mishra RC. A review article: on nanoemulsion (2014). World J Pharm Pharm Sci. 9:258-274.
[27]. Naz F, Ahmad FJ. Curcumin loaded colloidal

carrier system: formulation optimization, mechanistic insight, ex vivo and in vivo evaluation (2015).Int. J. Nanomedicine 10:4293-4307.

[28]. Patel NA, Patel NJ, Patel RP.Formulation and evaluation of curcumin gel for topical application (2008).Pharma. Dev. Tech.10:1-10.

[29]. Ritger PL, Peppas NA. A simple equation for description of solute release 1. Fickian and non-fickian release from non-swellable devices in the form of slabs, spheres, cylinders or discs (1987). J cont rel. 5: 23-36.

[30]. S Padsalg A, Patel K, Mokale V. Formulation, development and evaluation of Fluconazole gel in various polymer bases (2007). Asian J. Pharm. Sci. 1: 63-68.

[31]. Sanjay J, Pawan B, Shailendra M, Prashant G. A review on emulgel, as a novel trend in topical drug delivery system (2019). Recent Trends Pharma. Sci. Res. 1:30-39.

[32]. Sankha B, Prajapati BG. Formulation and optimization of celecoxib nanoemulgel (2017). Asian J Pharm Clin Res. 10:353-365.

[33]. Savale KS. Curcumin as a model drug: conformation, solubility estimation, morphological, in-vitro and in-vivo bio distribution study (2017). J. pharm. Sci. tech.7:31-35.

[34]. Shadab MD et al. Improved analgesic and anti-inflammatory effect of diclofenac sodium by topical nanoemulgel: formulation development- in vitro and in vivo studies (2020). J. Chem. 1-10.

[35]. Shah A, Thakkar V, Gohel M, Baldaniya L, Gandhi T. Optimization of self-micro emulsifying drug delivery system containing curcumin and artemisinin using d-optimal mixture design (2017). Saudi J. Med. Pharm. Sci 3: 388-398.

[36]. Shailendra P, Sayantan M, Preeti K. Emulgel: a novel approach for topical drug delivery system (2015). World J. Pharm. Res.4:209-223.

[37]. Syed HK, PEH KK. Antibacterial activity of curcumin and solubility-enhanced curcumin in microemulsion: a comparative study (2018). Lat. Am. J. Pharm. 37:1468-77.

[38]. Sugumar S, Ghosh V, Nirmala MJ, Mukherjee A, Chandrasekaran N. Ultrasonic emulsification of eucalyptus oil nanoemulsion: antibacterial activity against staphylococcus aureus and wound healing activity in wistar rats (2014). Ultra. Sono. Chem. 21:1044-1049.

[39]. Kaur LP, Guleri TK. Topical Gel: A Recent Approach for Novel Drug delivery (2013). Asian j. biomed. pharm. 3: 10-15.

Table

Table no.1 List of equipment's

Sr. no.	Equipments	Manufacturer
1	Electronic Balance	Shimadzu
2	UV-Visible Spectrophotometer	Jasco (V-630)
3	FT-IR	Bruker FTIR (alpha 2), Shimadzu
4	Zeta sizer	Malvern
5	Incubator shaker	Electro Lab
6	Centrifugation Machine	Remi Instrument Ltd
7	Magnetic stirrer	Remi Instrument Ltd
8	Homogenizer	Remi Instrument Ltd
9	Viscometer	Brook field Viscometer
10	pH meter	Eutech Instrument Oaktan
11	Incubator	Remi Instrument Ltd
12	Digital antibiotic zone reader	Chemiline

Table no.2 Manufacturing formula for nanoemulsion formulations

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Formulation code	CUR (mg)	IPM (ml)	Tween 80 (ml)	PEG400 (ml)	Water (ml)	TEA
FE1	100	26.70	20	20	33.3	q.s
FE2	100	23.34	21.68	21.68	33.3	q.s
FE3	100	20	23.35	23.35	33.3	q.s
FE4	100	16.66	25.2	25.2	33.3	q.s
FE5	100	13.34	26.68	26.68	33.3	q.s
FE6	100	10	28.35	28.35	33.3	q.s

Table no. 3 Manufacturing formula for gel base

Formulation code	Carbopol 934 (%)	Methyl paraben (%)	Propyl paraben (%)
G1	1	0.2	0.2
G2	1.5	0.2	0.2

