International Journal of Research in Advent Technology, Vol.6, No.8, August 2018
E-ISSN: 2321-9637
Available online at www.ijrat.org

Development and Evaluation of Amlodipine Besylate Laden *In-Situ* Film

Sayantan Mukhopadhyay [1], Nidhi Dobhal [2], Preeti Kothiyal [3]
Department of Pharmaceutics[1,2,3] , Shri Guru Ram Rai Institute of Technology and Science, Shri Guru Ram Rai University, Dehradun, Uttarakhand, India[1, 2,3]
Email: sayantan.pharmaceutics@gmail.com[1], nidhidobhal94@gmail.com[2]

Abstract - Film forming formulation are well-defined as non-solid dosage form that produce a substantial film *in-situ* after application on the skin or any other body surface. In the current research, the *in-situ* film of Amlodipine besylate were prepared. The polymeric solution of carbopol and HPMC with ethanol, applied to the skin as a semisolid and form an almost invisible film *in-situ* by solvent evaporation method. The prepared drug loaded gels and *in-situ* films were subjected for various evaluation parameters like, spreadability, pH, viscosity and thickness of film, weight uniformity, percent moisture content, moisture uptake, drug content, water vapour transmission, rolling ball, bio adhesion and *in-vitro* diffusion study respectively. The evaluation shows viscosity of gel 1416.75 to 1701 cps, pH of gel 6.63±0.124 to 7.2±0.081, spreadability 7.83±0.12 to 11.09±0.75cm^2, thickness of film were less than 0.2 mm, weight uniformity 0.027±0.001 to 0.054±0.001gm, percent moisture content 9.06±0.53 to 21.12±0.78%, moisture uptake 12.32±0.23 to 23.08±0.72%, drug content 76±1.63 to 88.6±0.4%, water vapour transmission 9.15±0.02 to 19.2±0.13 %, rolling ball 2.36±0.004 to 3.56±0.020 inch. The *in vitro* diffusion study of amlodipine laden *in-situ* film were carried out and the percent cumulative drug release from the *in-situ* film was in the range of 82.54 – 91.52% during the study period. From experimental study it was concluded that amlodipine laden *in-situ* film was successfully delivered drug via transdermal route, hence reduce first pass metabolism and improved patient compliances.

Keywords: *In-situ* film; Amlodipine besylate; Solvent evaporation.

1. INTRODUCTION:

The topical delivery system refers to a method wherein the formulation is applied to the superficial areas such as the skin, eyes, nose and vagina to treat ailments. [1] The topical route provide enormous and diverse surface for the application of drug. The topical delivery also offer an unconventional route other than the oral route and for the parenteral route. [2]
Amlodipine besylate is the antihypertensive and anti anginal drug, a third-generation dihydropiridin which act as a calcium channel blocker. Amlodipine besylate bind the nondihydropiridin binding site and dihydropiridin binding site and block the voltage dependent calcium channel. [3]

Amlodipine is conventionally deliver through oral route in a tablet dosage form but it has slow absorption and having first pass metabolism. So its required alternative route of drug delivery. Several scientific work was reported where amlodipine was delivered via topical route. One of the promising route is transdermal delivery and they are define as the dosage forms designed to deliver a therapeutically effective amount of drug across a patient's skin to systemic circulation at a predetermined rate over a prolonged period of time[4]. But the transdermal delivery have numerous drawback such as irritation in skin, cause a trouble when applied in the curved area of the body, extreme discomfort when peel off from the body and these formulations are difficult in preparation. These drawbacks can be overwhelmed by the other formulation such as gel or ointments [5]. The gel are usually comfortable to wear but may leave a tacky or greasy feel after application. So the development of TDDS formulations recently has been focused on employing several polymer gels as a film-forming agent [6].

