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Research Article

COMPARATIVE QUALITY EVALUATION OF MARKETED FORMULATION OF DICLOFENAC DIETHYLAMINE GEL

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Abstract

Diclofenac diethylamine (DDA), a non-steroidal anti-inflammatory agent is frequently prescribed for the long term treatment of rheumatoid arthritis, osteoarthritis and ankylosing spondylitis. Three different marketed DDA gel formulations of different brands were evaluated to check their critical quality control parameters. Different brands of formulations were characterized for their drug diffusion study, texture analysis profile, viscosity, pH etc. The statistical methods like correlation coefficient, confidence interval (95% CI) were applied for their dosage form performance aspect in comparision with reference product.The results of all three brands revealed that Brand B is superior to Brand A and Brand C in terms of drug diffusion properties (96.8%, 50min) in comparision with reference product profile, texture analysis (TPA-450 and Compresion- 550) and viscosity (2300cP).

Keywords: Diclofenac diethylamine Gels, Viscosity, Texture profile analysis, Diffusion study, Comparative evaluation.

Introduction:

In-vitro performance tests for solids, such as dissolution tests, have been extensively used as a tool in the development of drug formulations, in quality control procedures to ensure batch-to-batch uniformity, to monitor changes in the process and formula and to predict the in-vivoperformance of the product. A new test to evaluate the performance of semisolid products with the same aims as the dissolution test has been the subject of extensive discussion [1]. Currently, the performance testing system employing the vertical diffusion cell (VDC) is commonly applied to semisolid products, specifically creams, ointments and gels, and also to lotions. This procedure quantifies the release of the active component from the formulation, which diffuses through a membrane into a receptor solution [2]. In 2009, the United States Pharmacopeia Forum suggested the use of VDCs with synthetic membranes to test the performance of topical products [2]. In vitro release is one of several standard methods which can be used to characterize performance characteristics of a finished topical dosage form, i.e., semisolids such as creams, gels, and ointments. Important changes in the characteristics of a drug product formula or the thermodynamic properties of the drug(s) it contains should show up as a difference in drug release. Release is theoretically proportional to the square root of time (/t) when the formulation in question is in control of the release process because the release is from a receding boundary (SUPAC-SS, CMC 7). The aim of the present study was comparative evaluation of different marketed Diclofenac diethyl gel formulations (Brand A, Brand B and Brand C) for their various physicochemical properties like pH, viscosity, texture profile analysis, drug diffusion study along with reference product as per Center for Drug Evaluation and Research (CDER) guidelines and requirenments. This study will come out with best dosage form performance based on above criteria studied.

Material and Methods:

Diclofenac diethylamine Gel formulations of different brands were purchased from pharmacy market. Franz diffusion cell (FDC-08, Orchid Scientific, Nashik, Maharashtra) was used for drug diffusion. pH was determined using pH meter (Type-362, Systronic India Ltd., Ahmedabad), Brookfield viscometer (DVE) was used to determine viscosity. Texture analysis was performed using (CT3 Brookfield texture analyzer), UV visible spectrophotome-

ter (A SHIMADZU 1650 Tokyo, Japan) with 10mm quartz cuvette. Diclofenac diethylamine was provided as a gift sample from Cadila Health Care, Ahmedabad, Gujarat. All reagents and chemicals used were of analytical grade.

Calibration curve of Diclofenac diethylamine

10mg of Diclofenac diethylamine drug is taken in 100ml of phosphate buffer 7.4 in volumetric flask. Serial dilutions of drug with phosphate buffer 7.4 were preparedand analyzed by UV spectrophotometer at 276nm and calibration curve was plotted using different absorbance values.

Characterization of different Brands of DDA gels formulation:

pH determination of gel formulations: The pH was measured for each gel. The electrode was inserted in to the sample for 10 min prior to taking the reading at room temperature [4,5,11].

Viscosity determination: The viscosity determination was carried out using a Brookfield Viscometer (DVE+ Pro model) using spindle number S-64 at 6 rpm at a temperature of 25±1.0º C. The determinations were carried out in triplicate and the average of three readings was recorded[5].