Table no. 4 Manufacturing formula for Nanoemulgel

Formulation code	Nanoemulsion (gm)	Gel base (gm)	Aloe vera (gm)
FG1	30	30	-
FG2	30	30	30
FG3	30	30	60
FG4	30	30	120
FG5	-	30	30
FG6	-	30	60
FG7	-	30	120
FG8	Marketed formulation		

Table no.5 Evaluation data of nanoemulsion

Formulation code	Droplet size (nm)	Poly dispersity index (PDI)	Zeta potential (mv)	Viscosity (cps)	Drug Content (%)	Flux µg/cm^2/hr
FE1	146.2	0.436	-2.80	4320±1.421	87.50±1.314	28.23
FE2	111.4	0.493	-5.81	4440±1.451	88.90±2.132	42.28
FE3	58.00	0.269	-9.05	4870±1.412	95.80±1.121	56.06
FE4	96.64	0.287	-6.39	6920±1.952	90.20±2.214	47.61
FE5	102.0	0.559	-3.82	7340±1.272	92.10±1.352	43.59
FE6	30.33	0.664	-5.16	7960±1.241	93.50±1.361	48.25

Values are expressed in mean ± SD, where n=3

Table no.6Thermodynamic stability study

Formulation code	Heating cooling cycle	Centrifugation	Freeze thaw cycle	Inference
FE1	×	√	×	Failed
FE2	×	√	×	Failed
FE3	√	√	√	Passed
FE4	√	√	√	Passed
FE5	×	√	×	Failed
FE6	√	√	√	Passed

Table no.7Evaluation of gel

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Formulation code	pH	Rheology
GF1	6.47	49400±2.312
GF2	6.49	55700±1.314

Values are expressed in mean ± SD, where n=3

Table no. 8 Nanoemulgel formulation evaluation data

Formulation code	Evaluation Parameter		
	pH	Rheology (Cps)	Spreadability gm.cm/sec
FG1	6.49±1.20	48900±1.121	26.6±1.21
FG2	6.27±2.18	50500±1.101	29.7±2.19
FG3	6.43±2.15	49200±2.130	24.5±1.18
FG4	6.48±1.16	46800±2.214	30.5±2.17
FG5	6.47±1.17	47800±2.254	26.6±2.20
FG6	6.50±1.19	50300±1.221	22.30±3.22
FG7	6.29±1.20	50400±1.231	25.6±2.12

Values are expressed in mean ± SD, where n=3

Table no.9 In-vitro drug permeation study

Time (Hrs)	Nanoemulgel formulation	Marketed formulation
0	0	0
1	60.54	45.44
2	118.58	90.58
3	172.79	135.6
4	225.56	178.45
5	275.79	215.78
6	330.58	255.25
7	380.48	298.68
8	423.78	345.58
9	452.89	375.45
10	475.89	395.78
Flux = 40(µg/cm^2/hr)		17.37

Table no.10 Evaluation of nanoemulgel as compared to marketed

Sr. No.	Evaluation Parameters	Nanoemulgel	Marketed formulation
1	pH	6.48±1.16	6.49±1.546
2	Viscosity(cps)	46800±2.214	50876±2.262
3	Spreadability(gm.cm/sec)	30.5 ± 2.17	29.68± 1.27
4	Zone of Inhibition(mm)	8.25±1.21	7.0±1.11
5	Flux(µg/cm^2/hr)	40±1.72	17.37±1.52

FIGURE

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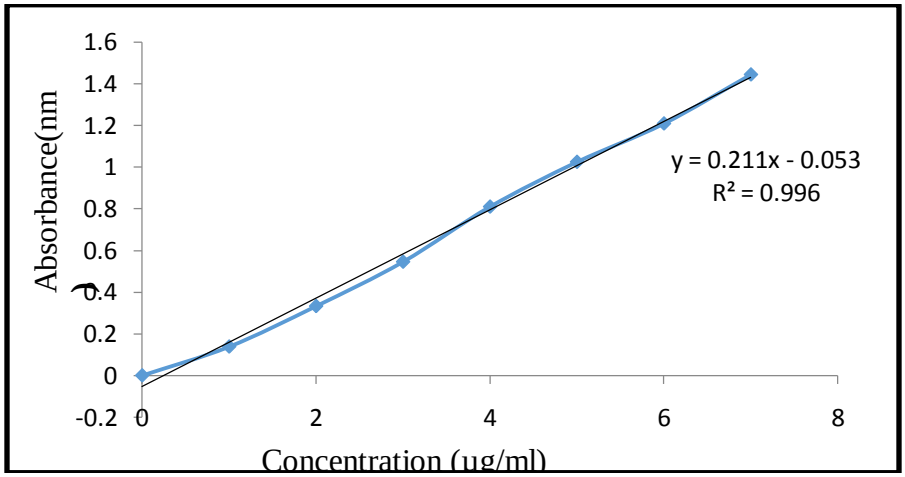