Incorporation of the drug in a film forming gel would facilitate prolonged contact of the drug on the skin. Film forming formulation are defined as non-solid dosage form that produce a substantial film in-situ after application on the skin or any other body surface. The polymeric solution is applied to the skin as a liquid/semisolid and form an almost invisible film *in-situ* by solvent evaporation [2][5]. In this study the amlodipine besylate *in-situ* film was prepared by using combination of carbopol 934 as a gelling agent and HPMC E50LV as a film forming polymer in which the PEG 400 and glycerine was used as a plasticizers. The effect of different permeation enhancer were studied in this research.

2. MATERIALS AND METHOD

2.1 *Preparation of in-situ film*

The *in-situ* film is formed by the solvent evaporation technique. In this method, amlodipine besylate laden film was prepared using different polymer. The Carbopol 934 (1.5%w/v) and HPMC E50 LV (6%w/v) was dissolved in ethanol. The ethanolic solution of HPMC was slowly added to the carbopol ethanolic solution. The drug was added to polymeric solutions of the carbopol and HPMC with continuous stirring for 20 minutes followed by addition of PEG 400, glycerol and various skin permeation enhancer. The composition in this study was shown in Table 1.

Table 1- Composition table of *in-situ* film formulation

S.No	Ingredients	Formulation code			
		FF1	FF2	FF3	FF4
1.	Amlodipine besylate(%)	0.2	0.2	0.2	0.2
2.	HPMC(E50LV(%w/v)	6	6	6	6
3.	Carbopol 934(%w/v)	1.5	1.5	1.5	1.5
4.	PEG 400(%v/v)	10	10	10	10
5.	Glycerine(%v/v)	0.4	0.4	0.4	0.4
6.	Ethanol(%v/v)	40	40	40	40
7.	Water(%v/v)	60	60	60	60
8.	Propylene glycol PG(%v/v)	5	-	-	-
9.	Tween 80(%v/v)	-	5	-	-
10.	Isopropyl mystriate(%v/v)	-	-	3	-
11.	Oleic acid(%v/v)	-	-	-	7

3. EVALUATION PARAMETERS:

3.1. Evaluation *parameter of gel:*

3.1.1. *Spreadability studies*: Spreadability of the amlodipine besylate gel was determined by spreading the 500mg of gel on a glass plate and then another glass plate was sited on it. 500g of weigh was put on the upper glass plate. After 5min the increase in diameter was noted and spreadability of gel was calculated by given formula [7].

$A = \pi r^2$

3.1.2. *Viscosity estimation:* The viscosity estimation of amlodipine besylate gel was executed on brookfield viscometer. The spindles was rotated at different rpm. [8]

3.1.3. *pH determination:* By digital pH meter, the pH of different gel formulations was determined.[9]

3.2. *Evaluation parameter of film:*

3.2.1. *Thickness of film:* The thickness of prepared *in-situ* film was determined by using digital micrometer screw gauge. The average thickness was determined and reported. [10]

3.2.2. *Weight uniformity*: Weight uniformity of film was determined by taking a weight of specific area of a film from different parts of the film by using digital balance. The average weights was then calculated from individual weights. [11]

3.3.3. *Percent moisture content*: The prepared *in-situ* film was cut into the area of $1cm^2$ and weighed individually. The weighed films were kept in a desiccator which have fused calcium chloride at room temperature for 24hrs. The films were reweighed after 24hrs. The percent moisture content was calculated from the given formula. [12]

Percent moisture content =

$$\frac{\text{Initial weight} - \text{Final weight}}{\text{Final weight}} \times 100$$

3.3.4. *Percent moisture uptake*: The weighed films were kept in a desiccator which contain saturated solution of potassium chloride at room temperature

for 24hrs to maintain 84% RH. The films were reweighed after 24hrs and percent moisture uptake was calculated from the given formula [13].

