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FORMULATION AND EVALUATION OF MATRIX TYPE TRANSDERMAL PATCHES OF BENAZEPRIL HYDROCHLORIDE

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Abstract:
In the present study an attempt has been made to formulate and evaluate the transdermal patches of benazepril hydrochloride using various types of polymers (Eudragit L100, Eudragit S100 and ethylcellulose).
The results of compatibility studies by Fourier transform infrared spectroscopy and differential scanning Calorimetry which shows no interaction between the drug and polymers.
The polymers Eudragit L100 and Eudragit S100 used for the formulation of transdermal patches exhibited milestone film forming property as compared to ethylcellulose.
The patches designed by using Eudragit L100 and Eudragit S100 were thin, flexible, smooth and transparent where as the patches prepared by using ethylcellulose were thin, flexible, smooth and opaque.
The weight variation test showed less variation in weight and suggesting uniform distribution of drug and polymer over the mercury surface.

Keywords: *Transdermal patches, Benazepril hydrochloride, polymers Eudragit L100, Eudragit S100, drug and polymer.*

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INTRODUCTION:

For several decades treatment of an acute disease or a chronic illness has been mostly accomplished by delivery of drugs to patients using various pharmaceutical dosage forms, including tablets, capsules, pills, suppositories, creams, ointments, liquids, aerosols, and injectables, as drug carriers. This type of drug delivery system is known to provide a prompt release of drug. Therefore to achieve and maintain the drug concentration within the therapeutically effective range that needed for treatment, it is often necessary to take this type of drug delivery system several times a day. This results in a significant fluctuation in drug levels. They have resulted in the development of new techniques for drug delivery. This results in a significant fluctuation in drug levels. However it was needed to development of precise and effective novel techniques for drug delivery. These techniques are capable of controlling the rate of drug delivery, sustaining the duration of therapeutic activity, and/or targeting the delivery of drug to a tissue.

The goal of developing new pharmaceutical medication systems is to improve drug delivery in the body (for example, oral controlled-release, inhalation, implant, and transdermal delivery systems) [1]. In comparison to oral dosage forms, there is a lot of room for new topical and transdermal drug delivery solutions to evolve. Therefore, pharma industries spend a huge amount of money to trigger the delivery of drug via transdermal delivery systems/techniques.

With a 30–40% growth in novel transdermal medications in recent years, the transdermal patch industry has approached £2 billion [2].The medications' adverse effects are thought to be reduced in transdermal dosing formulations [3]. When there is a significant first-pass metabolism that can metabolise medications early, the transdermal method of delivery is beneficial, furthermore, they can help with drug release over a longer period of time (up to one week), which is extremely beneficial for non-compliant individuals. [4]. However, not all medications can be formed into a transdermal patch due to physical, chemical, and biological constraints. A medication must have a molecular size of less than 500 Da to be delivered via transdermal route.

A problem with transdermal medication delivery is that only a small number of pharmaceuticals are available that can be delivered this way since drugs with molecular sizes greater than 500 Da usually do not permeate the skin's stratum corneum (SC). Because of the poor solubility of such moieties in both lipids and water, medications with a very high or low partition coefficient cannot reach blood circulation, but pharmaceuticals that are very sensitive to high temperatures can be delivered this way.

Considering the magnificent significance of transdermal preparation, in this study, an attempt has been made to formulate and evaluate the transdermal patches of benazepril hydrochloride using various types of polymers.

MATERIALS AND METHODS:

All the chemicals and solvents used in this study were of analytical grade.

Benazepril Hydrochloride and Ethyl cellulose were gift samples from Safe Tab Life Science, Puducherry, India while Eudragit L100 and Eudragit S100 were gift samples from from Orchid Pharmaceuticals, Chennai, India. Dibutyl phthalate, Poly ethylene glycol, Dimethyl sulfoxide and some other crucial chemicals and solvents were purchased from the authentic suppliers.

