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FABRICATION AND *IN-VITRO* CHARACTERISATION OF TRANSDERMAL PATCH USING JACKFRUIT MUCILAGE AS NATURAL POLYMER

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ABSTRACT

The present investigation highlights the formulation and characterization of transdermal patch of Acyclovir using *Artocarpus heterophyllus lam* mucilage as a natural polymer. The jackfruit mucilage was obtained from ripen fruit pulp of *Artocarpus heterophyllus lam* belonging to family Moraceae. The mucilage was isolated by maceration technique. The mucilage was characterised for purity, pH of mucilage, swelling index, ash value, Angle of repose, true density, bulk density, tapped density, Hausner's ratio and Carr's consolidation index. The films were prepared by solvent casting method and optimized by using 3² full factorial design and evaluated for physical parameters such as flatness, folding endurance, thickness, tensile strength, moisture content, moisture uptake, skin irritation potential of polymer matrices. The factorial design gave the 9 batches, depending on the desirability the solutions generated by the expert design the F1 batch was selected and there results of thickness (0.57 ± 0.0057 mm), folding endurance (311 times), %drug content (98.06 ± 0.02%), In-vitro drug release (95.26 ± 0.046%), Moisture content (7.53 ± 0.01%), Surface pH (6.38±0.058), Swelling index (7.13 ± 0.01), Tensile strength 24.88 ± 0.16 (N/25.4 mm). The transdermal patch using jackfruit mucilage shows the 100 % flatness. Thus from the present work it can be concluded that the Jackfruit mucilage can be a promising polymer for the film formation and thus should be explored in future as an inert suitable pharmaceutical excipient.

Keywords: Transdermal Patch, Film former, Jackfruit mucilage, folding endurance, Factorial design.

INTRODUCTION

Advanced techniques in biomaterials have resulted in the formulation of novel dosage form more pertinent to the transdermal, meeting the challenges of the physiochemical properties of the drug entity itself and achieving the therapeutic aim of the drug delivery system. The transdermal route has been used for many years to deliver drugs, which undergo first-pass metabolism. The transdermal route has the advantage of allowing excellent accessibility and reasonable patient compliance. Skin region offers attractive route of administration for local or systemic drug delivery. Recently interests have been focused on the delivery of drug to transdermal membrane by the use of bioadhesive material from natural sources. Several transdermal formulations are available and drug delivery via transdermal membrane is gaining importance as a novel route of drug administration.

A transdermal patch or skin patch is a medicated adhesive patch that is placed on the skin to deliver a time released dose of medication through the skin and into the bloodstream. Transdermal drug delivery is the application of drug on the skin surface so that it can permeate through the skin and reaches the systemic circulation at sufficient concentration to ensure therapeutic efficacy.

Now a day's natural mucilage are gaining importance as promising biodegradable polymeric materials. Many studies have been carried out in

the fields including food technology and pharmaceuticals using mucilage as polymers. It is clear that mucilage have many advantages over synthetic materials. Various applications of mucilage have been established in the field of pharmaceutical formulation of novel drug delivery systems, biotechnological applications and other delivery systems. Therefore, in the years to come, there will be continued interest in natural mucilage and their modifications aimed at the development of better materials for drug delivery systems.

The Artocarpus heterophyllus lam (jackfruit) mucilage is natural mucilage obtained from the ripe fruit pulp of the Artocarpus heterophyllus lam belonging to family Moraceae. The mucilage consists of carbohydrates. The literature survey revealed that it has potential as a film former and also as the tablet binder. Thus it can be explored for the formulation of transdermal patch using Acyclovir as a model drug for extended delivery of drug. The present investigation highlights the formulation and characterisation of transdermal Acyclovir patch of using Artocarpus heterophyllus lam mucilage as a natural polymer and it optimized by factorial design.

MATERIAL AND METHODS Materials

Acyclovir was obtained as a gift sample from Zim laboratories, Kalmeshwer, Nagpur. The fruits of Artocarpus heterophyllus (Jackfruit) were collected in the month of July-August, 2014 from the Hingna Market, Nagpur near the area of Priyadarshini J L College of Pharmacy Nagpur. The Artocarpus heterophyllus lam plant was authenticated by Senior Botanist Dr. Dongarwar Rashtrasant Tukadoji Maharaj Nagpur sir. University, Botany department Nagpur. Glycerin, Methyl paraben, procured by S.D. fine chemical's Mumbai. All reagents were AR grade. The drug samples were characterized by means of UV spectrophotometric method along with determination of solubility and pH analysis for their authentication.