Percent moisture uptake =

$$\frac{\text{Final weight} - \text{Initial weight}}{\text{Initial weight}} \times 100$$

3.3.5. *Drug content*: 1cm^2 area of film was dissolved in a methanol in specific volume. The solution was shaken and sonicated for 15minutes. The solution was filtered and the drug content was analyse spectrophotometrically at 360nm wavelength. [14]

3.3.6. *Water vapour transmission*: The glass bottle was filled with the calcium chloride and with the help of the adhesive the film was placed over the edge. Then the bottle was weighed and for 24hrs placed in the humidity chamber. After 24hrs the glass bottle was removed and the film was reweight. [12]

$$\text{Water vapour transmission} = \frac{W}{S \times T}$$

Where, W = increased weight of film after 24hr.
S = area of film exposed
T= exposure time.

3.3.7. *Rolling ball tack test*: It is an adhesive test of film in which the stainless steel ball of 7/16 inches is discharged on an inclined track, the ball moves down and comes into contact with flat, upward facing adhesive. The total distance travels by ball in the adhesive suggest the measurement of tack, which is expressed in inch. [14]

3.3.8. *Bio adhesion force*: By using a modified balance method the bio adhesion was measured. The two pans of balance were detached and replaced with the 100ml beaker and a glass side on right and left side respectively. On left side of the balance the 20g of weight was suspended. Another slide was placed below the hanged slide. The hairless goat skin were attached with both slides. One gram of gel was applied between the two goat skins. A force was applied to form bio adhesion bond, and then slowly water was added on right side beaker, till the gel was separated from one face of goat skin attached. Volume of water added was converted to mass. This gave the bio adhesive strength of gel in grams. [15]

3.3.9. *In-vitro diffusion study*: The *in-vitro* release of amlodipine besylate from the prepared *in-situ* film was performed by using Franz diffusion cell. The formulation was applied on the dialysis membrane which was placed between the donor and receptor compartment. The donor compartment was filled with the phosphate buffer pH 7.4 as a diffusion media. The assembly was kept on magnetic stirrer and solution was stir continuously. The sample was withdrawn at the predetermine time interval and the sink condition was maintain throughout the process. The sample was analysed by the UV spectrophotometry at 360nm and then cumulative drug release was determined. [16]

4. RESULT AND DISCUSSION

4.1. *Spreadability studies*:

The spreadability of gel range from 7.83±0.12 to 11.09±0.75cm^2 (Table 2) which indicate that prepared gel is easily spreadable by small amount of shear.

Table 2. Spreadability studies of gel of formulation FF1-FF4

S.no	Formulation code	Spreadability(cm^2) (Mean±S.D)
1.	FF1	7.83±0.123
2.	FF2	8.56±0.312
3.	FF3	8.33±0.210
4.	FF4	11.09±0.75

4.2. *pH determination*:

pH of all formulation were found in the range of 6.63±0.124 to 7.2±0.081 (as shown in table 3) which match with the physiological pH of skin (pH 6.8) that indicated it does not cause any irritation on application.

Table 3. pH determination of gel of formulation FF1-FF4

S.no	Formulation code	pH value (Mean±S.D)
1.	FF1	6.73±0.047
2.	FF2	6.86±0.047
3.	FF3	6.63±0.124
4.	FF4	7.2±0.081

4.3. *Viscosity estimation*:

From the result 1416.75 to 170 cps (Table 4 and fig.1) it was concluded that all the formulation follow non-newtonian pseudoplastic flow, a shear thinning behaviour, as with increase in the shear rate the apparent viscosity of gel was reduced.