Preparation of matrix type transdermal patches

Matrix type transdermal patches (F_1-F_{18}) containing benazepril hydrochloride are prepared by solvent casting technique employing a mercury substrate [5]. Polymer solutions are prepared using ethanol as solvent [6]. To the polymeric solution known weight of drug (benazepril hydrochloride 69.3 mg) is added and mixed slowly with a glass rod for 20 minutes until a homogenous drug polymer solution is formed. Then plasticizer and permeation enhancer of required quantity are added and mixed thoroughly. The resulting homogenous drug-polymeric solution is poured on a mercury substrate (area of 13.86 cm^2) in a petridish and dried at room temperature [7].The rate of evaporation of solvent is controlled by inverting a funnel over the petridish. The film formation is noted by observing the mercury surface after complete evaporation of the solvent [8]. After drying at room temperature for 24 hours, membranes are taken out, packed in aluminium foil [9] and stored in desiccator until further use.

Drug polymer interaction using FT-IR spectroscopy

The IR Spectral analysis of benazepril hydrochloride alone showed that the principle peak were observed at wave number of 3504.43, 3111.282978.18, 2805.01, 1735.99, 1676.20,1643.41, 1317.43, 1200.20, 858.35, and 765.77 (cm^{-1}) which indicates the presence of Carboxylic acid O-H stretch, Secondary amine N-H

stretch, Methyl C-H stretch, Methylene C-H stretch, Carboxylic acid C=O stretch, Keto C=O stretch, Aromatic C=C ring stretch. aromatic C-H stretch respectively. The results were shown in Figures3A, 3B, 3C, 3D, 3E, 3F & 3G. In the IR spectra of the physical mixture of benazepril hydrochloride with ES100, EL100 & EC the principle peak were observed nearly at the same wave number as in benazepril hydrochloride pure drug. However some additional peaks were observed with physical mixture, which could be due to the presence of polymers. The result of FTIR study suggested that there is no interaction between the drug and polymers used in the present study[10].

Differential Scanning Calorimetry Study

The physicochemical compatibility between the drugs and polymers used in patches is studied by using DSC studies. The DSC of the pure drug and physical mixtures of drug: polymer at 1:1 ratio is carried out. The sample is heated between 50ºC and 250º at the rate of 10ºC/min in an atmosphere of nitrogen (20 ml/min). The thermograms obtained for the drug, polymers, physical mixture of drugs with polymers are compared.

Percentage of moisture content and moisture uptake

The prepared films are weighed individually and kept in a desiccator containing silica gel at room temperature for 24 hours. The films are again weighed and the percentage moisture content is calculated using the formula.

$$\text{Percentage Moisture Content} = \frac{\text{Initial weight} - \text{Final weight}}{\text{Final weight}} \times 100$$

1) *Ex-vivo* permeation studies:

The *ex vivo* skin permeation experiments are conducted using vertical type Franz diffusion cells having receptor compartment capacity of 15 ml [11].

Preparation of rat skin membrane

Permeation studies are carried out after obtaining ethical committee clearance (Ref. No. 14024/E1/4/2011). Wister strains of male albino rats weighing between 105-120 g are used for this study. Membrane for the permeability studies is full thickness skin from the abdominal region of the rats. The hair present over the skin is removed by trimming and careful shaving so that the skin is not damaged. The skin is excised from rat after anaesthetizing. The epidermis is prepared surgically by heat separation technique, which involved soaking the entire abdominal skin in water at 60ºC for 45 sec, followed by careful removal of the epidermis. The excised skin samples are then stored in refrigerator at 0 - 4ºC and are used within three days [12].