Spreadability: It is expressed in terms of time in seconds taken by two slides to slip off from gel and placed in between the slides under the direction of certain load, lesser the time taken for separation of two slides, better the spreadability[6,8,13]. It is calculated by using following formula:

S = M. L / T

Where, M = weight tied to upper slide

L = length of glass slides

T = time taken to separate the slides.

Grittiness: Smears of gels were prepared on glass slide and observed under the microscope for the presence of any particle or grittiness [7].

Homogeneity: All developed gels were tested for homogeneity by visual inspection after the gels have been set in the container[7,11].

Assay of formulations: Drug content in gel was estimated by dissolving known quantity of gel (1g) in suitable quantity of phosphate buffer pH 7.4. Aliquots of different concentrations were prepared by suitable dilutions after filtering the stock solution and absorbance was measured at 276nm using spectrophotometer [13].

Texture Profile analysis: Texture profile analysis (TPA) of prepared gel was carried out by using Brookfield texture analyzer (CT3). TPA is a set of definitions relating the sensory properties of a product to the instrumental properties, which can be calculated from the results of a two cycle texture profile analysis test. Texture analyzers perform this test by applying controlled forces to the product and recording its response in the form of force, deformation and time.

The textural analysis was performed in the compression mode in a CT3 texture analysis by carrying out a penetration test using a cylindrical probe (13mm diameter), a penetration depth of 15 mm and velocities of 3 mm/s. After penetrating the sample, the probe returned to a position 30mm above the platform surface. From the graphic force versus distance obtained, the maximum force and negative force were noted. These parameters were correlated with firmness and adhesiveness, respectively. All the measurements were performed in triplicate[9,10].

Drug diffusion study: Phosphate buffer of pH 7.4 was used for in vitro release as a receptor medium. The pretreated skin of albino mice(or cellophane membrane) was used in diffusion cell. The gel sample was applied on the skin(or cellophane membrane) and then fixed in between donor and receptor compartment of diffusion cell. The receptor compartment contained phosphate buffer of pH 7.4. The temperature of diffusion medium was thermostatically controlled at 37 ± 1ºC by surrounding water in jacket and the medium was stirred by magnetic stirrer.The sample at predetermined intervals were withdrawn and replaced by equal volume of fresh fluid. The samples withdrawn were spectrophotometrically estimated at 276nm against their respective blank[11].

Various models were tested for explaining the kinetics of drug release. To analyze the mechanism of the drug release rate kinetics of the dosage form, the obtained data were fitted into zero-order, first order, Higuchi, Hixon-Crowell model and Korsmeyer-Peppas release model [7].

Zero order release rate kinetics

To study the zero–order release kinetics the release rate data are fitted to the following equation:

F=K0.t…… (Eq. 1)

Where 'F' is the drug release, 'K0' is the release rate constant and't' is the release time. The plot of percentage drug release versus time is linear.

First order release rate kinetics

The release rate data were fitted to the following equation:

Log (100 – F) = K t…… (Eq. 2)

A plot of log % drug release versus time is linear.

Higuchi release model

To study the Higuchi release kinetics, the release rate data were fitted to the following equation, where, 'k' is the Higuchi constant. In higuchi model, a plot of percentage drug release versus square root of time is linear.

$F1 = Q = K t^{½}$…… (Eq. 3)

Where, 'k' is the Higuchi dissolution constant.

Hixon-Crowell model

To study the Hixon-Crowell release kinetics, the release rate data were fitted to the following equation:

W01/3 – Wt1/3 = k t…… (Eq. 4)

Where, 'Wo' is the original mass/weight of drug, 'Wt' is the mass/weight at't' time, 'k' is Hixon-Crowell constant. In this model (Wo 1/3 – Wt 1/3) versus time is linear.

