EVALUATION AND *IN-VIVO* SKIN IRRITATION STUDY OF TRANSDERMAL PATCHES CONTAINING FLUFENAMIC ACID

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ABSTRACT

A transdermal patch is medicated adhesive patch that is placed on the skin to deliver a specific dose of medication through the skin and into the blood stream. In this system the drug therapy can be stopped instantly in situation where drug input is no longer desirable. The system allows reduce frequency of dosing which is particular favorable Conventional systems of medication that require multi dose therapy are having many problems. The controlled drug delivery is a newer approach is to deliver drug in to systemic circulation at a predetermined rate. Our system should duplicate continuous intravenous infusion, which not only by passes hepatic 'first-pass' elimination but also maintains a constant, prolonged and therapeutically effective drug level in the body.

KEYWORDS: Transdermal, Drug Delivery, hepatic First pass, therapeutically, intravenous. Infusion

1.INTRODUCTION:

The transdermal route has become one of the most successful and innovative drug delivery system for research in pharmaceutical sciences. Transdermal drug delivery provides a leading edge over injectables and oral routes by increasing patient compliance and avoiding first pass metabolism respectively.[1,2]A transdermal patch is medicated adhesive patch that is placed on the skin to deliver a specific dose of medication through the skin and into the blood stream. In this system the drug therapy can best opped instantly in situation where drug input is no longer desirable.[3,4] The system allows reduce frequency of dosing which is particular favorable Conventional systems of medication that require multi dose therapy are having many problems.[5,6] The controlled drug deliveryis a newer approach is to deliver drug in to systemic circulation at a predetermined rate.Our system should duplicate continuous intravenous infusion, which not only by passes hepatic 'first pass' elimination but also maintains a constant, prolonged and therapeutically effective drug level in the body.[7,8,9]

2. MATERIALS AND METHODS:

2.1 SEM STUDY OF FLUFENAMIC ACID PATCHES

The surface morphology of the transdermal patches (F5) was studied before and after permeation studies using scanning electron microscopy (Zeiss Evo 50). Samples were mounted on an aluminum stub using a double -sided adhesive tape and making it electrically conductive by coating with a thin layer of gold palladium alloy in vacuum. The scanning electron microscope was operated at an acceleration voltage of 15 kV.

2.2 In-VitroRelease kinetics

The release studies from formulated patches were carried out by using Franz diffusion cell. Prepared patch was placed between the donor compartments of diffusion cell separated by dialysis membrane. The receptor region comprising of buffer containing magnetic bead, which is operated by magnetic stirrer, for stirring. Periodically l ml

of aliquot sample was taken out from the receptor compartment at graded time intervals and same is replaced with phosphate buffer pH 7.4, analysis was done using UV/Visible spectrophotometer at 288 nm against buffer as a reference. The drug release data of all formulations were fitted to various mathematical models such as zero order as cumulative % of drug released vs. time, first order as log cumulative % of drug remaining vs. time.

2.3 STABILITY STUDIES OF FLUFENAMIC ACID PATCHES:

Stability studies of formulations was conducted according to ICH guidelines by storing at 40 °C and 75 % RH for 3 months. The samples were withdrawn at 30, 60 and 90days and evaluated for physical appearance and drug contents. The ex vivo permeation study was performed after 90 days and compared with fresh batch.

2.4 EX-VIVO PERMEATION STUDIES OF FLUFENAMIC ACID PATCHES:

2.4.1 Preparation of Rat Skin:

The rats were sacrificed by excess ether inhalation and the abdomen carefully shaved with a razor after removal of hair by electric clippers. A full thickness skin was excised from the shaved abdominal site which is followed by removal of fat adhering to dermis with a scalpel. Any trace of fat adhering to skin was then finally removed by wiping it with cotton swabs soaked in isopropyl alcohol. Finally, skin was rinsed with phosphate buffer solution pH 7.4 and stored at -20°C in aluminium foil till used and always used within a week.

2.4.2 Cumulative amounts of drug permeated

The amount of drug transferred was estimated by taking 5ml of the sample at graded time intervals up to 24 hrs, the volume was reloaded with an equal withdrawn volume of buffer. The absorbance was measured at 288 nm spectrophotometrically. The graph was plotted between the Cumulative amounts of drug transferred in µg/cm^2 against time.