Methods Extraction of mucilage

Artocarpus heterophyllus (Jackfruit) lam mucilage is extracted by maceration process. The ripe fruit pulp of Artocarpus heterophyllus lam was separated and the seeds were removed and the pulp was soaked in distilled water with occasional shaking. The soaked pulp was ground in grinder and kept for 24 hr with occasional stirring. After 24 hr material was squeezed through a muslin cloth to separate the marc from filtrate. The filtrate was precipitated with the suitable organic solvent. The precipitated fibres were separated by decanting the organic solvent and then the mucilage was dried. The extracted mucilage was stored in desicator till further use at room temperature.

Preparation of transdermal patch

For the present work the solvent casting method was selected for the preparation of transdermal films as this method is easy and can be done at laboratory level because it does not required bulky apparatus. In solvent casting method water soluble polymer was dissolved in a separate beaker to form a homogenous viscous solution. Other ingredients, including Acyclovir were dissolved in small proportion of aqueous solvent using magnetic stirrer. These viscous solutions were mixed thoroughly and then were degassed under vacuum. The resulting bubble free solutions were poured onto a glass mould and were kept in oven. Dried films were cut into the desired shapes and sizes for the intended application.

Drug excipients compatibility studies

Pure drug (Acyclovir), *Artocarpus heterophyllus lam* (Jackfruit) mucilage and their physical mixture were examined by using Fourier Transform Infrared (FTIR) using potassium bromide pellet method and Differential scanning calorimetry (DSC).

Experimental design

The application of factorial design gives a statistically systematic approach for the formulation and optimization of transdermal patch with desired thickness, folding endurance and % drug release. The formulations were fabricated according to a 3² full factorial design, allowing the simultaneous evaluation of two

formulation (independent) variables such as amount of jackfruit (X1) and amount of plasticizer (X2) and their interaction. The experimental designs with corresponding formulations are outlined in table 4. The three dependent variables that were selected for study were thickness (Y1), folding endurance (Y2) and % drug release (Y3). STAT-EASE, design expert 8.0.0 software was used for generation and evaluation of the statistical experimental design. The design including investigated factors and responses are show in table 2 and 3. For optimization purpose effect of various independent variables upon measured responses was studied using following mathematical equation i.e. multiple liner regression analysis (MLRA) involving independent variables and their interaction for various measured responses generated by 3² full factorial design.

 $Y = \beta 0 + \beta_1 X_1 + \beta_2 X_2 + \beta_3 X_1 X_2 + \beta_4 X_1^2 + \beta_5 X_2^2 + \beta_6 X_1 X_2^2 + \beta_7 X_1^2 X_2 + \beta_8 X_1^2 X_2^2$

Where, Y is the dependent variable, while β_0 is the intercept is the arithmetic average of all quantities outcomes of 9 runs, $\beta 1$ to β_8 are the coefficients computed from the observed experimental values of the independent variables. The terms X_1X_2 and X_i^2 (i=1, 2) are the interaction and polynomials terms, respectively. The statistical validation of the polynomials was established on the basis of vates's ANOVA provision in the Design Expert software and it is shown in table 6. Subsequently, feasibility as well as grid search was performed to locate the composition of optimum formulations. Also three-dimensional (3-D) responses surface graphs were generated by the Design Expert software and it is shown in figure 7-12 and the results of responses is shown in table 5. Table 3 summarize accounts of the 9 experimental run studied, their factor combinations, and the translation of coded level to the experimental unit employed during study.

Characterisation of Transdermal Patch Weight uniformity

For weight uniformity determination, three films of the size 10 mm diameter were weighed individually using digital balance (Wensar, PGB200 MUMBAI) and the average weight was calculated with \pm S.D. Result as shown in table 9.

Thickness

Thickness of the films was measured using digital vernier calliper (Mitutoyo, japan). The thickness was measured at three different sites of the films and average was taken with \pm S.D. Result as shown in table 9.

Swelling index

The agar solution was prepared by dissolving 0.2 gm of agar in 10 ml of warmed phosphate buffer pH 7.4, and then this solution was poured into a petri dish and allowed to cool. After determining initial patch diameter, each patch was allowed to swell on it's respective surface of gel. The diameter of patches was determined after 2, 5 and 7 hrs. The results were recorded as the mean value of three reading. The swelling studies were performed for 7 hrs. Because a residence time of 7 hrs were recommended. Result as shown in table 9.

Surface pH

To determine surface pH, three patches of each formulation were allowed to swell for 2 hrs on the surface of agar plate. Surface pH was measured by using pH paper placed on the surface of swollen patch. Result as shown in table 9.