Korsmeyer and Peppas release model

The release rate data were fitted to the following equation, Where, Mt /Mμ is fraction of drug released 'k' is the release constant, 't' is the release time, 'n' is diffusion exponent, if n is equal to 0.89, the release is zero order. If n is equal to 0.45 the release is best explained by Fickian diffusion, and if 0.45 < n < 0.89 then the release is through anomalous diffusion or non-fickian diffusion (swellable and cylinder Matrix). In this model, a plot of log (Mt/Mμ) versus log (time) is linear. The data from In-Vitro Drug diffusion studies of gels was fitted to Zero-order, First release.

Statistical comparison of the in vitro drug diffusion data

The statistical tests were used to compare the in-vitro release rates is a non-parametric statistical method, based on a standard confidence interval (CI) procedure. This test is related to the Wilcoxon Rank Sum/Mann-Whitney rank test, applied to the data for the log of the slope (release rate). For the release rate of the Brand (B) run to be within the 90% CI in relation to the reference (R) test run, the values for the B/R ratio should lie within the limits of 75% - 133% (FDA, 1997)[14].

Result and Discussion:

Standard calibration curve along with their values of DDAwereshown inTable 1and Figure 1, respectively. Calibration curve was plotted by taking concentration of drug (5-30μg/ml) on x-axis and on y-axis absorbance. The curve is linear and follow beers' and Lambert's law.

Table 1 : Standard plot values of Diclofenac diethylamine

Sr. No.	Concentration (μg/ml)	Absorbance
1	0	0
2	5	0.144
3	10	0.29
4	15	0.491
5	20	0.655
6	25	0.824
7	30	0.921

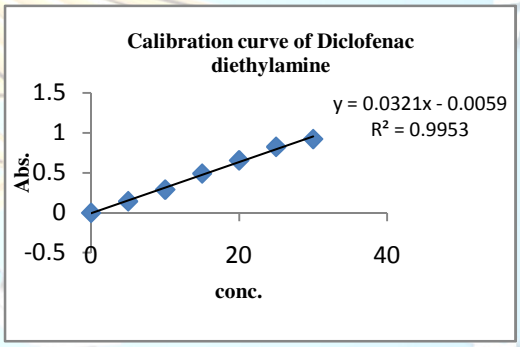

Figure 1: calibration curve of Diclofenac diethylamine in phosphate buffer pH 7.4

The pH value of all gel was in range of 5-6 which is considered acceptable to avoid the risk of irritation upon application to the skin.The results are summarized in Table 2.

Viscosity study results were mentioned in Table 2. It indicated that Brand A showed viscosity value of 4933 cP, for Brand B it was 2300 cP and for Brand C the value was 7400 cP. The results revealed that Brand C shows more viscosity as compare to Brand A and Brand B. The possible reason behind variation in viscosity is because of different viscosity forming agents used in the various marketed formulation and long term storage. Based on results, Brand B is showing moderate viscosity which is good for its consistency and applicability.

The spreadability is very much important characteristics of gel formulation which impart the flow of gel formulation from container. The values of spreadability were shown in Table 2. The Brand B are having higher spreaded value (2) and as compare to Brand A (1.8) and Brand C (1.5) respectively. It shows that Brand B was found to be good.
Results of homogeneity and grittiness are given in Table 2. All the gels were found to be homogenous with absence of lumps. All the gels were clear and transparent. Grittiness was observed in none of the gels.
Result of drug content analysis of different marketed formulations was shown in Table 2. The drug content of the gel was estimated and results were within the official limits of 90 to 110% (schedule C, D and C act 1945 for the different gel formulations). The drug content determination of Brand B showed 96.73% while it was 89.11% for Brand A and 85.4% for Brand C. It shows Brand C shows failing for the assay.