2.4.3 Drug Flux

The drug flux (µg /hr/ cm^2) at steady state was calculated by dividing the slope of the linear portion of the curve by the area of the exposed skin surface. The target flux can be calculated by the following equation

JTarget=CssClTBW/A

Were,

A = effective surface area of the transdermal patch

BW= average human body weight of 70 kg
Css=the steady state plasma concentration of Flufenamic acid
ClT= documented total clearance of Flufenamic acid

2.4.3.1 Lag time (T_{lag})

The lag time (T_{lag}) was determined by extrapolating the linear portion of the cumulative amount permeated versus time curve to the abscissa.

2.4.3.2 Enhancement Factor

The effectiveness of various permeation enhancers was determined by comparing drug flux in the presence and absence of each permeation enhancer, and obtained ratio was known as the enhancement factor (EF).

2.5 In-Vivo Study

To determine the irritant effect or any chance of edema with the use of transdermal patches, primary skin irritancy test was evaluated according to Draize test. Transdermal patches were applied onto the dorsal skin of albino rats (150-250 g), which was shaved on the previous day of the study. The rats were divided into five groups (six animals in each group). The animals of Group-I were served as control without any treatment. The medicated tape (USP adhesive tape) was applied to Group-II animals. The blank patches and Flufenamic acid patches (F5) were applied on to the animals of Group III and IV respectively. To Group-V (standard), 0.8 % v/v aqueous solution of formalin as standard irritant was applied. The animals were applied with new patch/ formalin solution each day upto 7 days and skin irritation (erythema and edema) was evaluated by visual scoring The scores were given for erythema from 0 to 4 depending on the degree of erythema as follows: no erythema 0, slight erythema (barely perceptible- light pink) 1, moderate erythema (dark pink) 2, moderate to severe erythema (light red) 3, severe erythema (extreme

redness)4. The edema scale was:0,none;1,slight;2,well defined;3, moderate; and 4, severe. After visual evaluation of skin irritation, the animals were sacrificed.

3. RESULTS AND DISCUSSION

3.1 SEM Study

The surface morphology of the transdermal patches before and after *in vitro* permeation study was scanned using a scanning electron microscope (SEM).

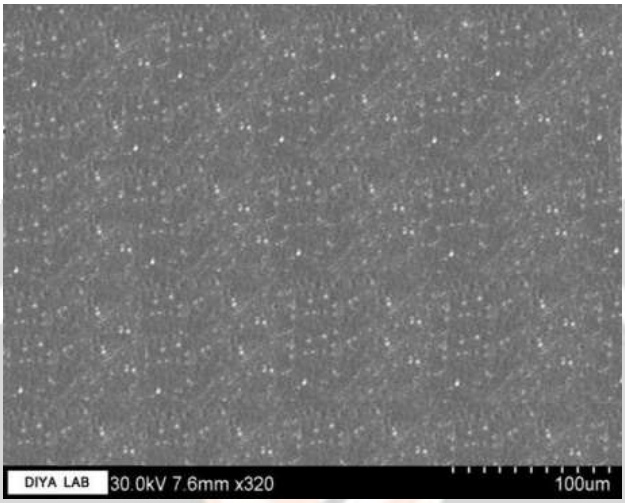

Figure 1: SEM photographs of Flufenamic acid patch (F5) before permeation study

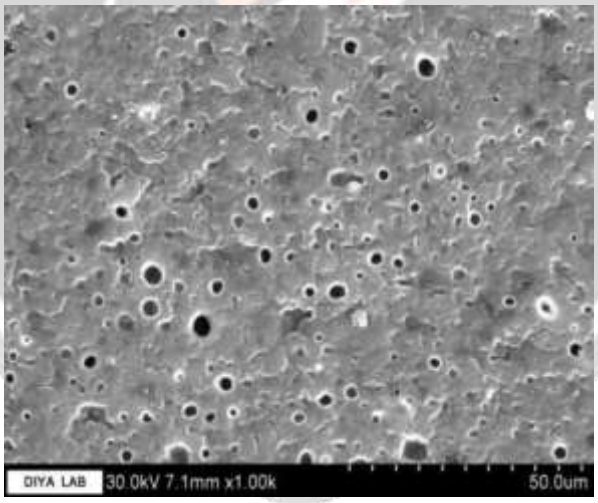

Figure 2: SEM photographs of Flufenamic acid patch (F5) after permeation study.