Permeation studies

The receptor compartment is filled with 15 ml of Phosphate buffered saline pH 7.4. The transdermal patches with backing membrane are firmly pressed onto the centre of the rat skin. Once adhesion to the skin surface had been confirmed, the skin is quickly mounted on the diffusion cell receptor compartment such that the patch is tightly secured over the flange aperture. The donor compartment is then placed in position and the two halves of the cell are clamped together. The whole assembly is placed over a magnetic stirrer. The dissolution medium in the receptor compartment is stirred constantly using a magnetic bead. The samples of 0.5 ml are withdrawn at regular time intervals of 1 hour and analyzed for drug content. Receptor phases are replenished with equal volume of fresh receptor medium at each time interval [13]. Each permeation experiment is repeated three times. The cumulative amounts of drug permeated and corrected for acceptor sample replacement is plotted against time.

Determination of permeation parameters

For the determination of permeation parameter, the cumulative amount of drug permeated across the skin is plotted against time. The steady state flux (J) is calculated from the slope of the linear region of the above plot. The lag time (T) is calculated by extrapolating the linear region of the curve to the X – axis [14]. The permeability coefficient (K_p) is calculated from the ratio of flux to drug concentration in the donor chamber.

Statistical analysis

Graph Pad In Stat Version 3.0 Software is used for statistical analysis. The cumulative amount permeated and flux values obtained are tested for the determination of significant differences using a one-way analysis of variance (ANOVA).

2) *In vitro* drug release studies:

The *in-vitro* drug release study for the transdermal patches are carried out using modified paddle over disc assembly (USP Apparatus 5) [15]. The disc apparatus (European Pharmacopoeia 5.0) consists of mesh screen made of stainless steel clamped in the watch glass using nylon clips. The transdermal patch of specified area is pasted over a small piece of aluminium foil (backing layer) to prevent two dimensional releases. The transdermal patch with

backing layer is placed between inert stainless steel mesh and watch glass exposing the patch to the medium. It is also ensured that the patch does not float inside the disc assembly. The disc assembly containing transdermal patch is placed at the bottom of the dissolution vessel, with the mesh facing upwards, under the rotating paddle. The dissolution medium used is 900 ml of Phosphate buffered saline pH 7.4. The apparatus is equilibrated to the temperature of 32 ± 0.5^0c operated at 50 ± 1 rpm. The dissolution study is carried out for 12 hours. 5 ml of samples are withdrawn at regular intervals of 15 minutes for 1 hour and then 30 minutes for next 11 hour. The same volume of corresponding dissolution medium is replenished to maintain sink condition. The amount of benazepril hydrochloride released is determined by measuring the absorbance of the samples at 241.5 nm using UV-Visible spectrophotometer. Each test is performed in triplicate.

3) Study of drug release kinetics

In order to investigate the drug release mechanism from patches, the percentage cumulative drug release data is analyzed with following mathematical model [15].

Table-1 Order of reaction

Model	Equation
Zero order Kinetics	$Q = Q_0 - K_0t$
First order kinetics	$Q = Q_0 (1 - e^{-K/t})$
Higuchi square root model	$Q_t = K_H t^{1/2}$
Korsmeyer-peppas model	$Q_t / Q_\infty = K_k t^n$
Hixson-Crowell cube root model	$\sqrt[3]{Q0} - \sqrt[3]{Qt} = KHC^t$

Where,

Q_t = Amount of drug released at time t
Q_0 = Initial amount of drug
K_0, K_1, K_H , K_{HC} and K_K are the coefficients of equation. The most appropriate model is selected on the basis of goodness of fit test. The zero order kinetic describes the systems in which the drug release rate is independent of its concentration. The first order kinetics describes the system in which drug release rate is concentration dependent. Higuchi model describes the release of water – soluble drug from an insoluble matrix as a diffusion process based on the Fick's law and is square root time dependent. The Hixson – Crowell cube root law describes the drug release from a system depends upon the change in surface area or diameter of particle or system and involves no diffusion mechanism. Korsmeyer – Peppas model describes the fraction of drug release relates exponentially with respect to time. This model is generally used to analyze the release of pharmaceutical polymeric dosage forms, when the release mechanism is not well known or when more than one type of release phenomena could be involve.