Table 4. Measurement of average viscosity of gel of FF1-FF4

S.no	Formulation code	Average of viscosity(cp)
1.	FF1	1701
2.	FF2	1416.75
3.	FF3	1654.75
4.	FF4	1495.75

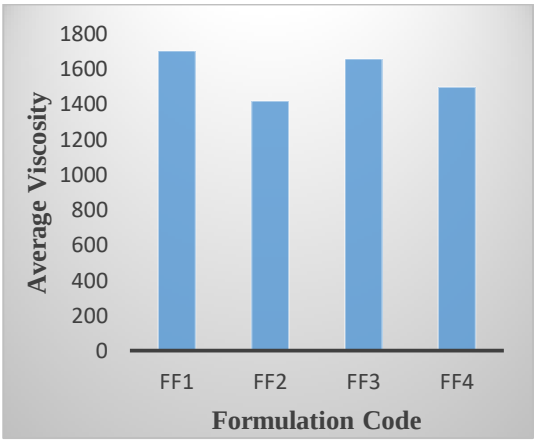

Fig 1. Average viscosity of gel and formulation FF5-FF9

4.4. *Thickness of film*:

The thickness of all the formulations of *in-situ* film was less than 0.2mm.

4.5. *Weight uniformity*:

The result indicated that as the polymer concentration remain fixed there is no significant difference between the weights of the prepared films, it was ranged between the 0.027±0.001 to 0.054±0.001 (Table no.5).

Table 5. Weight Uniformity of *in-situ* film of FF1-FF4

S.no	Formulation code	Weight uniformity(gm) (Mean±S.D)
1.	FF1	0.054±0.001
2.	FF2	0.045±0.001
3.	FF3	0.034±0.002
4.	FF4	0.027±0.001

4.6. *Percent moisture content*:

The percent moisture content was found to be 9.06 ± 0.53 to 21.12 ± 0.78 % (Table no.6 and fig 2). The film should be stable, dry and fragile if the moisture content is found to be lower.

Table 6. Percent moisture content of *in-situ* film of FF1-FF4

S.no	Formulation code	Percent moisture content (Mean±S.D)
1.	FF1	21.12±0.78
2.	FF2	16.22±0.73
3.	FF3	12.7±1.08
4.	FF4	9.06±0.53

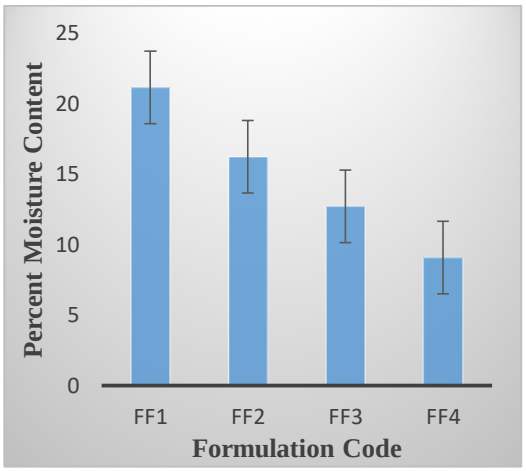

Fig 2. Percentage moisture content of prepared formulations of *in-situ* film

4.7. *Percent moisture uptake*:

The moisture uptake in the film ranged from 12.32 ± 0.23 to 23.08±0.72% (shown in table 7 and fig 3).The decrease in the amount of moisture uptake decrease the risk of microbial contagion in the film and its bulkiness.

Table 7. Percent moisture uptake of *in-situ* film of FF1-FF4

S.no	Formulation code	Percent moisture uptake (Mean±S.D)
1.	FF1	23.08±0.72
2.	FF2	21.33±0.20
3.	FF3	15.34±0.71
4.	FF4	12.32±0.23

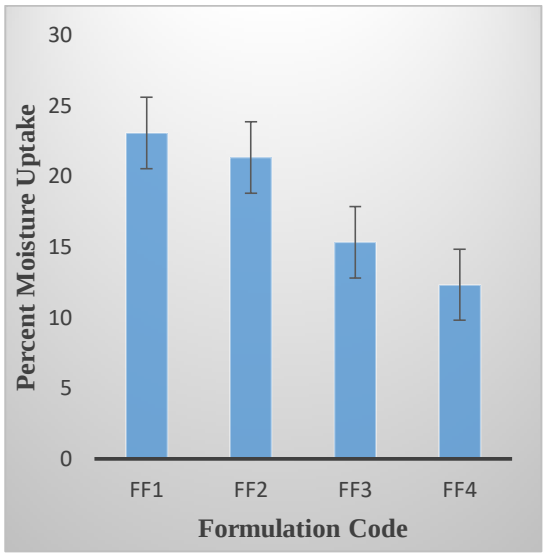

Fig 3. Percentage moisture uptake of prepared formulations of *in-situ* film

4.8. *Drug content*:

The drug content of the film was ranged from 76±1.63 to 88.6±0.47% (Table 8 and fig. 4). The result of drug content indicate that the drug is uniformly dispersed in formulation.

Table 8. Drug content of *in-situ* film of FF1-FF4

S.no	Formulation code	Drug content (Mean±S.D)
1.	FF1	88.6±0.47
2.	FF2	76±1.63
3.	FF3	85.5±0.44
4.	FF4	83.3±0.25

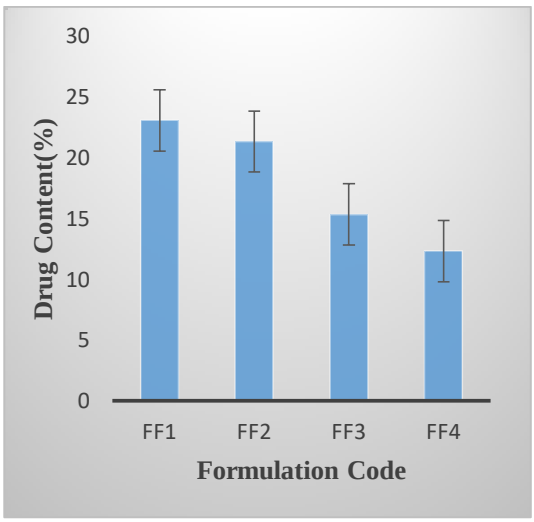

Fig 4. Drug content of prepared formulations of *in-situ* film

4.9. *Water vapour transmission rate*:

Water vapour transmission rate was found to be 9.15 ± 0.02 to 19.2 ± 0.13% (Table no. 9), the results indicate that all the films were permeable to water vapour and the degradation of the polymer matrix hence the release of the drug through the film takes place.

Table 9. Water vapour transmission of *in-situ* film of FF1-FF4

S.no	Formulation code	Water vapour transmission (Mean±S.D)
1.	FF1	19.2±0.13
2.	FF2	17.55±0.22
3.	FF3	9.15±0.02
4.	FF4	11.12±0.05

4.10. *Rolling ball tack test*:

Rolling ball tack was found to be 2.36±0.004 to 3.56±0.020inch (Table no 10). From the result it was observed that the tack of the film is suitable and good to adhere over a skin surface.

Table 10. Rolling ball tack test of *in-situ* film of FF1-FF4

S.no	Formulation code	Rolling ball tack test(in inches) (Mean±S.D)
1.	FF1	2.36±0.004
2.	FF2	3.12±0.016
3.	FF3	3.54±0.032
4.	FF4	3.56±0.020

4.11. *Bio adhesion force*:

The bio adhesion force ranged from 10.62 ± 0.36 to 16.84±0.13g (shown in table no 11) bio adhesion strength was enough to retain the film in the skin for a sufficient period of time.

Table 11. Bio adhesion test of *in-situ* film of FF1-FF4

S.no	Formulation code	Bio adhesion force (g) (Mean±S.D)
1.	FF1	12.80±0.09
2.	FF2	15.06±0.02
3.	FF3	16.84±0.13
4.	FF4	10.62±0.36

4.12. *In vitro release study*:

The *in vitro* diffusion study of amlodipine besylate laden *in-situ* film were carried out and the percent cumulative drug release from the *in-situ* film was observed minimum in formulation FF2 i.e. (82.54%) and maximum in formulation FF1 i.e. (91.52%) during the study period. The order of the drug release was found to be FF1> FF4> FF3> FF2 where the amount of drug release after 24 hrs were 91.52%, 87.54%, 85.07%, and 82.54% respectively (reported in table 12 and fig 5).