3.2 In-Vitro Release study

The percentage of drug release order was as follows
:F5>F4>F3>F2>F1
Formulations F5 showed the greatest percentage of drug release values. It was revealed that the ratio between drug: PVP K90 at 1:2 was found to be the most effective in increasing the drug release from patch.

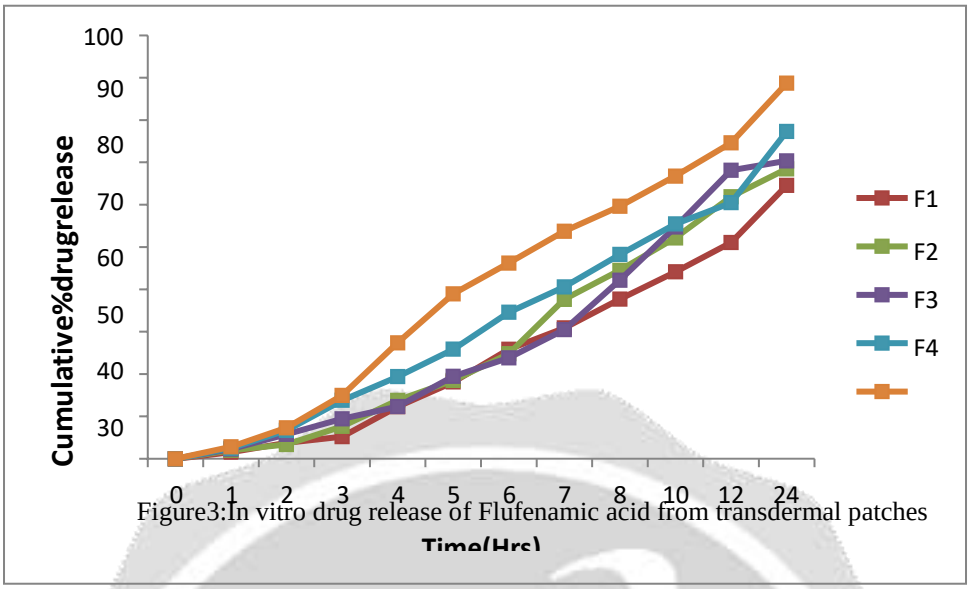

Figure3:In vitro drug release of Flufenamic acid from transdermal patches

RELEASE KINETICS OF FLUFENAMIC ACID PATCHES

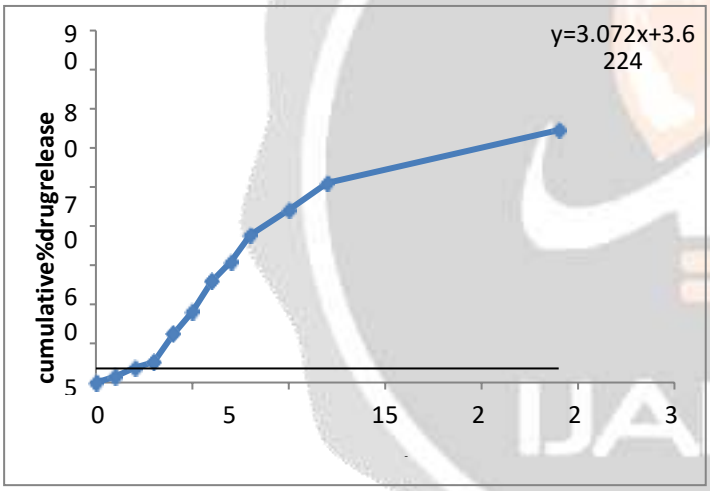

Figure4: Zero order release kinetics F1

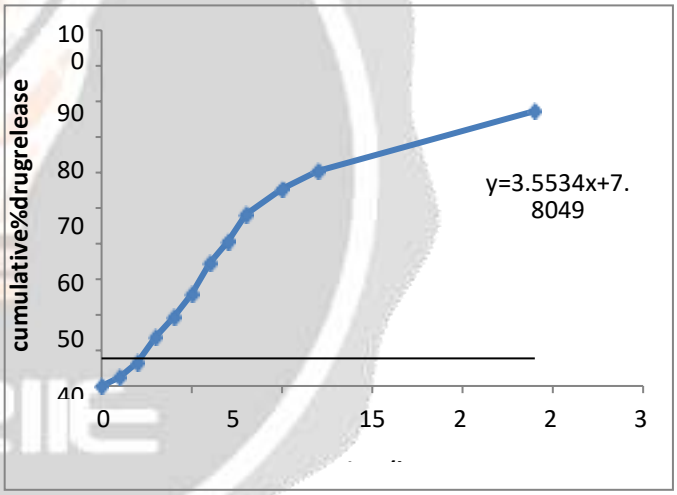

Figure5: Zero order release kineticsF4