RESULT AND DISCUSSION:

The mercury substrate method has been utilized for the preparation of transdermal patches that yielded opaque, smooth, flexible, non-sticky and uniform patches in case of ethylcellulose polymer where as transparent, smooth, flexible, non-sticky and uniform patches in case of Eudragit polymer. The results were shown in Table-2. The results indicated that the method used for casting the film on a mercury substrate was found to be satisfactory.

The weight of the patches were ranged from 13.60 ± 0.4898 mg to 21.80 ± 0.2160 mg for ethyl cellulose patches prepared with DMSO (F_1, F_3, and F_5), 12.20 ± 0.4326 mg to 20.06 ± 0.7930 mg for ethyl cellulose patches prepared without DMSO (F_2, F_4, and F_6), 24.76 ± 0.4189 mg to 30.80 ± 0.5099 mg for Eudragit S100 patches prepared with DMSO (F_7, F_9, and F_{11}), 23.46 ± 0.6182 mg to 28.60 ± 1.4445 mg for Eudragit S100 patches prepared without DMSO (F_8, F_{10}, and F_{12}), 11.70 ± 0.7118 mg to 26.03 ± 0.8178 mg for Eudragit L100 patches prepared with DMSO (F_{13}, F_{15}, and F_{17}), 11.30 ± 0.0471 mg to 24.90 ± 0.3741 mg for Eudragit L100 patches prepared without DMSO (F_{14}, F_{16}, and F_{18}). The results were shown in Table-3 and Figure-1. From the results it was observed that the weight of the patches containing dimethyl sulfoxide (F_1, F_3, F_5, F_7, F_9, F_{11}, F_{13}, F_{15}, and F_{17}) was greater than that of patches prepared without permeation enhancer (F_2, F_4, F_6, F_8, F_{10}, F_{12}, F_{14}, F_{16}, and F_{18}). The thickness of patches varied from 0.07 ± 0.005 mm to 0.31 ± 0.017 mm. The results were shown in Table-3 and in Figure-2. Folding endurance measures the ability of patch to withstand rupture. The results of folding endurance were shown in Table-3. The folding endurance of Eudragit patches (F_7 to F_{18}) was ranged from 32.00 ± 0.6432 to 247.0 ± 1.6329 where as it was ranged from04.33 ± 0.4714 to 35.30 ± 1.2472 for the patches prepared with Ethylcellulose (F_1 to F_6) From the results it was observed that the folding endurance was found to be high in patches containing Eudragit polymer when compared to the patches containing ethyl cellulose polymer. This was due to

less film forming property of cellulose derivative when compared to Eudragit. All the formulations showed 100% flatness.

The percentage moisture content in the patches was found to be low and ranged fro1.52 ± 0.061 to 3.66 ± 0.43%. The results were shown in Table-3 and Figure-3.

The drug content of all the patches (F_1 to F_{18}) was in the range of 75.78 ± 0.63 to 95.95 ± 1.09%. The results were shown in Table-3. The results suggest that the process employed to prepare the patches shown uniform drug content, with minimum batch variability [16].

Known concentration (10µg/ml) solution of benazepril hydrochloride in phosphate buffered saline pH 7.4 was scanned to find out the λ_{max} and it was found to be 241.5 nm. The absorbance's were measured at λ_{max} of 241.5nm. The correlation coefficient was found to be 0.9996. The results were shown in Table-4 and Figure-4.

However The IR Spectral analysis of benazepril hydrochloride alone showed that the principle peak were observed at wave number of 3504.43, 3111.28, 2978.18, 2805.01, 1735.99, 1676.20,1643.41, 1317.43, 1200.20, 858.35, and 765.77 (cm^{-1}) which indicates the expected functional groups of drug.

In Differential Scanning Calorimetry thermogram the endothermic melting transition of benazepril hydrochloride was observed at 162.27 ºC.

***In vitro* drug release studies**

The *in-vitro* drug release study for the transdermal patches are carried out using modified paddle over disc assembly (USP Apparatus 5). The objective was to estimate, characterize and rationalize the drug release from matrix film [17].