Percent moisture absorbance

The percentage moisture absorption test was carried out to check the physical stability of the transdermal patch at high humid conditions. Three 1cm diameter patches were cut out and weighed accurately. Then the patches were placed in desiccator containing saturated solution of aluminium chloride, keeping the humidity inside the desiccator at 75 %. After 3 days the films were removed, weighed and percentage moisture absorption was calculated. Average percentage moisture absorption of three films was calculated by following formula:

% Moisture absorbance = Final weight - Initial weight x 100

Final weight

Percentage moisture loss

Percentage moisture loss was measured to check the integrity of films at dry condition. Three 1cm diameter patches were cut out and weighed accurately and kept in desiccators containing fused anhydrous calcium chloride. After 3 days the films were removed, weighed. Average percentage moisture loss of three films was calculated by following formula:

% Moisture loss =

Final weight - Initial weight x 100 Final weight

Folding endurance

The flexibility of films can be measured quantitatively in terms of folding endurance. Folding endurance of the films was determined by repeatedly folding (1cm*1cm) films at the same place till it was broken. The number of times films were folded at the same place without breaking gave the value of folding endurance. Result as shown in table 9.

Flatness test

Three longitudinal strips were cut from each film at different portion like one from the centre, other one from the left side, and another one from the right side. The length of each strip was measured and the variation in length because of nonuniformity in flatness was measured by determining percent constriction, with 0% constriction equivalent to 100% flatness. Result as shown in table 9.

Drug content uniformity

The films were tested for drug content uniformity by UV-spectrophotometric method. Films of size (4.53 cm) diameter were cut from three different places from the casted films. Each film was placed in 100 ml volumetric flask and dissolved in phosphate buffer (pH 7.4) and 1 ml was taken and diluted with phosphate buffer (pH 7.4) up to 10 ml. The absorbance of the solution was measured at 252 nm using UV spectrophotometer (shimadzu 1601). The percentage drug content was determined using the standard graph and the same procedure was repeated for three films. Result as shown in table 9.

Tensile strength and % elongation

Tensile strength is the maximum stress applied to a point at which the film breaks. Elongation is defined as a measure of the capacity of a patch to deform prior to failure. Tensile strength and of the percent elongation patches were determined on tensile strength testing apparatus. Rectangular patch strips of 25.4 mm X 50mm were fixed between the jaws of the instrument. The load on the strip was gradually increased to a maximum at a speed of 50mm/min. and the change in the length of the strips that occur with increasing stress was measured i.e. it is calculated by the applied load at rupture divided by the cross-sectional area of strip. Tensile strength and percent elongation of three patches of each batch were measured. Result as shown in table 9.

In-vitro drug release

USP dissolution model is used for the in- vitro drug release study. The paddle over disc method (USP apparatus V) can be employed for assessment of the release of the drug from the prepared patches. Dry films of known thickness is to be cut into definite shape, weighed, and fixed over a glass plate with an adhesive. The glass plate was then placed in a 250-mL of the dissolution medium or phosphate buffer (pH 7.4), and the apparatus was equilibrated to $32\pm 0.5^{\circ}$ C. The paddle was then set at a distance of 2.5 cm from the glass plate and operated at a speed of 50 rpm. Samples (5mL aliquots) were withdrawn at appropriate time intervals up to 24 hr and analyzed by UV spectrophotometer. The experiments were performed in triplicate and the mean value was calculated. Graph is shown in figure 13 & 14. Result as shown in table 9.

Stability studies

To assess the stability of formulation stability studies were done as per ICH guidelines. The formulated transdermal patches were wrapped in aluminium foil and stored at $40\pm0.5^{\circ}$ c and $75\pm5\%$ RH for period of one month. After an interval of 15 days the patches were tested for physical appearance, weight variation, thickness and drug content uniformity. Result as shown in table 10.

RESULTS AND DISCUSSION Drug-Excipients Interaction Study

The drug-excipients interaction was performed by

FTIR and DSC and it is shown in figure 1-6.





Figure 1: FTIR of Acyclovir



Figure 2: FTIR of Artocarpus heterophyllus lam mucilage



Figure 3: FTIR of combination of Acyclovir and Artocarpus heterophyllus lam mucilage



Vidyadevi Bhoyar et al. / Pharmacophore 2015, Vol. 6 (6), 267-280





Figure 5: DSC of Artocarpus heterophyllus lam mucilage



Figure 6: DSC of combination of Acyclovir and Artocarpus heterophyllus lam mucilage

These results indicated partial amorphisation and solublization of Acyclovir due to the processing and absence of any additional peak indicated that there was no interaction between the drug and jackfruit mucilage in the formulation.