Table 2: Assessment of various parameters for different marketed formulation of Diclofenac diethylamine gel

Formulation	pH	Viscosity	Spreadability	Homogeneity	Grittiness	Drug content
Brand A	6	4933	1.8	+++	-	89.11
Brand B	5.5	2300	2	+++	-	96.73
Brand C	5.7	7400	1.5	++	-	85.4

+ Satisfactory, ++ good, +++ Very good; - no grittiness

The result for TPA of all three brands is given in Figure 2, 3 and 4, respectively. TPA results of gel showed that the area under the positive curve is a measure of the energy required to deform the sample to the defined distance. Texture profile analysis (TPA) was performed using a CT3 Texture Analyzer in TPA mode. At least five replicate analyses of each sample were performed at temperatures of 25 °C and 35°C. From the resulting force–time plots, the hardness (the force required to attain a given deformation), compressibility (the work required to deform the product during the first pass of the probe) and adhesiveness (the work necessary to overcome the attractive forces between the surface of the sample and the surface of the probe) were derived. As per results obtained, TPA for Brand B (450) is superior to the brand A (550) and brand C (2000) because Brand B peak value was less than Brand A and Brand C. The results for compression mode of texture of all three brands were mention in Figure 5, 6 and 7 for Brand A, B and C, respectively.

Spreadability test was performed by using CT3 Texture Analyzer in Compression mode. The maximum force value on the graph is a measure of the firmness of the sample at the specified depth. The area under the positive curve is a measure of the energy required to deform the sample to the defined distance (Hardness Work Done). Vishal Pande et al. 2014 have reported [17] that the firmness and energy required deforming a sample to a defined depth grades samples in order of spreadability. A higher peak load (firmness) and hardness work done value indicate a less spreadable sample. Conversely, a lower peak load (firmness) value coupled with a lower hardness work done value indicates a more spreadable sample. As per results obtained, compression for Brand B (550) is superior then the brand A (8000) and brand C (800) because Brand B peak value was less than Brand A and Brand C.

Texture profile analysis and Compression:

In-vitro drug release study was included with square root of time function and the same procedure is necessary for the linearization of the data and for better predictivity with high degree of accuracy (Higuchi, 1962; Guy, Hadgraft, 1990; Bemvindo, 2006). The representation of the amount released per unit area (μg/cm^2) as a function of the square root of time allows the drug release rate for a semisolid product to be obtained from the straight line slope (FDA, 1997) [20]. According to Higuchi (1962), there is a linear relationship between the amount of drug released/diffused and √t when the diffusion through this semisolid matrix is the limiting step of the process and in these circumstances no other parameter (e.g. receptor medium, membrane) has a significant effect. Also, this