Figure no.1 Determination of λ_{max} and calibration curve of CUR in phosphate buffer pH 7.4

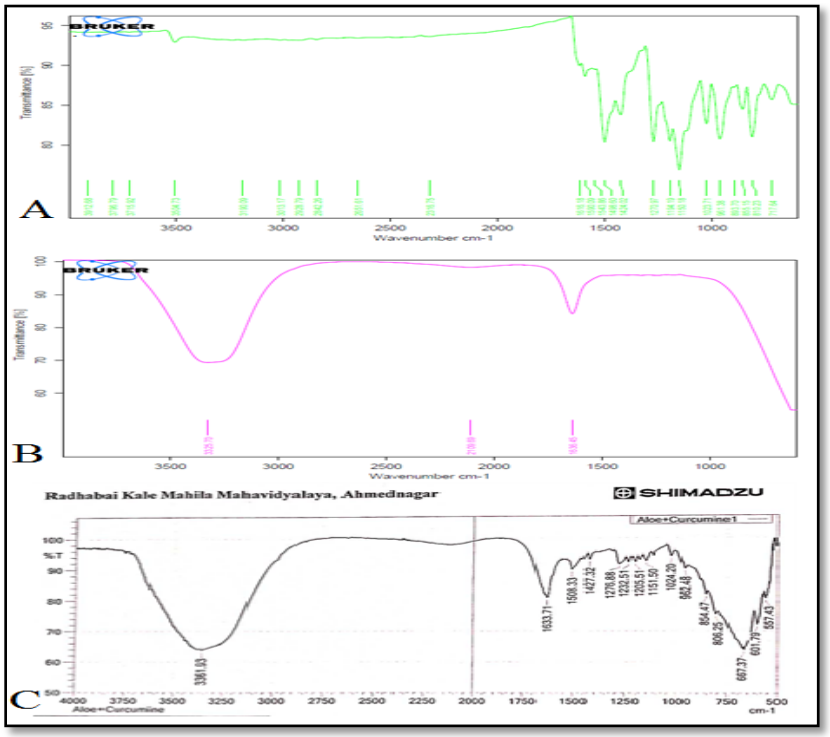

Figure no.2 FTIR spectra of- A)CURB) Aloe C)CUR + aloe vera

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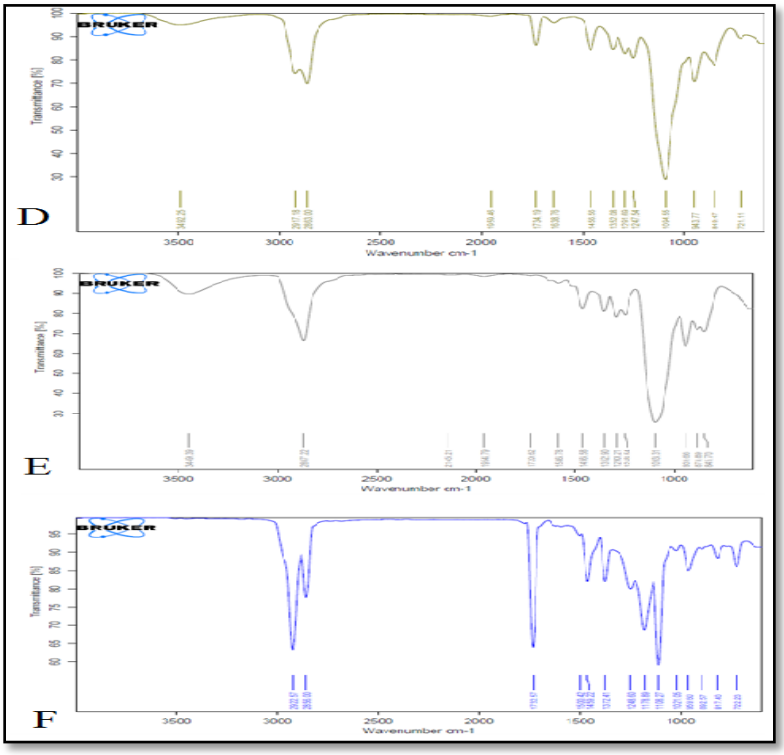