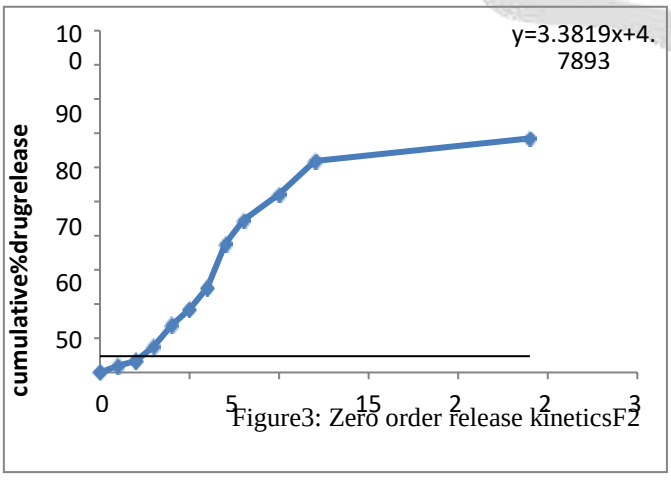

Figure3: Zero order release kineticsF2

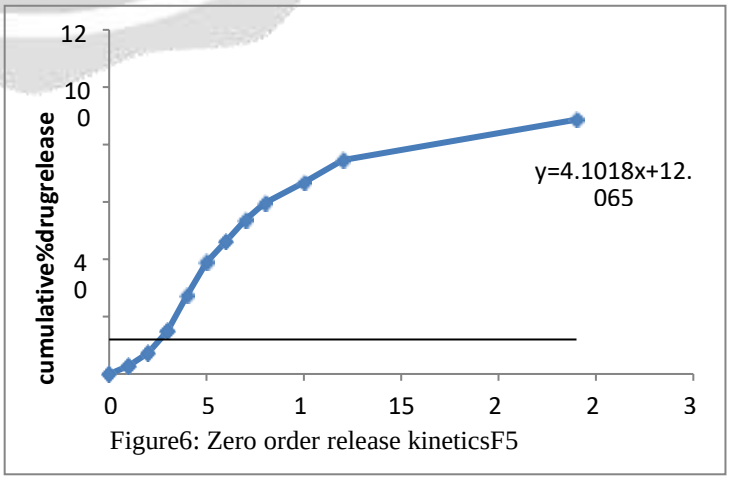

Figure6: Zero order release kineticsF5

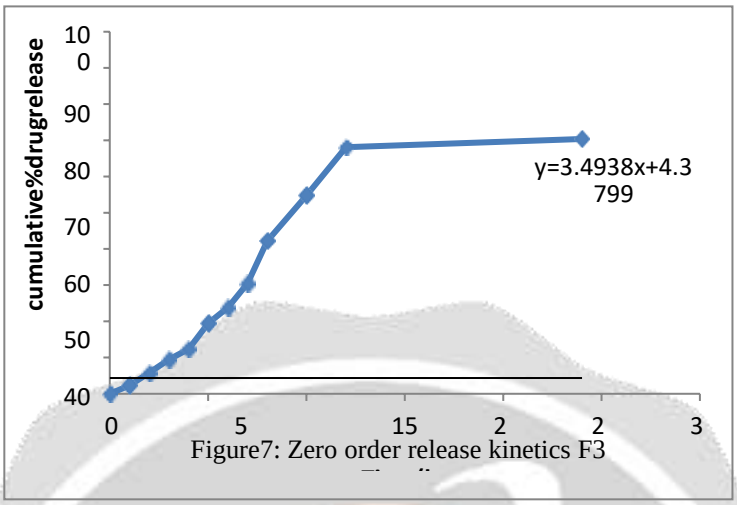

Figure7: Zero order release kinetics F3

3.3 STABILITY STUDIES OF FLUFENAMIC ACID PATCHES:

The stability study of optimized formulation (F5) was conducted according to ICH guidelines ;the formulation was stored at40°C and 75% relative humidity for 3 months. The result indicated that no change in physical appearance was observed after 90 days. The drug content of the patch was found 97.11, 96.91 and 96.84% after 30, 60 and 90 days respectively, indicated that no significant change after 3 months.