Preformulation study of acyclovir Solubility

Sr. No	Solvents	Results
1	Water	Slightly soluble
2	Dimethyl sulphoxide	Completely soluble
3	Organic solvents(Ethanol)	Insoluble

Table 1: Solubility of drug

Acyclovir is slightly soluble in water.

Melting point

The melting point of the Acyclovir was $250-257^{\circ}$ c.

Partition coefficient

Partition coefficient was determined as ratio of concentration of drug in n-octanol to the concentration of drug in water and the value is reported as 0.025 as log P and the partition coefficient observed 0.029.

Preparation and optimization of transdermal patch using jackfruit mucilage with acyclovir as a drug The transdermal films can be prepared by using solvent casting method. The procedure is mentioned in section 6.6 of experimental work. For the fabrication of transdermal patch the raw batches were prepared, in which the different concentration of jackfruit mucilage starts from the 0.5% w/v to 10% w/v were used. Also various types of plasticizers were used such as polyethylene glycol, Tween 80, Dibutyl-phthate, Glycerin but among these plasticizers the Glycerin gives the best results. The raw batch which gives the best result with 10% w/v of jackfruit mucilage and 8% v/v of glycerine. Acyclovir which is a BCS class III drug having high solubility low permeability was selected as the drug candidate for the transdermal patch. Acyclovir has low water solubility but it is soluble in DMSO completely. Also DMSO increases the permeability of drug through skin. So the purpose of drug solubility as well as the permeability of acyclovir is solved by use of DMSO.

Optimization

Table 2: Independent and Dependent variables

Factors	Low level (-1)	Middle level (0)	High level (1)
X ₁ - Amount of jackfruit (%w/v)	8	10	12
X ₂ - Amount of plasticizer (%v/v)	5	7	9

Table 3: Factors and Levels with their Real and Coded Values

Independent variables	Dependent variables
X ₁ - Amount of jackfruit mucilage (%w/v)	Y ₁ - Thickness (nm)
X ₂ - Amount of plasticizer (%v/v)	Y ₂ - folding endurance (no.)
	Y_3 - % drug release (%)

Batch code	Amount of jackfruit	Amount of	Drug	Dimethyl	Methyl	Distilled
	mucilage (X1)	plasticizer (X2)	(mg)	sulphoxide	paraben	water
	(%w/v)	(%v/v)		(ml)	(gm)	(ml)
F1	8	5	20	3	0.025	q.s 10
F2	8	7	20	3	0.025	q.s 10
F3	10	7	20	3	0.025	q.s 10
F4	12	5	20	3	0.025	q.s 10
F5	12	7	20	3	0.025	q.s 10
F6	10	9	20	3	0.025	q.s 10
F7	8	9	20	3	0.025	q.s 10
F8	10	5	20	3	0.025	q.s 10
F9	12	9	20	3	0.025	q.s 10

 Table 4: Formulation table

Table 5: Results of responses

Responses	F1	F2	F3	F4	F5	F6	F7	F8	F9
Thickness (mm)	0.57	0.68	0.74	0.78	0.7	0.75	0.8	0.72	1.00
Folding endurance (no.)	315	29	145	10	95	65	23	150	256
Drug release (%)	95.26	51	79	25	74	72	34	83	89

Polynomial equation generated by expert design of final optimized batch

(Note: Value of P-value i.e. prob > F less than 0.0500 indicate model terms are significant.) The model equation relating thickness as response became:

Thickness: (significant value= 0.0433)

THICKNESS =
$$+0.75+0.072X_1+0.080X_2$$
 ...

.....eq (1)

The model equation relating folding endurance as response became: **Folding Endurance:** (significant value= 0.0253)

FOLDING ENDURANCE= +120.89-1.00X1-0.2183X2+134.50X1X2

.....eq (2)

The model equation relating % drug release as response became: **% Drug Release:** (significant value=0.0296)

% DRUG RELEASE= +66.92+1.29X1-1.38X2+31.32X1X2eq (3)

In equations (1) positive sign for coefficient of X2 indicates that the thickness of transdermal patch increases when concentration of plasticizer (glycerin) is increased and positive sign for coefficient of X 1 indicate positive effect of jackfruit concentration on thickness. In equation (2) negative sign for coefficient of X 1 indicates that the folding endurance increases when concentration of jackfruit mucilage decreases and negative sign for coefficient of X2 indicates that folding endurance of transdermal patch increases when concentration of plasticizer (glycerin) decreases. In equations (3) positive sign for coefficient of X1 indicates that the % drug release of transdermal patch increases when concentration of jackfruit mucilage is increased and negative sign for coefficient of X1 indicate that the % drug release sign for coefficient of X1 indicate that the % drug release when concentration of plasticizer (glycerin) decrease.