Figure no.3 FTIR spectra of D) CUR +Tween 80 E)CUR +PEG40

A) CUR +IPM

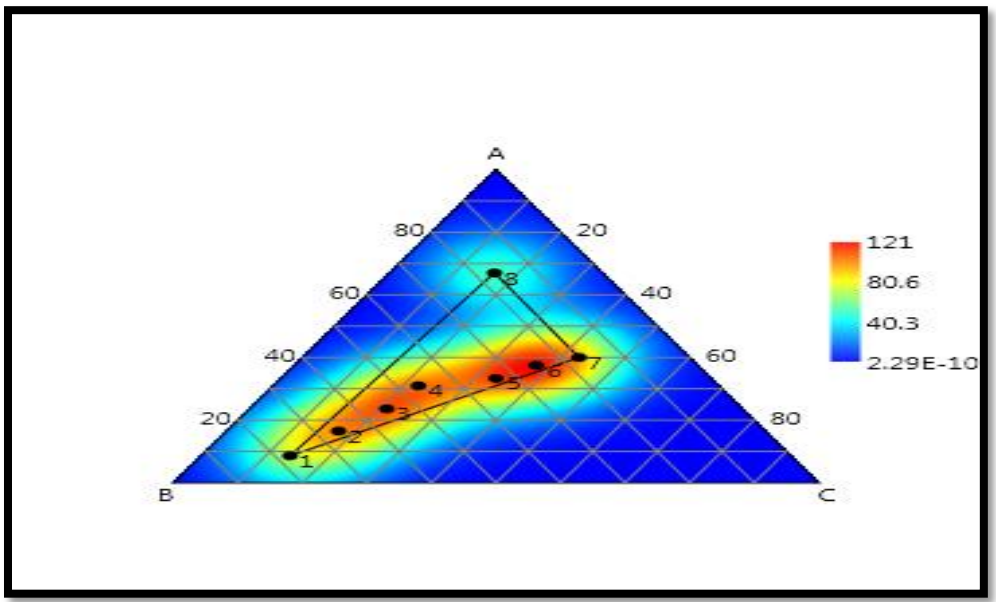

Figure no.4 Construction of pseudo-ternary phase diagram

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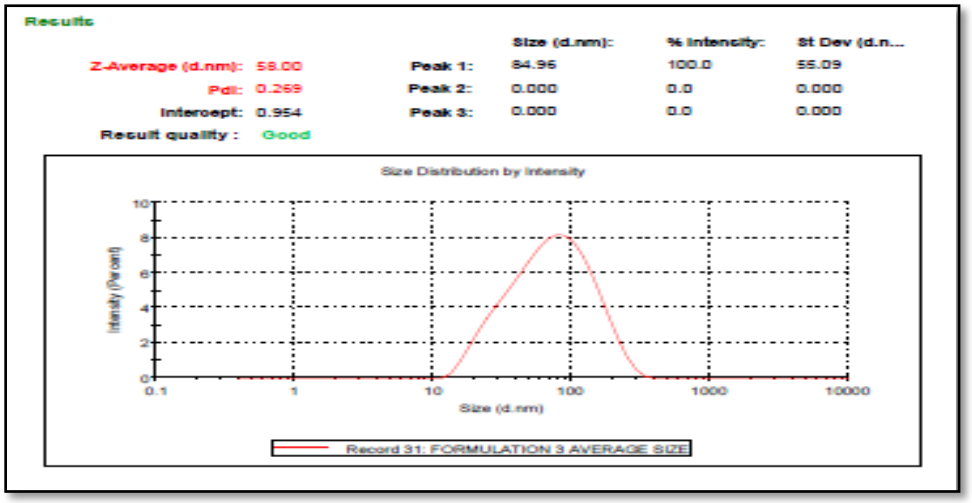