S.No.	Time interval	Physical appearance	Drug contents(%)
1.	0 days	Uniform	0
2.	30 days	Uniform	97.11
3.	60 days	Uniform	96.91
4.	90 days	Uniform	96.84

Table6: Data of the stability of Flufenamic acid formulation(F5)

3.4 Ex-Vivo Study

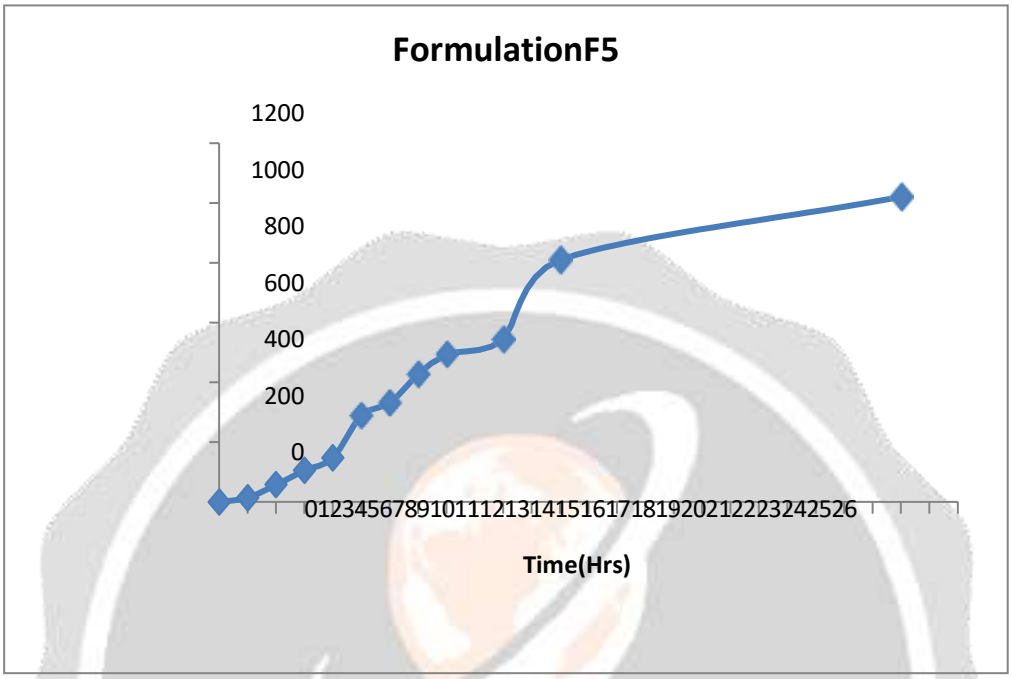

Figure 8. Ex vivo permeation studies of Flufenamic acid through rat skin from transdermal patches

3.4.1 Lagtime(T_{lag})

The lag time for Formulation 9 was found to be 0.72 ± 0.58 hours.

3.4.2 Enhancement Factor

The obtained value of Enhancement Factor was 4.36.