Table 12. *In vitro* release study of formulations FF1- FF4

S.No	Time (hrs)	Cumulative Drug Release (%)			
		FF1	FF2	FF3	FF4
1.	0.5	3.621	2.147	5.058	-
2.	1	6.501	4.821	7.184	2.482
3.	1.5	7.824	5.943	8.871	5.762
4.	2	9.212	7.119	10.53	8.658
5.	3	11.54	12.82	11.90	9.690
6.	4	12.88	14.06	13.85	10.92
7.	5	15.23	17.62	16.60	12.93
8.	6	18.79	20.82	19.78	13.58
9.	7	22.65	23.94	21.58	18.66
10.	8	27.82	26.83	25.60	24.69
11.	9	32.54	28.47	30.18	27.93
12.	10	35.89	31.09	32.12	30.98
13.	24	91.59	82.54	85.07	87.54

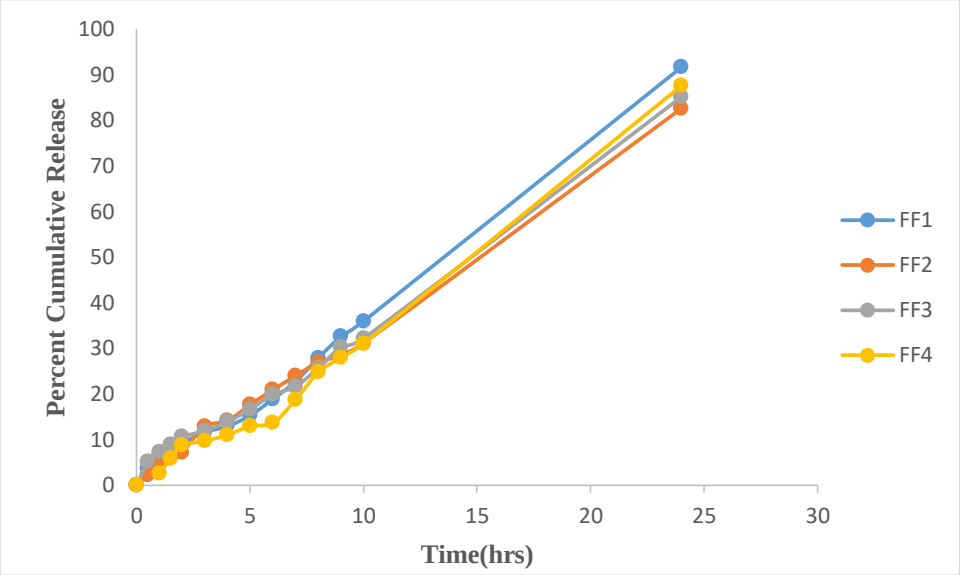

Fig 5. *In-vitro* drug release profile of formulations FF1 to FF4

The above result shows that the formulation containing propylene glycol (FF1) as a permeation enhancer show maximum drug release. On the basis of evaluation parameter such as spreadability, viscosity, drug content, percent moisture content and *in-vitro* drug diffusion the FF1 is the best optimized formulation.

5. CONCLUSION:

From the experimental study it was concluded that amlodipine besylate was successfully delivered in a form of *in-situ* film via topical route. The drug was released at a controlled rate by pass the first pass metabolism, hence increase its bioavailability and may provide better therapeutic compliance.

REFERENCES

[1] 1] Singh Malik, D., Mital, N. and Kaur, G., 2016. Topical drug delivery systems: a patent review. Expert opinion on therapeutic patents, 26(2), pp.213-228.