Figure no.6 Particle size analysis of formulation FE3

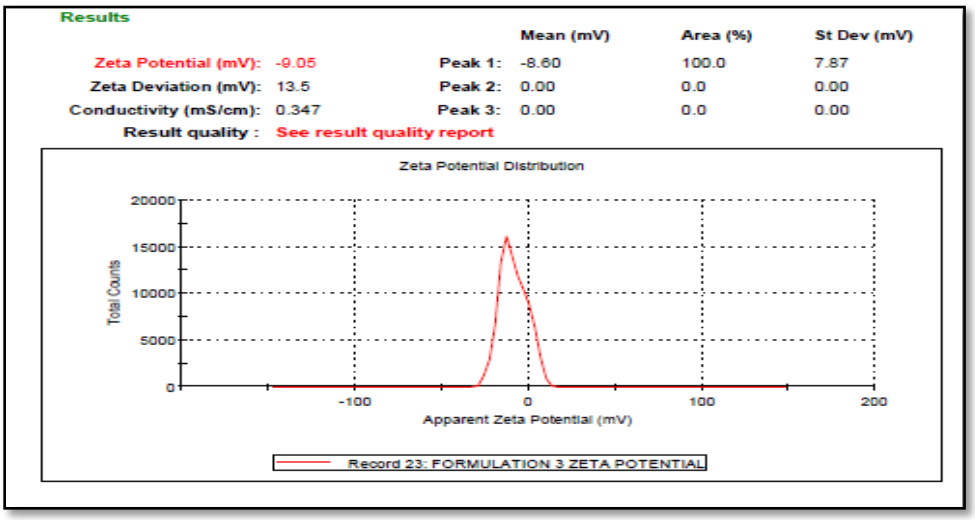

Figure no.7 Zeta potential of formulation FE3

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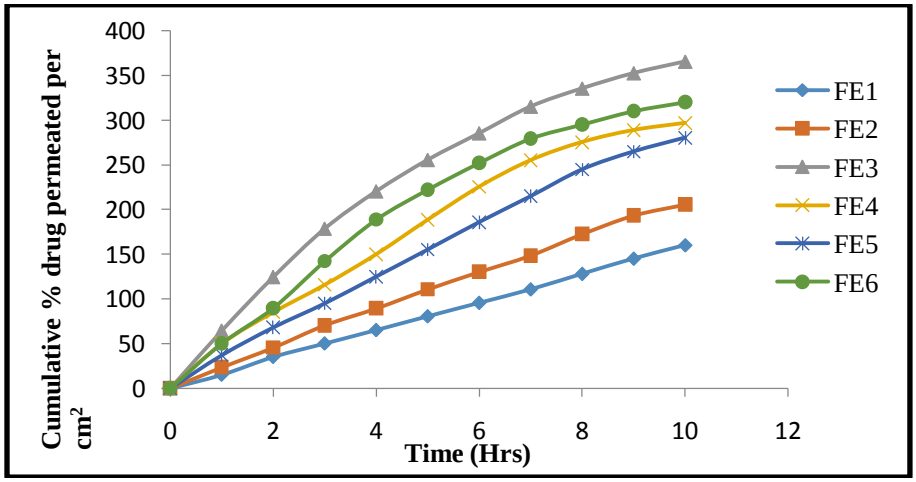