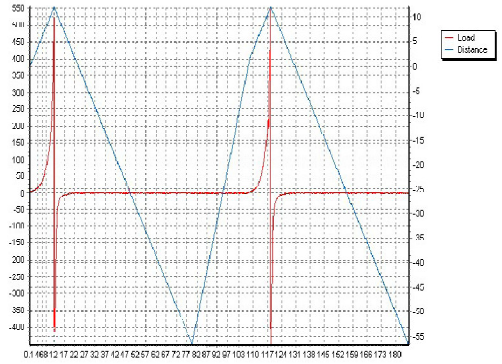

Figure 2: Brand A

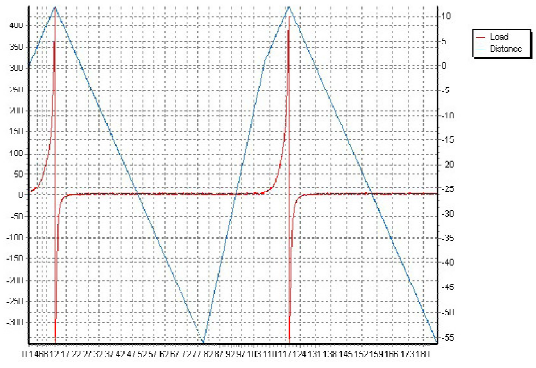

Figure 3: Brand B

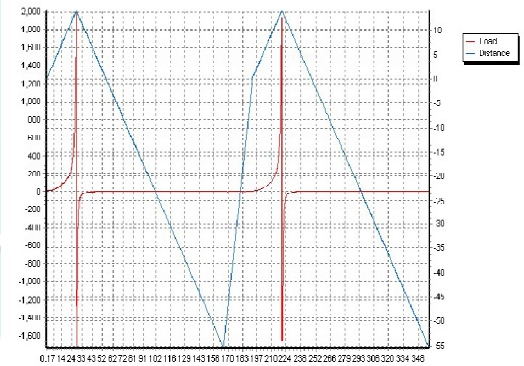

Figure 4: Brand C

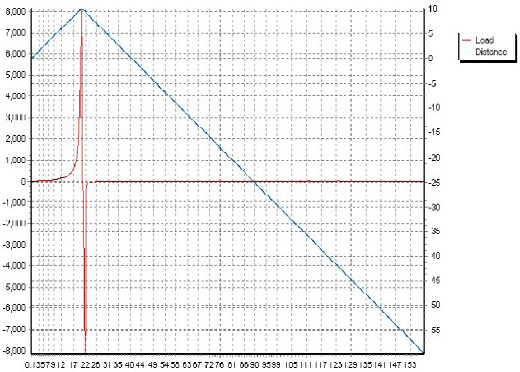

Figure 5: Brand A

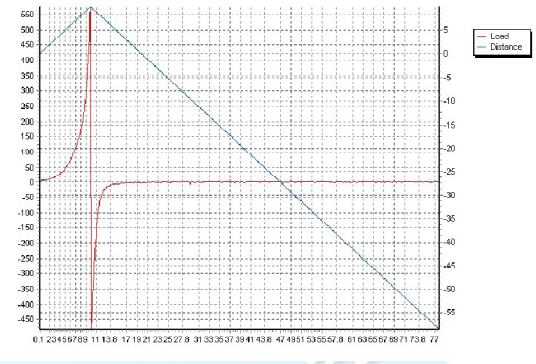

Figure 6: Brand B

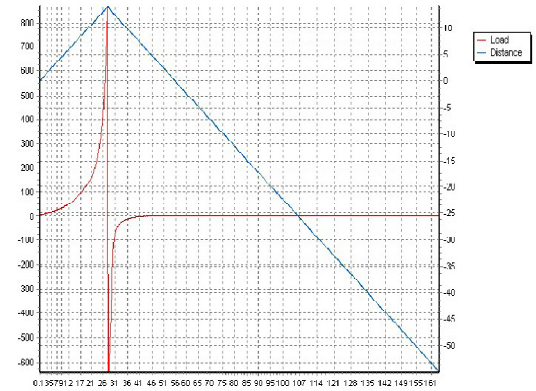

Figure 7: Brand C

relationship has proved to be valid and linear for topical formulations where the percentage of drug released is less than approximately 30% of the amount applied in the donor compartment, forming the matrix, for an infinite drug dosage. Thus, the diffusion coefficient is not concentration-dependent [1,20]. The results of in-vitro drug diffusion study were carried out upto 50 min and their results are summarized in Table 3 and figure 8. Percentage of drug released at 50 min by Brand A (78.5%) Brand B (96.8%) and Brand C (72.6%). The order of release of the drug from various gel formulationswas as follows: Brand B > Brand A > Brand C. It was concluded that gel of Brand B released DDA high rate which was found to be better compared to other marketed products[12].

According to results obtained for diffusion study of all marketed formulations, the drug released follows Korsmeyer and Peppas model [7].

Table 3 – Percentage drug release from different marketed formulations

Sr.no.	Time (Min)	Reference product	% drug release Brand A	% drug release Brand B	% drug release Brand C
1	0	0	0	0	0
2	10	8.76	20.9	22.6	22.3
3	20	14.14	30.8	38.9	35.6
4	30	23.58	48	45.5	48.2
5	40	30.93	56.8	58.9	52.5
6	50	38.38	78.5	96.8	72.6