[2] 2] Kathe, K. and Kathpalia, H., 2017. Film forming systems for topical and transdermal drug delivery. Asian journal of pharmaceutical sciences.

[3] 3] Kaynak, M.S., Bogacz, A., Stelmasinski, M. and Sahin, S., 2011. Bioavailability File: Amlodipine/Biyoyararlanim Dosyasi: Amlodipin. FABAD Journal of Pharmaceutical Sciences, 36(4), p.207.

[4] 4] Dobhal Nidhi, Mukhopadhyay Sayantan., 2018 Transdermal Drug Delivery – Its Approaches, Utility And Current Advancement, Indo American Journal Of Pharmaceutical Sciences.

[5] 5] Bajaj, H., Kumar, T. and Singh, V., 2016. Film forming gels: a review. Research journal of pharmaceutical biological and chemical sciences, 7(4), pp.2085-2091.

[6] 6] Guo, R., Du, X., Zhang, R., Deng, L., Dong, A. and Zhang, J., 2011. Bioadhesive film formed from a novel organic–inorganic hybrid gel for transdermal drug delivery system. European Journal of Pharmaceutics and Biopharmaceutics, 79(3), pp.574-583.

[7] 7] Tasdighi, E., Azar, Z.J. and Mortazavi, S.A., 2012. Development and in-vitro evaluation of a contraceptive vagino-adhesive propranolol hydrochloride gel. Iranian journal of pharmaceutical research: IJPR, 11(1), p.13.

[8] 8] Dantas, M.G.B., Reis, S.A.G.B., Damasceno, C.M.D., Rolim, L.A., Rolim-Neto, P.J., Carvalho, F.O., Quintans-Junior, L.J. and Almeida, J.R.G.D.S., 2016. Development and evaluation of stability of a gel formulation containing the monoterpene borneol. The Scientific World Journal, 2016.

[9] 9] Makwana, S.B., Patel, V.A. and Parmar, S.J., 2016. Development and characterization of in-situ gel for ophthalmic formulation containing ciprofloxacin hydrochloride. Results in pharma sciences, 6, pp.1-6.

[10] 10] Cherukuri, S., Batchu, U.R., Mandava, K., Cherukuri, V. and Ganapuram, K.R., 2017. Formulation and evaluation of transdermal drug delivery of topiramate. International journal of pharmaceutical investigation, 7(1), p.10.

[11] 11] Alam, M.I., Alam, N., Singh, V., Alam, M.S., Ali, M.S., Anwer, T. and Safhi, M.M., 2013. Type, preparation and evaluation of transdermal patch: A review. World J. Pharm. Pharm. Sci, 2, pp.2199-2233.

[12] 12] Darwhekar, G., Jain, D.K. and Patidar, V.K., 2011. Formulation and evaluation of transdermal drug delivery system of clopidogrel bisulfate. Asian Journal of Pharmacy and Life Science ISSN, 2231, p.4423.

[13] 13] Prajapati, S.T., Patel, C.G. and Patel, C.N., 2011. Formulation and evaluation of transdermal patch of repaglinide. ISRN pharmaceutics, 2011.

[14] 14] Dhiman, S., Singh, T.G. and Rehni, A.K., 2011. Transdermal patches: a recent approach to new drug delivery system. Int J Pharm Pharm Sci, 3(5), pp.26-34.

[15] 15] Varma, V.N.S.K., Maheshwari, P.V., Navya, M., Reddy, S.C., Shivakumar, H.G. and Gowda, D.V., 2014. Calcipotriol delivery into the skin as emulgel for effective permeation. Saudi Pharmaceutical Journal, 22(6), pp.591-599.

[16] 16] Taksande, J.B., Gupta, M., Trivedi, R.V., Wadher, K.J. and Umekar, M.J., 2016. Formulation and characterization of organic-inorganic hybrid film for transdermal drug delivery. Journal of applied pharmaceutical research, 4(3), pp.8-15.