Figure no. 8 In-vitro drug permeation study

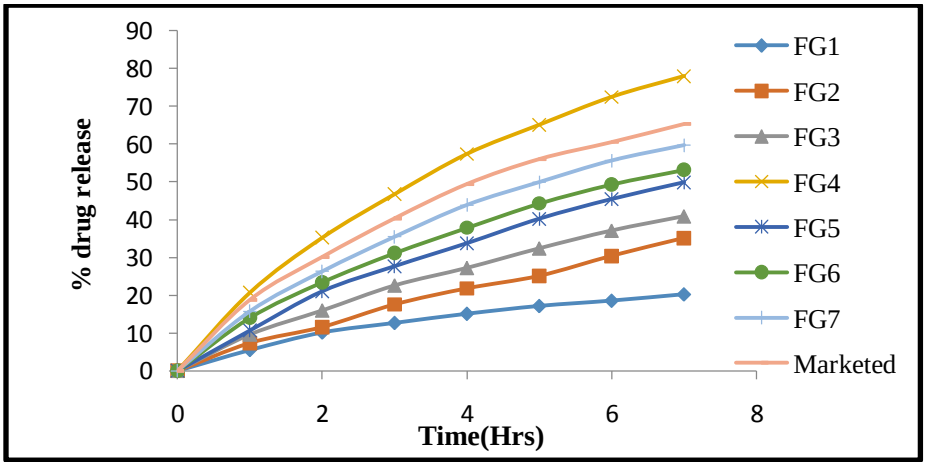

Figure no.9 In-vitro drug release study of nanoemulgel

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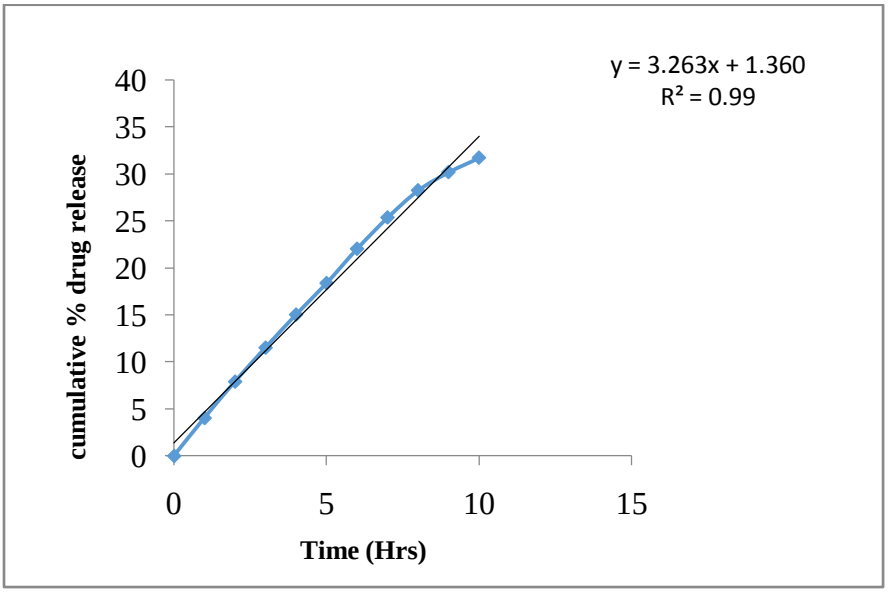