Table-2 FORMULATIONS AND PHYSICAL APPEARANCE

S.NO	FORMULATION CODE	PHYSICAL APPEARANCE
1	F_1-EC 2.5% (with DMSO)	opaque, smooth, nonsticky and flexible
2	F_2- EC 2.5% (without DMSO)	opaque, smooth, nonsticky and flexible
3	F_3- EC 3.0% (with DMSO)	opaque, smooth, nonsticky and flexible
4	F_4- EC 3.0% (without DMSO)	opaque, smooth, nonsticky and flexible
5	F_5- EC 3.5% (with DMSO)	opaque, smooth, nonsticky and flexible
6	F_6- EC 3.5% (without DMSO)	opaque, smooth, nonsticky and flexible
7	F_7- ES100 4.0% (with DMSO)	Transparent, smooth, nonsticky and flexible
8	F_8- ES100 4.0% (without DMSO)	Transparent, smooth, nonsticky and flexible
9	F_9- ES100 4.5% (with DMSO)	Transparent, smooth, nonsticky and flexible
10	F_{10}- ES100 4.5%(without DMSO)	Transparent, smooth, nonsticky and flexible
11	F_{11}- ES100 5.0% (with DMSO)	Transparent, smooth, nonsticky and flexible
12	F_{12}- ES100 5.0%(without DMSO)	Transparent, smooth, nonsticky and flexible
13	F_{13}- EL100 4.5% (with DMSO)	Transparent, smooth, nonsticky and flexible
14	F_{14}- EL100 4.5%(without DMSO)	Transparent, smooth, nonsticky and flexible
15	F_{15}- EL100 5.0% (with DMSO)	Transparent, smooth, nonsticky and flexible
16	F_{16}- EL100 5.0%(without DMSO)	Transparent, smooth, nonsticky and flexible
17	F_{17}- EL100 5.5% (with DMSO)	Transparent, smooth, nonsticky and flexible
18	F_{18}- EL100 5.5%(without DMSO)	Transparent, smooth, nonsticky and flexible

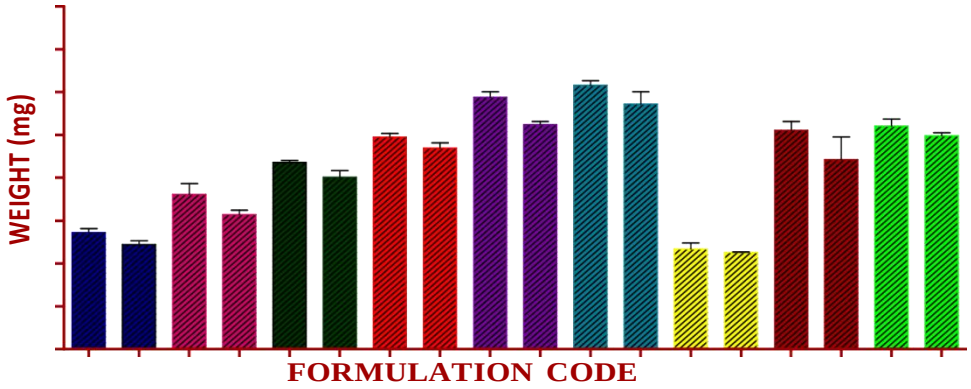
Table-3 CHARACTERISATION OF BENAZEPRIL HYDROCHLORIDE: TRANSDERMAL PATCHES