3.4.3 *Ex vivo* permeation studies of fresh batch and 3 month old batch

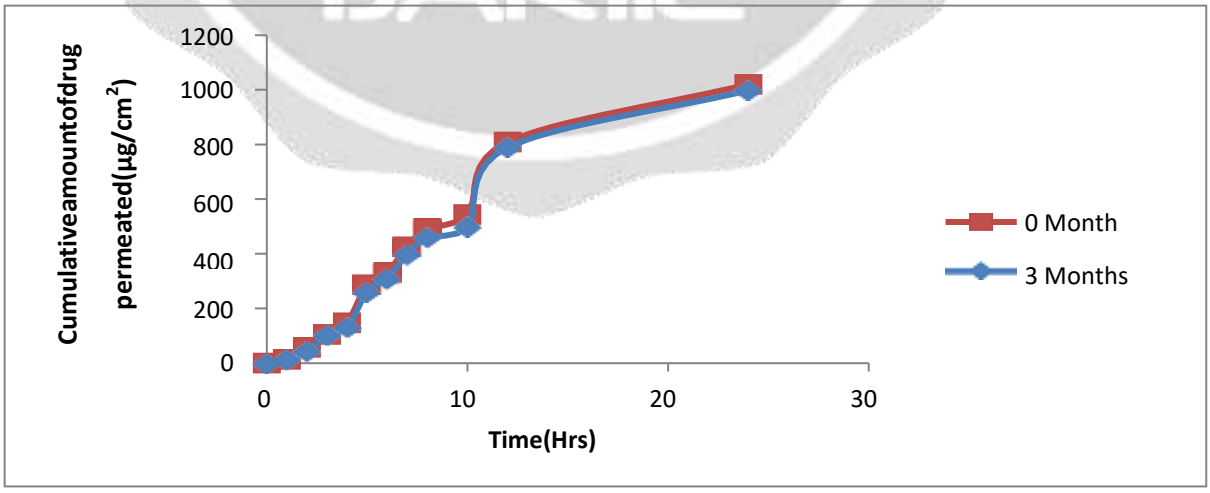

Figure9: *Ex vivo* drug permeation profile of fresh and 3 month old transdermal patch of optimized formulation (F5)

3.5 IN-VIVO IRRITATION STUDIES OF FLUFENAMIC ACID PATCHES

The skin irritation test of Flufenamic acid transdermal patches (F5) was performed on dorsal skin of albino rats in comparison with USP adhesive tape and standard irritant formalin (0.8%).

S. No.	Control		USPAdhesive Tape		BlankPatch		FormulationF5		Formalin	
	Erythema	Edema	Erythema	Edema	Erythema	Edema	Erythema	Edema	Erythema	Edema
1	0	0	2	1	1	0	1	0	3	3
2	0	0	1	1	2	1	2	1	3	4
3	0	0	2	1	1	1	1	1	3	4
4	0	0	1	2	1	0	1	1	4	3
5	0	0	0	1	1	2	0	2	3	4
6	0	0	1	0	2	0	2	0	3	3
Avg.	0.00	0.00	1.16	1.00	1.33	0.66	1.66	0.83	3.16	3.50

Table1:Results of skin irritation study of Flufenamic acid patches

The scores were given for erythema from 0 to 4 depending on the degree of erythema as follows: no erythema 0, slight erythema (barely perceptible- light pink) 1, moderate erythema (dark pink) 2, moderateto severe erythema (light red) 3, severe erythema (extreme redness) 4. The edema scale was:0,none; 1,slight; 2,well defined; 3,moderate; and 4, severe. The skin irritation score (erythema and edema) was found to be less than 2. According to Draize *et al.* compound which producing score of less than 2 are considered negative Hence, the prepared transdermal patches of Flufenamic acid were free of skin irritation.

4. SUMMARY AND CONCLUSION

The result indicated that the drug is uniformly distributed in prepared transdermal patch and after permeation study; it was observed that the drug is released from the patch on to the skin,which can be then permeated through skin in to the systemic circulation.

The patches were subjected to permeation study through rat abdominal skin using modified Franz diffusion cell. The diffusion kinetics from all the formulation obeyed zero order kinetics, diffusion as mechanism to drug release following non Fickian diffusion.

5. REFERENCES

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