Figure no.10 Zero order model

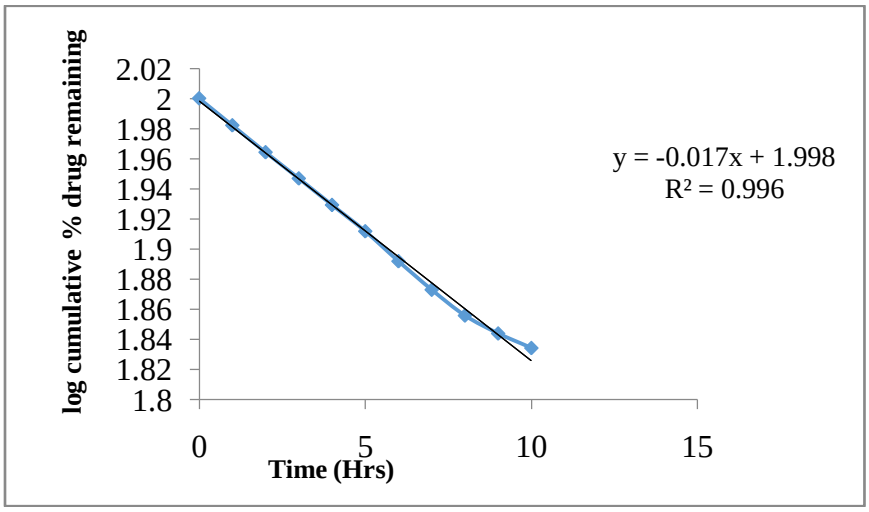

Figure no.11 First order model

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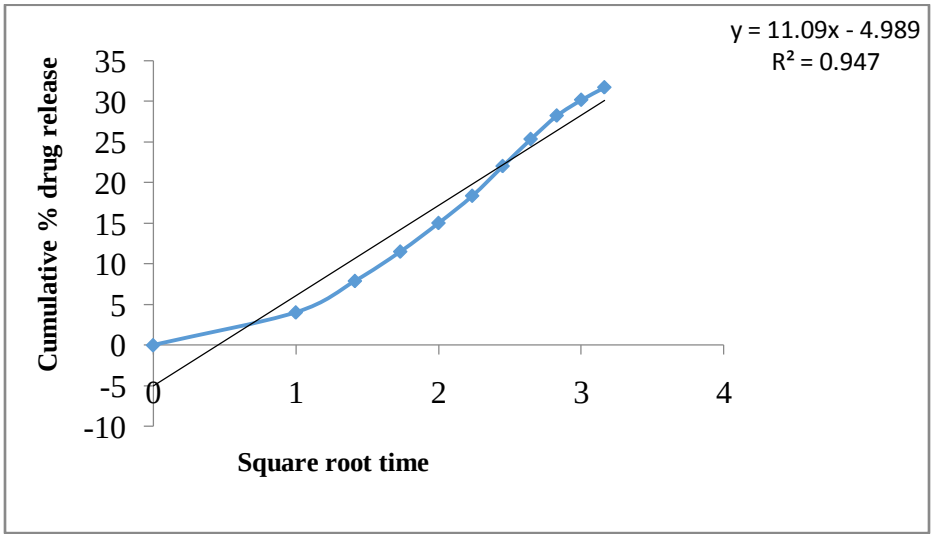

Figure no. 12 Higuchi model

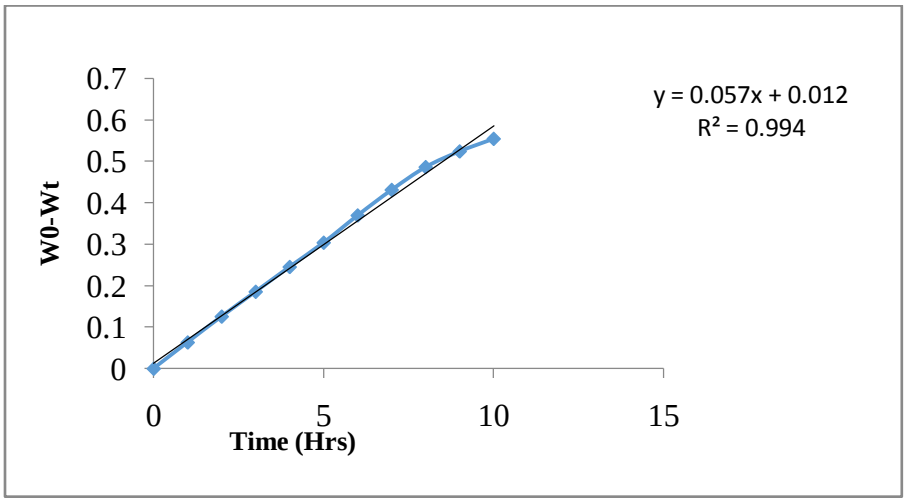

Figure no.13 Hixson Crowell model

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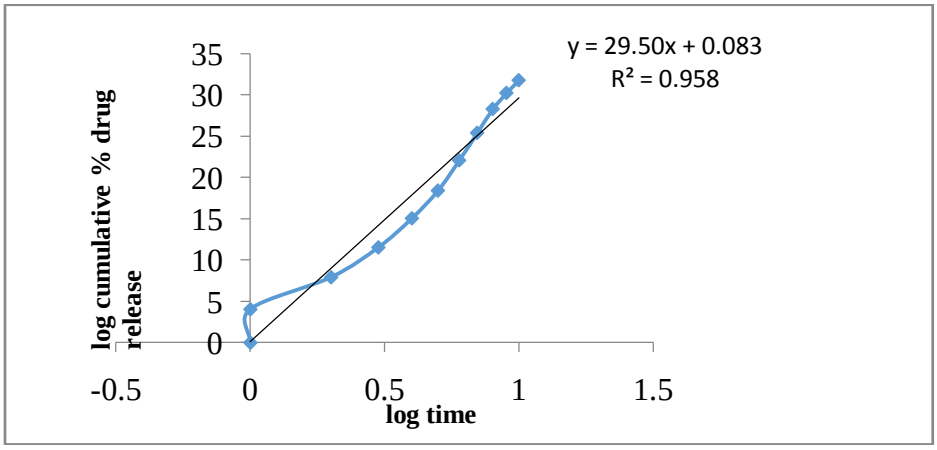