Formulation Code	Weight ± S.D* (mg)	Thickness ± S.D*(mm)	Folding Endurance ± S.D* (No. of times)	Moisture content ± S.D* (%)	Drug content ± S.D* (%)
F_1	13.60 ± 0.4898	0.08 ± 0.005	09.33 ± 0.4714	2.95 ± 0.086	84.00 ± 2.52
F_2	12.20 ± 0.4326	0.07 ± 0.005	04.33 ± 0.4714	2.50 ± 0.061	85.83 ± 1.01
F_3	18.06 ± 1.2656	0.09 ± 0.005	06.66 ± 0.4714	1.86 ± 0.123	89.38 ± 2.29
F_4	15.70 ± 0.5099	0.08 ± 0.005	05.66 ± 0.4714	2.15 ± 0.171	84.67 ± 2.89
F_5	21.80 ± 0.2160	0.10 ± 0.011	35.30 ± 1.2472	1.69 ± 0.173	75.78 ± 0.63
F_6	20.06 ± 0.7930	0.09 ± 0.010	07.33 ± 0.4714	1.52 ± 0.061	87.36 ± 1.24
F_7	24.76 ± 0.4189	0.26 ± 0.005	46.00 ± 0.8164	3.66 ± 0.430	92.08 ± 0.86
F_8	23.46 ± 0.6182	0.20 ± 0.010	32.00 ± 0.6432	3.56 ± 0.318	81.98 ± 1.86
F_9	29.40 ± 0.6539	0.27 ± 0.015	189.3 ± 1.2472	3.42 ± 0.291	91.40 ± 1.24
F_{10}	26.23 ± 0.3299	0.23 ± 0.005	145.0 ± 3.9665	2.39 ± 0.158	84.50 ± 1.86
F_{11}	30.80 ± 0.5099	0.29 ± 0.005	152.6 ± 2.0548	2.23 ± 0.232	82.48 ± 3.33
F_{12}	28.60 ± 1.4445	0.28 ± 0.011	149.5 ± 2.3342	1.99 ± 0.323	87.36 ± 0.71
F_{13}	11.70 ± 0.7118	0.13 ± 0.011	247.0 ± 1.6329	3.03 ± 0.174	83.32 ± 2.29
F_{14}	11.30 ± 0.0471	0.12 ± 0.015	197.0 ± 0.8164	2.66 ± 0.039	87.32 ± 0.63
F_{15}	25.53 ± 1.0498	0.15 ± 0.005	221.0 ± 1.6329	2.52 ± 0.196	95.95 ± 1.09
F_{16}	22.13 ± 2.6599	0.14 ± 0.010	171.7 ± 0.9428	2.48 ± 0.399	85.68 ± 1.26
F_{17}	26.03 ± 0.8178	0.18 ± 0.005	129.3 ± 0.4714	2.34 ± 0.070	77.77 ± 1.24
F_{18}	24.90 ± 0.3741	0.17 ± 0.017	105.7 ± 1.2472	2.25 ± 0.037	86.52 ± 1.04

Table-4 CALIBRATION CURVE OF BENAZEPRIL HYDROCHLORIDE USING PHOSPHATE BUFFERED SALINE PH 7.4

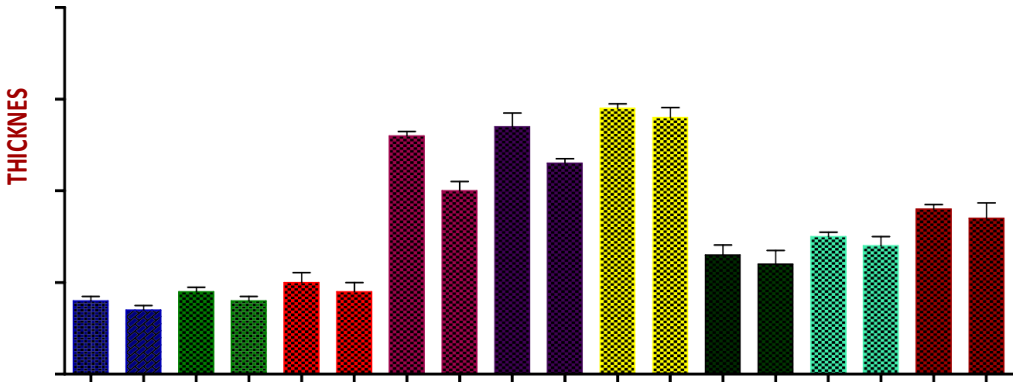
Sl.no	Concentration µg/ml	Absorbance ± S.D*
1	2	0.043 ± 0.0000
2	4	0.083 ± 0.0016
3	6	0.128 ± 0.0049
4	8	0.163 ±0.0016
5	10	0.198 ± 0.0008
6	12	0.247 ± 0.0058
7	14	0.285 ± 0.0049
8	16	0.333 ± 0.0060
9	18	0.371 ± 0.0071
10	20	0.414 ± 0.0104
		$\gamma = 0.9996$