Figure 7: Contour plot of thickness



Figure 8: 3D Graph of thickness





Figure 9: Contour plot of folding endurance Figure: 10 3D graph folding endurance



3. % Drug release





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Source		Sum of Squares	Degree of freedom (d.f.)	Mean Square	F-value	P-value i.e. probe>F
Thickness	Model	0.069	2	0.035	5.54	0.0433(S)
	X ₁ - Amount of jackfruit mucilage (%w/v)	0.031	1	0.031	4.93	0.0689(NS)
	X ₂ - Amount of plastcizer	0.038	1	0.038	6.15	0.0478(S)
Folding	Model	75227.00	3	25075.72	7.71	0.0235(S)
endurance	X ₁ - Amount of jackfruit mucilage (%w/v)	6	1	6	1.846E-003	0.9674(NS)
	X ₂ - Amount of plastcizer	2860.17	1	2860.17	0.88	0.3913(NS)
	X ₁ X ₂	72361.00	1	72361.00	22.26	0.0053(NS)
% Drug	Model	3943.87	3	1314.62	7.13	0.0296(S)
release	X ₁ - Amount of jackfruit mucilage (%w/v)	9.98	1	9.98	0.054	0.8252(NS)
	X ₂ - Amount of plastcizer	11.37	1	11.37	0.062	0.8137(NS)
	$X_1 X_2$	3922.52	1	3922.52	21.28	0.0058(NS)

Vidyadevi Bhoyar *et al. / Pharmacophore* 2015, Vol. 6 (6), 267-280 Table 6: Summery of ANOVA for the Response Parameters

X1, X2 represents Amount of jackfruit mucilage, Amount of plasticizer, respectively X1X2 is the interaction effects.

S and NS are the significant and non significant, respectively.

d.f. indicate degree of freedom.

Table 7. Solutions generated by the expert design								
Sr. No	Jackfruit Mucilage	Glycerin	Thickness	Folding	% Drug	Desirability		
	(% w/v) X1	(% v/v) X2	(mm)	Endurance (no.)	Release (%)			
1	8	5	0.59722	278.22	98.314	0.962		
2	8.05	5.21	0.599002	274.857	97.5738	0.956		

Table 7: Solutions generated by the expert design

The software gave two solutions. From that first solution was selected because it has highest desirability. The solutions generated by the software are depicted in table 7. The results of the optimized batch with its predicted value, actual values are given in table 8.

In-vitro drug release (%)



Figure 13: % *In-vitro* drug release (F1-F4)



(hrs) Figure 14: % *In- vitro* drug release (F5-F9)

Table 8:	Results	of op	otimized	batch	F1
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Batch	Composition		Responses	Predicted Value	Actual Values
	X 1	X2			
Optimized	8	5	Thickness (mm)	0.59722	0.57
Batch F1			Folding Endurance (No.)	278.22	311
			% Drug Release (%)	98.314	95.26

Table 9: Characterization parameter of batch F1

Sr. No.	Characterization Parameter	Result
1	Thickness (mm)	0.57 ± 0.0057
2	Folding endurance	311
3	Weight uniformity (gm)	2.3066 ± 0.0015
4	% drug content (%)	98.05±0.02
5	Surface pH	6.38±0.058
6	Swelling index	7.13±0.01
7	Flatness (%)	100
8	Tensile strength (N/25.4mm)	24.88 ± 0.16
9	% Elongation (%)	274.60 ± 12.62
10	In-vitro drug release (%)	95.26±0.046
11	Skin irritation test	No irritation or redness on the skin
11	Moisture content (%)	7.53 ± 0.01

Data represented ± S.D (n=3)

Parameters	0 days	15 days	30 days				
Weight uniformity (gm)	2.3066 ± 0.0015	2.3062±0.0012	2.3059±0.0021				
Thickness (mm)	0.57 ± 0.0057	0.57±0.0054	0.568 ± 0.001				
Folding endurance	311	311	309				
In-vitro drug release (%)	95.26±0.046	95.09±0.026	95.03±0.025				
Surface pH	6.38±0.058	6.35±0.023	6.34±0.065				
% drug content (%)	98.05±0.02	98.01±0.05	97.26±0.01				

Vidyadevi Bhoyar *et al. / Pharmacophore* 2015, Vol. 6 (6), 267-280 Table 10: Stability studies

CONCLUSION

From the present work it can be concluded that the Jackfruit mucilage can be a promising polymer for the film formation and thus should be explored in future as an inert suitable pharmaceutical excipient.

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