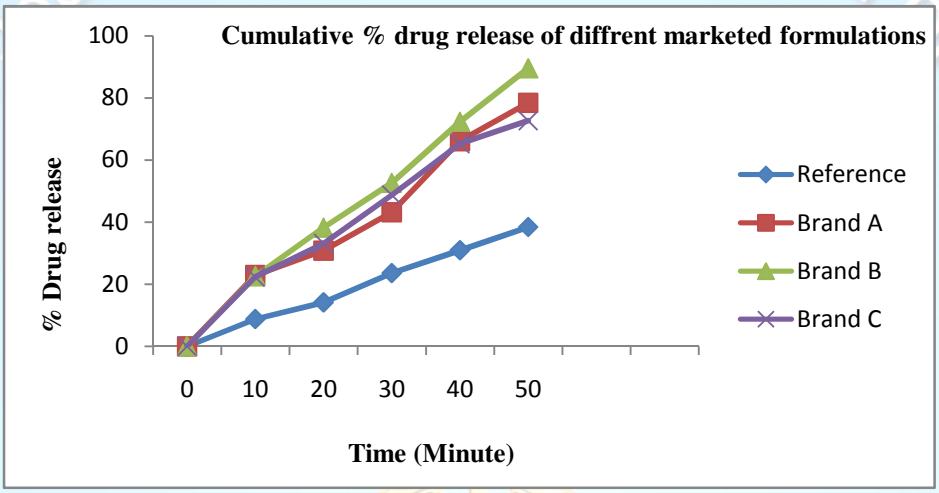

Figure 8: In-vitro drug release of DDA from different marketed formulations and reference product

Table 4 represents the results for drug release rate, correlation coefficient and lag time for all three products along with reference formulation. The lag time is defined as the time required for a system involving the passage of a substance through a membrane to reach equilibrium [15]. The lag time is calculated by extrapolating the linear regression line along with the time axis. An intercept of "x" which generally corresponds to a small fraction of an hour is a normal characteristic in release assays plots (FDA. 1997). The values in Table 4 showed that the products had a short lag time, that is, the diffusion equilibrium of the process was established within a short period of time. The release results can reflect the combined effect of several physical and chemical parameters, including the solubility, particle size and rheological properties of the dosage form. This explains the differences between the formulations evidenced in every test performed individually and this is considered as a test to be used for the "final quality control" (FDA, 1997)[16].

Table 4: Release rate, correlation coefficient and lag time of product

Brand	Release rate ($ug/cm^2/\sqrt{h}$)	Correlation coef-ficient		Lag time
Reference	637.18 ± 51.52	+0.6548	-0.0108	5.56
Brand A	700.27 ± 32.52	+0.5156	-0.0936	6.02
Brand B	475.35 ± 15.82	+0.5771	-0.1051	4.75
Brand C	354.25 ± 10.25	+0.5243	-0.0683	2.25

Moreover, the different time release of drug compared to reference product showed statistically significant in drug release profile. The results of confidence interval (CI at 95% level) were also mentioned in Table 5. Not any of the brand products is similar to the reference product because not any of the brand products is lies within the stated limit that is 75% and 133.33% (FDA, 1997). The C.I. is a range of value, above and below a finding in which the actual value is likely to fall. The C.I. represents the accuracy or precision of an estimate. 95% of CI would contain the population mean and a 0.95 probability[12]. Based on results obtained in

Table 5, we can come to final conclusion that all marketed formulations are not similar with reference formulation because none of them showed their values within the stated limit.

Conclusion

The purpose of the study was to carry out the comparative quality control evaluation of different marketed DDA gel formulations. Texture Profile Analysis (TPA) was carried out to learn about texture and physical properties of formulations. Texture analysis showed Brand B was found to be good. The percentage of drug released by Brand B was found to be 96.8 which is better than Brand A and C. The order of release of the drug from various gel formulations was as follows: Product B > Product A > Product C. Not any of the Brand products were considered to be similar to the reference drug product. The difference between the fluxes of the products may be attributed to different vehicle components. Thus, different formulations may result in distinct amounts of drug diffusion into the skin and may, thus, exhibit different intensities of activity. This results obtained highlight the influence of excipients on the performance of topical products. However, in vitro release tests only provide an indication of the formulation performance.