***n = 3**



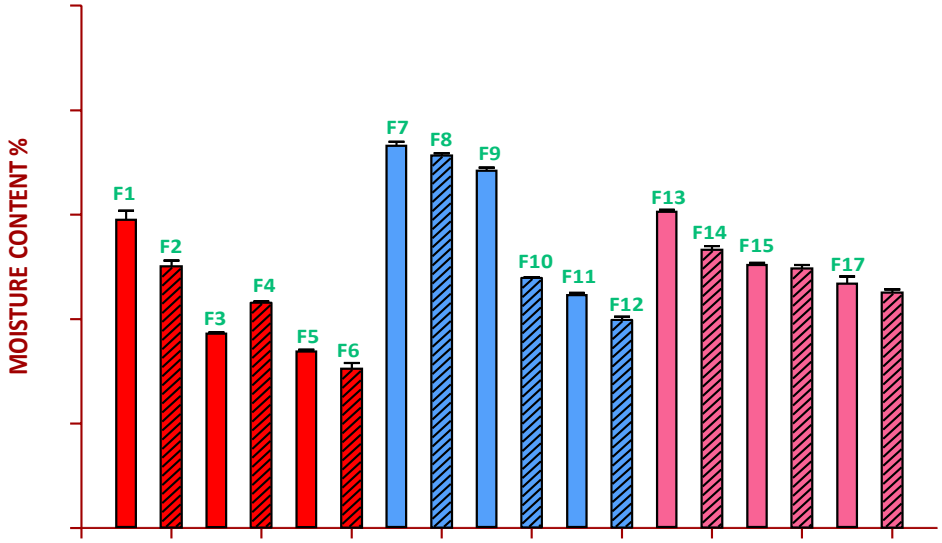
F1-EC 2.5% (WITH DM
F2-EC 2.5% (WITHOUT
F3-EC 3.0% (WITH DM
F4-EC 3.0% (WITHOUT
F5-EC 3.5% (WITH DM
F7-ES100 4.0% (WITH DMSO)
F8-ES100 4.0% (WITHOUT DMSO)
F9-ES100 4.5% (WITH DMSO)
F10-ES100 4.5% (WITHOUT DMSO)
F11-ES100 5.0% (WITH DMSO)
F13-EL100 4.5% (WITH DMSO)
F14-EL100 4.5% (WITHOUT DMSO)
F15-EL100 5.0% (WITH DMSO)
F16-EL100 5.0% (WITHOUT DMSO)
F17-EL100 5.5% (WITH DMSO)

FIGURE-1 WEIGHT VARIATION OF ALL THE FORMULATIONS



F1-EC 2.5% (WITH DMS
F2-EC 2.5% (WITHOUT
F3-EC 3.0% (WITH DMS
F4-EC 3.0% (WITHOUT
F5-EC 3.5% (WITH DMS
F7-ES100 4.0% (WITH DMSO)
F8-ES100 4.0% (WITHOUT DMSO)
F9-ES100 4.5% (WITH DMSO)
F10-ES100 4.5% (WITHOUT DMSO)
F11-ES100 5.0% (WITH DMSO)
F13-EL100 4.5% (WITH DMSO)
F14-EL100 4.5% (WITHOUT DMSO)
F15-EL100 5.0% (WITH DMSO)
F16-EL100 5.0% (WITHOUT DMSO)
F17-EL100 5.5% (WITH DMSO)