Figure no.14Korsmeyer- peppa's model

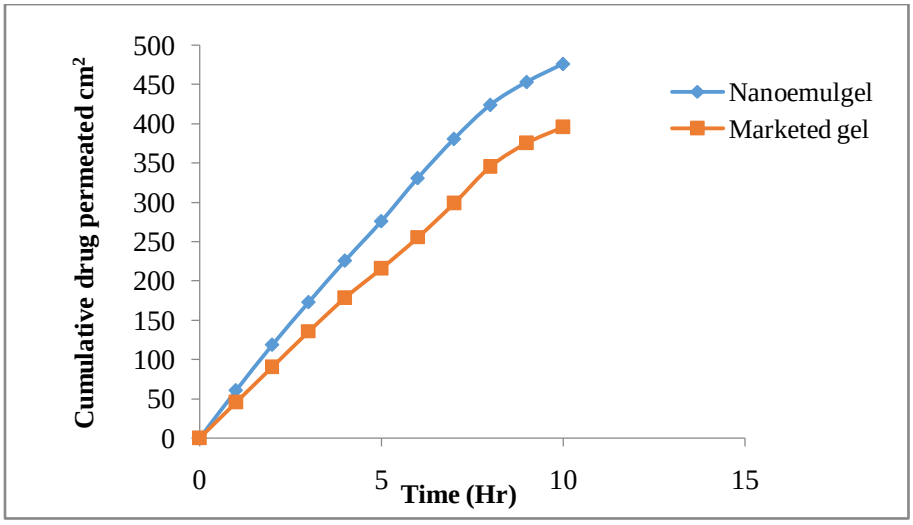

Figure no.15Invitro drug permeation study

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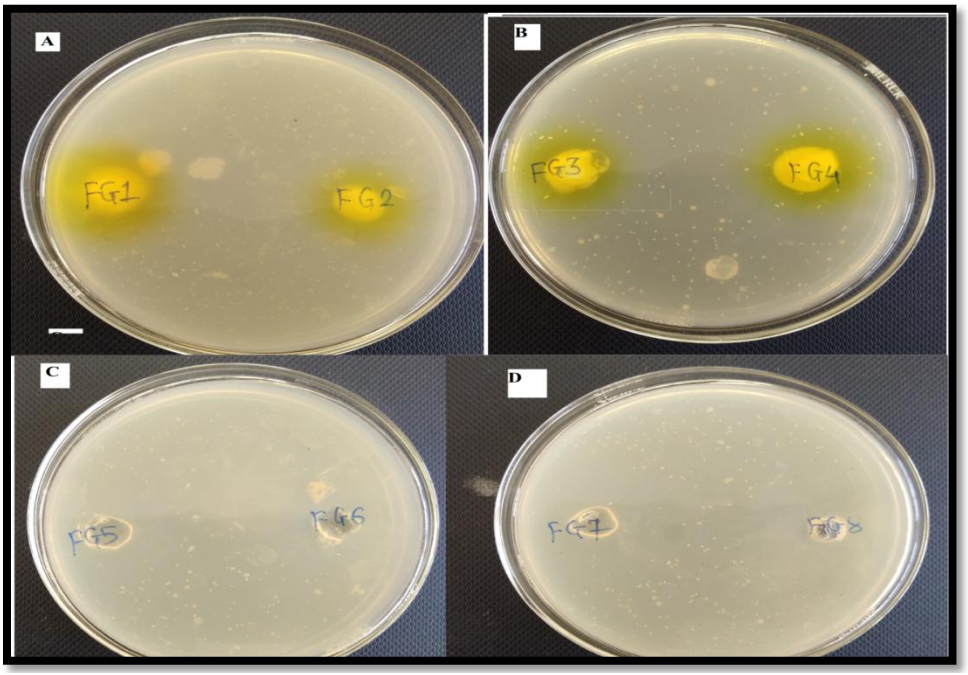

Figure no.16 Zone of inhibition study against Staphylococcus aureus

Note*: FG1 = NE, FG2= NE + Aloe vera (30 gm), FG3: NE + Aloe vera (60 gm), FG4 = NE + Aloe vera (120 gm), FG5 = Aloe vera (30 gm), FG6 = Aloe vera (60 gm), FG7 = Aloe vera (120 gm), FG8 = Marketed formulation