Table 5 : statistical testing of difference between reference and brand

Comparison	Limit value of the 90% confidence interval	Conclusion
Ref × Brand A	51.56 – 9.36	Not similar
Ref × Brand B	57.71 – 10.51	Not similar
Ref × Brand C	52.43 – 6.83	Not similar

***Requirement for similarity: values must be between 75% and 133.33% (FDA, 1997)**

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Reference:

1. Shah V.P. In-vitro release from semisolid form. In: Bronugh.L. ; maibach, H.I. (Eds.) percutaneous absorption. Boca Raton : Taylor and Francis group, 2005; p. 473-480
2. Ueda C.T. et al. A Topical and transdermal drug products, *Pharm. Forum* 2009;35(3):750-764
3. BRASIL. Resolution RDC n. 48, dated October 6, 2009. It provides for the realization of alteration, inclusion, suspension, reactivation, and post-registration, cancellation of medications and other measures. National agency for health surveillance, Brasilia, October 07 2009. Section 1, p.60-67
4. Shivhare U.D. et al. formulation development and evaluation of diclofenac sodium gel using water soluble polyacrylamide polymer, digest journal of nanomaterials and biostructure 2009;4:285-290
5. Ravindra R.P. et al. comparison of physical characteristics of vanishing cream base, cow ghee and shata dhauta ghrita as per pharmacopoeial standard, Int J pharm bio sci 2013 oct;4(4):14-21
6. Kaur L.P. et al. Development and evaluation of topical gel of minomidil from different polymer bases in application of alopecia. Int. J. pharmacy and pharm. Sci. 2010;2:43-47
7. Ashni verma et al. formulation, optimization and evaluation of clobetasol propionate gel, Int journal of pharmacy and pharmaceutics sci 2013; 5(4): 666-674
8. Bharat parashar et al. formulation and evaluation of gel containing miconazole nitrate an antifungal agent, Int journal of pharma research and review 2013;2(6):675-680
9. V.V. pande et al. fabrication and characterization of sertaconazole nitrate microsponge as a topical drug delivery system, Indian journal of pharmaceutical sci 2015;77(6):675-680
10. Damodar c goupale et al. evaluation of physical stability of oleogels containing diclofenac diethylamine. Research journal of pharmaceutical, biological and chemical sciences 2011;2(4):92-99
11. Baksh abrar et al. formulation and in-vitro evaluation of NSAIDs gel. International journal of current pharmaceutical research 2012;4(3):56-58

12. Karin goebel et al. In-vitro release of diclofenac diethylamine from gels: evaluation of generic semisolid drug product in brazil. Brazillian journal of pharmaceutical science 2013;49(2)
13. Ganesh dinkarrao basarkar et . development of microsperes containing diclofenac diethylamine as sustained release topical formulation. Bulletin of pharmaceutical research 2013;3(1):14-22
14. Shah, V.P. et al. In vitro release: Comparative evaluation of vertical diffusion cell system and automated procedure. Pharm. Dev. Technol. 2003;8(1):97-102
15. Aulton, M.E. Design of pharmaceutical forms. 2nd ed. Porto Alegre: Artmed, 2005, 677
16. Bemvindo, C.S. comparative study of cutaneous clearance and penetration of miconazole nitrate from commercial topical emulsions. Rio de Janeiro, 2006. 110 p. [Master's Degree Thesis, Federal University of Rio de Janeiro, Brazil]
17. Vishal pande et al. Design expert assisted formulation of topical bioadhesive gel of sertaconazole nitrate. Advance Pharmaceutical Bulletin 2014;4(2):121-130
18. Guy R.H et al. On the determination of drug release rates from topical dosage forms. Int. J. Pharm. 1990;60(2):R_1-R_3
19. Higuchi W.I. Analysis of data on the medicament release from ointments. J. Pharm. Sci. 1962;51(8):802-804.
20. Toscano C.: Development and validation of a system for evaluating the in-vitro release of benzoyl peroxide delivered in geysers. Rev. Bras. Cienc. Farm, 2001, 37(3) 341-346