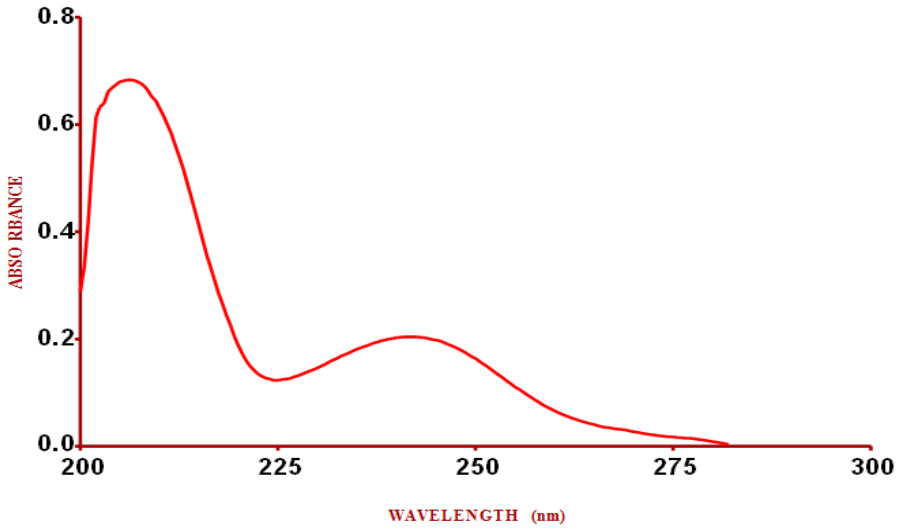
FIGURE-2 THICKNESS OF ALL THE FORMULATION



F1-EC 2.5% (WITH DMSO)
F2-EC 2.5% (WITHOUT DMSO)
F3-EC 3.0% (WITH DMSO)
F4-EC 3.0% (WITHOUT DMSO)
F5-EC 3.5% (WITH DMSO)
F6-EC 3.5% (WITHOUT DMSO)
F7-ES100 4.0% (WITH DMSO)
F8-ES100 4.0% (WITHOUT DMSO)
F9-ES100 4.5% (WITH DMSO)
F10-ES100 4.5% (WITHOUT DMSO)
F11-ES100 5.0% (WITH DMSO)
F12-ES100 5.0% (WITHOUT DMSO)
F13-EL100 4.5% (WITH DMSO)
F14-EL100 4.5% (WITHOUT DMSO)
F15-EL100 5.0% (WITH DMSO)
F16-EL100 5.0% (WITHOUT DMSO)
F17-EL100 5.5% (WITH DMSO)
F18-EL100 5.5% (WITHOUT DMSO)

FIGURE-3 MOISTURE CONTENT OF ALL FORMULATION

FIGURE-4 DETERMINATION OF λ_{max} OF BENAZEPRIL HYDROCHLORIDE IN PHOSPHATE BUFFERED SALINE OF pH 7.4



CONCLUSION:

The mercury substrate assay was victoriously used for the development of transdermal patches which provided opaque, smooth, flexible, non-sticky, and uniform patches. However, in ethyl cellulose polymer, patches were obtained as transparent, smooth, flexible, nonstick, and uniform. While in the case of Eudragit polymer, patches were observed as transparent, smooth, flexible, non-sticky, and uniform. The results were noted in Table-2. It was concluded that the method used for casting the film on a mercury substrate was found to be acceptable. The drug, benazepril hydrochloride was used to design the transdermal patches. However structure of the drug was successfully elucidated by FT-IR spectroscopic and Differential Scanning Calorimetry techniques.

The drug release study of transdermal patches was favorably achieved by using redesigned paddle over disc assembly (USP Apparatus 5) approach. Ultimately the objective was to estimate, characterize and rationalize the drug release from matrix film

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